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for acute poisoning by cyanides*

Volume I

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Efficacy and Safety of Antidotes for Acute Poisoning by Cyanides

CONTENTS

1. SUMMARY AND CONCLUSIONS

Cyanide poisoning is a very serious, albeit rare, event with possible neurological sequelae that may result in severe disability, and death. It can occur after exposure by ingestion, inhalation and dermal absorption of hydrocyanic acid, cyanide salts, or cyanogenic compounds such as acetone cyanohydrin, nitriles, biological substances like cassava or amygdalin (Vitamin B17), or sodium nitroprusside (SNP) (used as a medication for hypertension). Fire smoke inhalation may also cause cyanide poisoning under certain conditions of combustion.

Despite the 1993 overview on "Antidotes of Poisoning by Cyanide" (IPCS/CEC Evaluation of Antidotes Series) and the review of commonly used cyanide antidotes in the 2007 ECETOC Report in the JACC (Joint Assessment of Commodity Chemicals) series "Cyanides of Hydrogen, Sodium and Potassium, and Acetone Cyanohydrin" there has been no recognised consensus on the relative efficacy, efficiency, safety, and practicality of the various antidote treatments being used across the world. This situation although undesirable was sustained by the absence of any comprehensive review which was in turn due to the complexity of the clinical picture, since cyanide antidotes are employed in different circumstances of poisoning. In order to address this situation ECETOC established a new Task Force to review the available evidence on the efficacy, efficiency, safety, and practicality of antidote regimes under the different poisoning circumstances in which they were used. The four circumstances considered were direct poisonings with hydrocyanic acid or its salts, poisoning with cyanogenic compounds (above), fire smoke inhalation, and poisoning with initially unknown substances.

The Task Force reviewed the available literature until mid-2010 and collected further cases with a questionnaire from industry and Poison Control Centres. The Poisoning Severity Score (PSS), a well-established score system addressing all organ systems for grading of poisoning severity, was adjusted slightly to meet the specifics of cyanide poisoning (PSSa – 'a' denotes adapted).

The reported cases were then evaluated for antidote use. In the majority of cases single antidotes were given although in some combinations of antidotes were used. Cases were split into treatment groups based on the sequence in which different antidotes were administered, if more than one antidote was given. For each such sequence the efficacy based on the PSSa and the safety of the respective antidote or antidote combinations were described. This sequential view allowed in many cases for a statistical analysis of efficacy which was included in the single antidote assessments together with pharmaceutical data, pharmacokinetic data, safety/side effects, practicality, an overview of reported cases and case series. Single casuistic case reports shortly described in tables were collected in an appendix as summaries to make the antidote chapters more readable. An overall assessment was made for each antidote.

Pre-clinical antidotes were addressed based on data from animal experiments. In particular alpha-ketoglutarate and cobinamide, and to a lesser extent, dihydroxyacetone and pyruvate (pro-drugs) seem to have promising potential as cyanide antidotes.

It was apparent that although all direct acting antidotes (excluding sodium thiosulphate) may reverse cardiac arrest or at least facilitate resuscitation, they cannot prevent neurological sequelae due to hypoxia in all cases.

Sodium thiosulphate appears to act more rapidly than previously thought, but is effective, when administered alone, only in moderate poisoning (PSSa 2). Amyl nitrite, when administered alone, was found to be effective in moderate to severe poisonings (PSSa 2.5). Hydroxocobalamin, when administered alone, was found to be effective in severe poisonings (PSSa 3). The combinations of sodium nitrite and sodium thiosulphate, with or without amyl nitrite (PSSa 3), and of dimethylaminophenol and sodium thiosulphate (PSSa 2) were also found to be at least partially effective. These methaemoglobin forming agents all require combination with sodium thiosulphate to capture cyanide released during physiological methaemoglobin reduction. Dicobalt edetate has rarely been given, so that no comprehensive evaluation was possible. In any case, the intrinsic toxicity of dicobalt edetate significantly reduces its applicability. In regard of its effectiveness and low toxicity sodium thiosulphate can be given after all direct acting antidotes in situations when a delayed formation of cyanides cannot be excluded.

Oxygen, in contrast to the other antidotes discussed, has no effect on cyanide toxicokinetics, and is therefore not regarded as an antidote in its own right. Oxygen was only partially effective in severe poisonings (PSSa 3), and other antidotes were fully effective without oxygen. The administration of normobaric oxygen in combination with other antidotes to cyanide is very effective and possibly at least additive. It is however recommended that in conditions where oxygen is not available, treatment with antidotes to cyanide should not be delayed.

For the different circumstances of cyanide poisoning mechanism of cyanide formation, course, case series, case tables and an overview of antidote uses in the respective conditions were addressed, before recommending (a) specific antidote(s) for the condition. These recommendations could not only deal with efficacy and safety, but had also to account for practicility, e.g. cold storage requirement in a tropical country, or high price in developing countries. These combined aspects prompted recommendations for different situations / circumstances and severities shown in the following table.

Table 1: Antidotes[a] recommended for acute poisoning by cyanides

Circumstance [b]	Severity of poisoning	
	Mild or moderate	**Severe**
Direct		
HCN or its salts	None or STS (HOCO)	AN/SN or DMAP, followed by STS
Cyanogenic compound		
Cassava	None or STS	STS, in very rare most severe cases SN followed by STS
Laetrile/Amygdalin	None or STS	AN/SN/STS or HOCO(/STS) (or DMAP/STS)
Nitriles	None or STS	STS, in very severe cases SN/STS or HOCO(/STS) or DMAP/STS
SNP	STS	STS
Smoke		
CO and cyanide	None or STS	HOCO (followed by STS). Neither AN/SN nor DMAP
Unknown	None or STS	HOCO or AN, followed by STS. If then required SN/STS (or DMAP/STS)
Child	None or STS	HOCO and/or STS
Mass poisoning	None	AN (or DMAP i.m.)
First aider	None	AN

[a] STS, sodium thiosulphate; HOCO, hydroxocobalamin; AN, amyl nitrite; SN, sodium nitrite; DMAP, dimethylaminophenol; i.m., intramuscular (injection).
[b] HCN, hydrogen cyanide; SNP, sodium nitroprusside; CO, carbon monoxide.

The basis of cyanide poisoning treatment, independent of the source, are the usual measures of life support i.e. oxygen, and if required mechanical ventilation and safeguarding of adequate circulation by e.g. catecholamines.

2. INTRODUCTION

2.1 Background

Acute cyanide poisoning, while being a rare occurrence, is an extremely serious event due to the potentially fatal outcome, with a reported mortality rate ranging from 11 to 95% (Yen et al, 1995). While there is general agreement on the immediate supportive care in acute cyanide poisoning there is no such international agreement on the preferred first-line antidote treatment, with different countries favouring different antidote regimes. Currently applied antidote therapies include: Oxygen and sodium thiosulphate (STS) used throughout the world, amyl nitrite (AN) and sodium nitrite (SN) initially promoted in the USA, but in use worldwide, 4-dimethylaminophenol (DMAP) in Germany, dicobalt edetate (Co-EDTA) primarily in UK and France, and hydroxocobalamin (HOCO) in France, also spreading worldwide more recently. These seven antidotes have been shown to be of some use in the treatment of acute human cyanide poisonings, at least in specific situations.

In most instances cyanide antidotes are recommended and used as combinations, e.g. AN/SN/STS, DMAP/STS, or HOCO/STS. Since there have been no controlled studies on the efficacy of either single antidotes or combinations recommendations on their preferred use rely on expert opinion rather than on evidence-based medicine. Some authors have even recommended supportive treatment and oxygen only.

This situation does not reflect an absence of literature on cyanide antidotes. Quite the opposite, there are several publications by expert panels that have dealt with the issue of cyanide toxicity and poisoning, e.g. the IPCS' "Antidotes for Poisoning by Cyanide" (1993a) and the European Agency for the Evaluation of Medicinal Products "Guidance Document on the Use of Medicinal Products for the Treatment of Patients Exposed to Terrorist Attacks with Chemical Agents" (EMEA, 2003, 2006). There are also other monographs, guidelines and proceedings at a national level. There is a book on "Clinical and Experimental Toxicology of Cyanides", published in 1987, that addresses a number of issues regarding the mechanisms of toxicity, condition of cyanide poisonings, and antidotal treatment (Ballantyne and Marrs, 1987). There are also review articles that have been published, updating knowledge about old as well as new antidotes and even promising substances from preclinical studies.

The European and other producers of hydrogen cyanide and its salts having reviewed this situation concluded that the present divergent use of antidote regimes was due to the absence of a comprehensive evidence-based assessment of their efficacy and safety and that since this created confusion this was undesirable.

Since the earlier ECETOC review on the toxicology and ecotoxicology of Cyanides (ECETOC, 2007) contains only a short chapter describing the available cyanide antidotes and

following the IPCS publication on antidotes to poisoning by cyanides (IPCS, 1993a), some antidotes had been registered as licensed drugs and consequently a greater number of case reports had become available, ECETOC established a new Task Force to critically review the efficacy and safety of antidotes used for the treatment of acute cyanide poisoning taking into account the current state of knowledge.

As the success of the analysis would rely considerably on case study reports and many of these were recognised to be of limited detail, the task force decided to use the internationally accepted Poisoning Severity Score (PSS) scheme since it would allow inclusion of case studies for which only a few clinical data and the final outcome are known. The PSSalso allows addressing further issues, e.g. time-course of the poisoning, or grading the severity for each step in sequential antidotal treatment, or assessing the efficiency and safety of a treatment.

In establishing the scope of the review and the tructure of the report it was recognised that different cyanide compounds and circumstances of poisoning may require different antidotes. Namely hydrogen cyanide and its salts which are known to act rapidly while cyanogenic compounds liberate cyanide more slowly resulting in a delayed action. Furthermore cyanide poisonings might be combined with exposure to other toxic chemicals such as in the case of fire smoke, or the poisoning may be with an unknown substance (e.g. during terrorist attack) and the involvement of cyanide(s) may be purely circumspect.

The aim of the Task Force was therefore to review all retrievable data, published and in-company experiences, regarding the efficacy and safety of the seven listed cyanide antidotes, with the intention to identify (an) antidote(s) of choice taking into account further aspects like cost, availability, shelf life and ease of administration for each of the different cyanide poisoning circumstances:

- HCN and its salts (with immediate effect);
- cyanogenic compounds including cassava, nitriles, and nitroprusside (delayed effect);
- fire smoke: carbon monoxide [CO] and cyanide (mixed intoxication);
- poisoning with an (initially) unknown substance.

The target audience was identified as medical professionals (occupational physicians, nurses, paramedics and others), public health decision makers and chemical companies (including ECETOC members).

2.2 Terms of reference

- Review the available human and clinically relevant experimental literature and other available information (e.g. from industry and poison centres, including national reviews) on the use of antidotes for the treatment of acute cyanide poisoning.
- Evaluate the literature, summarising – for the four conditions [circumstances] of poisoning above – the evidence on efficacy and safety of these antidote treatments, and comment on their stability, storage, availability, and practice of use.
- Identify gaps in knowledge.
- Based upon the available information, make a scientific judgement as to whether or not (an) antidote(s) of choice can be recommended for each of the different types of poisoning.
- Write a report and advice on an appropriate communication / information strategy.

2.3 Pathophysiology, pharmacokinetics and antidotal mode of action for cyanide

This is a brief overview of cyanide pathophysiology and pharmacokinetics as far as they are relevant and necessary for an understanding of the mode of action of cyanide antidotes. For further details referral is made to the ECETOC JACC report no. 53.

Hydrocyanic acid (HCN) and cyanide-salts may be absorbed readily via inhalation, through the skin or by ingestion. Intoxications can occur accidentally, for example during use in a laboratory or industry, or with suicidal intention. The circumstances under which the exposure occurred are herin referred to as 'intoxication circumstances'. These factors often have significant influence on the time course of events and often the overall severity of the intoxication. Inhalation of HCN as gas is highly toxic and rapidly acting. Poisoning by dermal exposure is not as rapid and severe as if the poison is ingested orally since the skin retards absorption. Accidents with cyanides, either in laboratories or industry, usually do not result in lethal intoxications, as exposure may be limited or decontamination and/or therapeutical actions are taken immediately. In contrast, suicidal poisonings are often severe or fatal due amounts ingested, that are considerably in excess of minimum lethal doses. Individuals working in electroplating, precious metal mining, gold smithing and to a lesser extent chemists, biochemist and pharmacists working that work with cyanides are more prone to occupational cyanide poisoning. Since the marketing of cyanides is strictly regulated, relatives of these people tend to have more ready access to cyanide for suicide intentions than the general public.

In the body cyanide usually can be found as HCN (as it has a pK_a of 10^{-9}). The cyanide-ion (CN^-) has high affinity for and binds strongly to the central metal ion (Fe^{3+}) of metallo-enzymes to form cyanide complexes thus rendering those enzymes non-functional. As such cyanide is a potent inhibitor of cellular (mitochondrial) respiration by reversibly binding to the ferric ion of

the cytochrome aa$_3$ within the mitochondria (Ballantyne, 1987). Cyanide more avidly combines with the reduced rather than the oxidised cytochrome oxidase.

Tri-valent iron in their mitochondria is essential for most cells as part of the cytochrome *c*-oxidase (complex four) of the respiration-chain. ATP and thus biologically usable energy is created by passing a pair of electrons down the respiratory chain from NADH to molecular oxygen:

$$NADH + H^+ + 3\ ADP + 3Pi + \tfrac{1}{2}\ O_2 \rightarrow NAD^+ + H_2O + 3\ ATP \quad \text{(Equation 1)}$$

Changing the charge from the ferrous to the ferric form within the cytochrome a$_3$, the terminal oxidative respiratory enzyme, the final electron is transferred in the respiratory chain:

$$2\ \text{cytochrome a}_3\ (\text{ox})\ (Fe^{2+}) \rightarrow 2\ \text{cytochrome a}_3\ (\text{red.})\ Fe^{3+};\ 2e^- + \tfrac{1}{2}\ O_2 + 2H^+ \rightarrow H_2O)$$
$$\text{(Equation 2)}$$
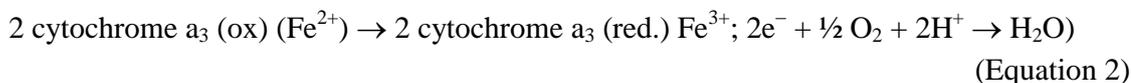
This reaction is blocked after cyanide intoxication. The higher the number of cytochrome *c* oxidase is in the cells the more cyanide will be there. So tissues with high energy consumptions as brain, kidney or liver are more susceptible to cyanide toxicity. As cyanide blocks the respiration chain no more ATP can be obtained and this leads to the typical 'inner asphyxia' (see above). This means that oxygen is present in the body but cannot be used for energy-generation in the cells. The patients appear clinically hypoxic, but at the same time there is no cyanosis of the skin because of high saturation even of the venous blood with oxygen. The difference between oxygen saturation in the arterial to the venous blood is decreased. As energy cannot be regained over the oxidative pathway any longer the cells have to switch to the anaerobic pathway. This results in a high lactate production and thus a metabolic (lactic) acidosis with a strong correlation between the severity of the cyanide intoxication and the blood-lactate levels (Baud et al, 2002). It should be rocognised, however, that lactic acidosis may be due to other clinical causes (cardiopulmonary arrest, extensive burns, respiratory impairment, methaemoglobinaemia) (Strickland et al, 1992), including other intoxications like CO (Katzman and Penney, 1993).

A much smaller part of tri-valent iron exists in the erythrocytes as methaemoglobin (MetHb). The use of MetHb-forming antidotes raises this fraction enabling CN$^-$ to be bound to Fe^{3+} because the pocket which protects Fe^{2+} in the haemoglobin (Hb) molecule is opened. In so doing cyanide is redistributed from the vital respiration-chain to the erythrocytes thereby decreasing the amount of free cyanide, which is the toxic agent.

Figure 1: Distribution of the cyanide ion in the body[a]

a ‹- - -› indicates equilibrium
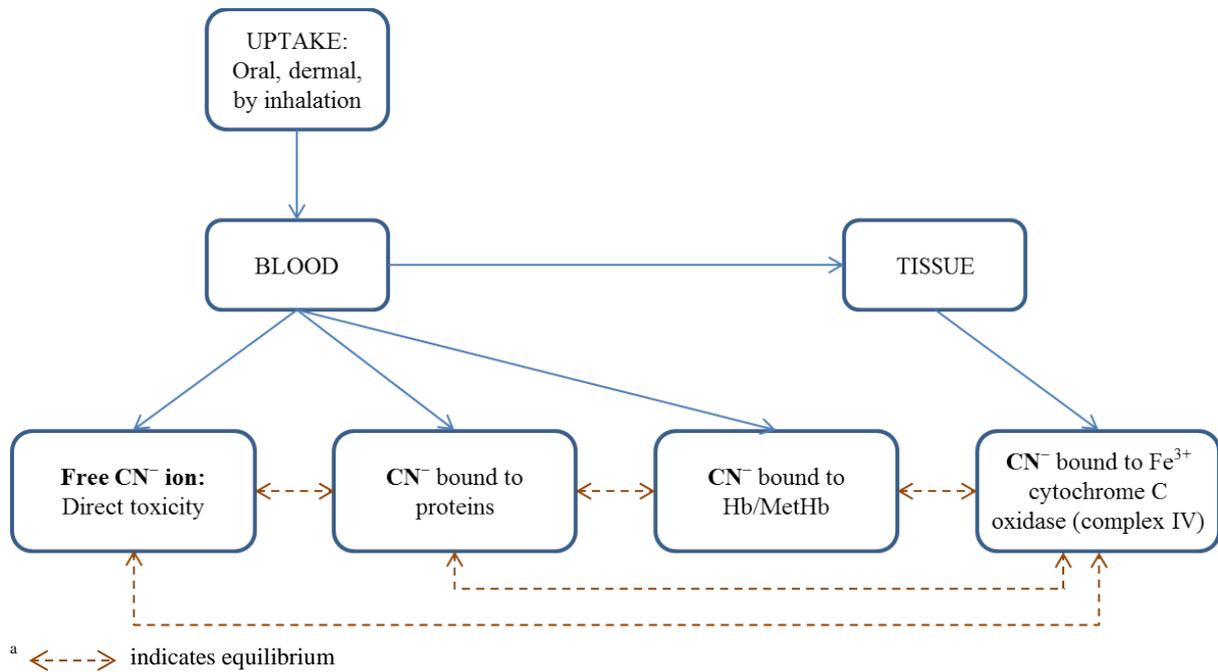
The majority of the cyanide (more than 90%) is detoxified in the liver by two sulphur-dependent enzymes called rhodanese and β-mercaptopyruvate sulphurtransferase. Both can be supported by using the antidote STS. Smaller amounts of cyanide can also be detoxified by using the cyanide-ion for physiological vitamin B_{12} formation.

Figure 2: Metabolism of cyanide and mode of action of cyanide antidotes
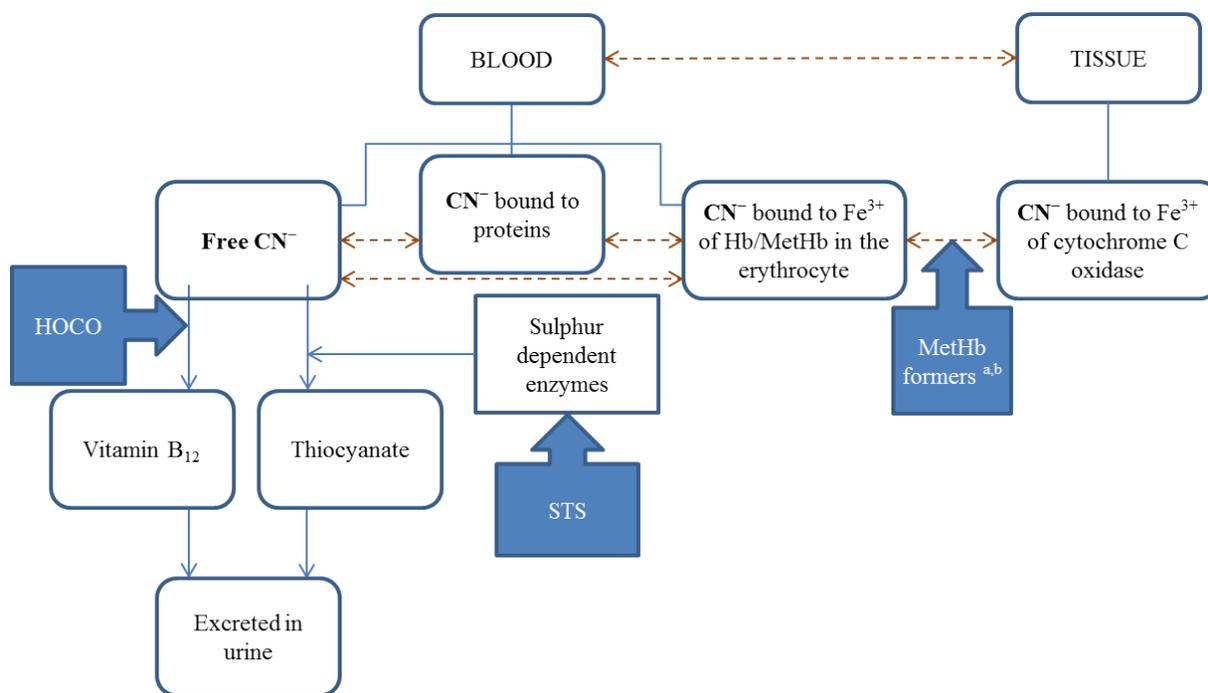
[a] MetHb formers (AN, SN, DMAP) shift this equilibrium quantitatively to the left (quantitative).
[b] CN^- is not excreted via this pathway.

Cyanide is converted to thiocyanate (SCN^-) in the liver and excreted in the urine with a half-life of 2.7 days (Schulz et al, 1979a). A smaller part (amount not exactly known) is excreted with the bile (Strickland et al, 1992). The main route of excretion of surplus vitamin B_{12} is through the kidneys.

The elimination of SCN^- and HOCO is hampered within patients who suffer from impaired liver and/or kidney function. The normal detoxification-capacity of the body is approximately 1 µg CN^-/kgbw/min (IPCS, 2004).

Figure 3: Elimination pathway for cyanide and its metabolites
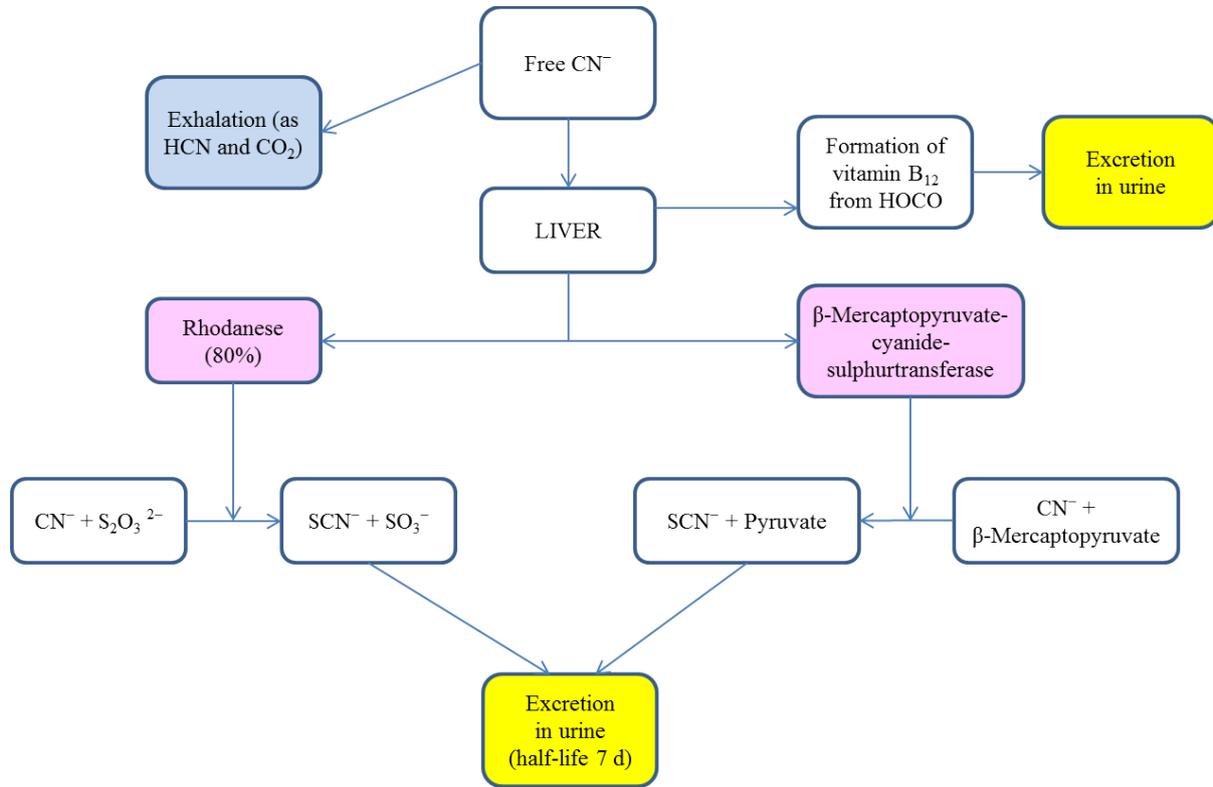
3. METHODOLOGY

Methodology includes the working method as well as the materials selected to address the terms of reference.

3.1 Selection of experts

Following expert nominations received from ECETOC member companies and through WHO-IPCS upon invitation by ECETOC's Secretary General, the ECETOC Scientific Committee appointed 7 members of the Task Force from academia and industry. The composition of the Task Force is listed at the end of this report.

3.2 Procedure

The Task Force held 14 one-day meetings to define a work plan, collect the literature and other information (see below), adapt the Poison Severity Score (PSS) (Persson et al, 1998) to cyanide poisoning (PSSa; defined in Section 3.4.4), agree statistical analysis of the sequential PSSa by antidotes and by poisoning circumstances, format a questionnaire, review and discuss draft chapters / sections of the report and agree final conclusions and recommendations.

For each antidote currently available information was gathered and efficacy and safety were assessed for the different conditions of poisoning.

For each condition of poisoning, the reported uses of antidotes (singly or in combination) were assessed accounting for availability, required speed of action, effectiveness and safety in order to arrive at an antidote recommendation.

The draft Task Force report was subject to peer review by ECETOC Scientific Committee members and external reviewers (both listed at the end of the report). The final report version was adopted after some minor amendments requested by the reviewers. Following final editing, the Secretariat published the report.

3.3 Materials

A search of the literature was made in 2008 and last updated by the middle of 2010, using the following key-words: "Poisonings or overdose or overdosage or intoxication" and one of the following items: "Hydrocyanic Acid; Hydrogen Cyanide; Cyanides; Nitriles; Acetonitrile; Propionitrile; Laetrile; Bitter Almond; Amygdalin; Nitroprusside; Cyanogenic compounds;

Oxygen; Nitrites; Thiosulphate / Thiosulfate; Dicobalt edetate; Dimethylaminophenol; Hydroxocobalamin; alpha-Ketoglutarate" in PubMed, Embase, Scopus, Toxline, French Poison Control Centres database, and since August 2000 "Current Awareness in Toxicology". A manual selection was performed to select publications meeting the inclusion criteria. Language was not a limitation for initial selection of the papers, which were reviewed if in English, French, German, Dutch, Italian, Spanish, Portuguese, Danish or Swedish. Other languages, especially Russian and Asian languages were excluded, if they had not been translated nor had an English abstract available. Particular attention was paid to avoid duplicates suggested by the same age, sex, city, and common authors in a reported case.

References and case reports that provided individual patient data were eligible for the study. In publications on series of cyanide poisonings, the publications were only considered if they provided individual data applicable to at least a single case.

In addition, unpublished cases from chemical companies or Poison Centres were collected, either self-reported with individual data or using the questionnaire developed by the Task Force (Appendix A). This was sent to chemical companies and Poison Information Centres. Four personal, unpublished cases were added as a result of this process.

3.3.1   Criteria for exclusion

Cases of poisoning resulting from exposure to cyanide derivatives not known to result in cyanide poisoning (e.g. ferrocyanide) and the unrelated substances cyanamide, cyanate, and isocyanate were not included. Furthermore, cyanide poisonings due to tobacco smoke inhalation were not included.

Experimental studies in animals and *in vitro* data were generally not included, and only considered in the absence of sufficient human data. Reports on analytical (measurement) methods were also excluded from this review. A list of all unquoted references is given in the second part of the bibliography.

3.4   *Methods*

A review and evaluation of the available human and clinically relevant experimental literature was performed by antidotes and by conditions. When data were lacking in humans, limited relevant experimental data were collected to ensure inclusion of basic knowledge. Gaps of knowledge were identified, corresponding to either the complete lack of data, data existing only in experimental animal studies, and data on combinations of antidotes regarding synergistic effects and sequence of application.

Summaries of casuistics are presented in Appendix C for all antidotes and poisoning circumstances.

3.4.1 By antidote

In this chapter the inherent properties, mode of action, pharmaceutical properties, practicality, efficacy and safety of the currently applied antidotes as well as any identified gaps in knowledge are described and discussed in a common twelve section format.

1. Introduction.
2. Identity.
3. Physico-chemical and pharmaceutical properties.
4. Practicality (use restrictions due to shelf-life, storage condition, stability).
5. Pharmacokinetics and metabolism (of the antidote: experimental and human data).
6. Mode of action (of the antidote), dosage and time of onset (of therapeutic principle).
7. Safety (toxicology – reported toxic and other adverse effects, including excipients).
8. Case series and casuistics [a] of cyanide / cyanogenic poisonings treated with the antidote (alone and in combination).
9. Occurrence (frequency and severity) of adverse effects in clinical use.
10. Assessment of efficacy and effectiveness [b] (of the antidote) in clinical use.
11. Gaps in knowledge.
12. Conclusion.

Detailed casuistics were assigned to Appendix C.

3.4.2 By poisoning circumstance

For the four poisoning circumstances – HCN and CN salts, cyanogenic compounds, mixed intoxications, and poisoning with unknown substances (categorised as cases with subsequent information that CN⁻ was involved, and cases in which no information on the causative poison was ever found) – the TF gathered information for description and discussion in a common nine section format.

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[a] Casuistics is the recording and study of cases of any disease. Case is an occurrence of a disease or disorder.

[b] The distinction between efficacy and effectiveness is explained in Section 3.4.4.

1. Introduction.
2. Identity.
3. Pharmacokinetics and toxicity.
4. Casuistics [a] (table and characteristic single cases).
5. Treatment and/or prophylaxis.
6. Practicality of antidote(s).
7. Occurrence (frequency and severity) of adverse effects in clinical use.
8. Assessment of efficacy and effectiveness of antidotes in clinical use.
9. Gaps in knowledge.
10. Discussion and conclusion.

Detailed casuistics were assigned to Appendix C.

3.4.3 Practicality

Aspects relating to practicality i.e. ease of practical use included recommendations by the supplier(s) for the following:

- Storage temperature / need for refrigeration etc.
- Protection from light.
- Shelf-life.
- Availability in different regions of the world, including regional cost aspects, if available.
- Ease of administration by non-clinically trained persons.

3.4.4 Assessment of efficacy and effectiveness

For the purpose of assessment, efficacy of an antidote should be distinguished from its effectiveness in clinical use and can be defined as follows:

- Efficacy (adjective efficacious) is the capacity to produce an effect. In this context, efficacy shows whether an antidote does work or does not work to counteract cyanide. In other words, it is the ability of an antidote to produce the desired detoxification effect, in expert hands and under ideal circumstances. This is shown by the sequential PSSa database (Section 3.4.4).

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[a] Casuistics is the recording and study of cases of any disease. Case is an occurrence of a disease or disorder.

- Effectiveness (adjective effective) refers to the ability of a drug [antidote] to produce a beneficial effect, under typical use circumstances. In this context, effectiveness shows that an antidote actually works in clinical use with regard to dosage, timing, practicality, and costs.

Note: Efficiency (adjective efficient) is, in general, the extent to which time or effort is well used for the intended task or purpose. It is a relative term which, in this context, describes the efficacy relative to a drug (antidote) which has the highest observed efficacy.

The Poisoning Severity Score (PSS) is a classification scheme for grading cases of poisoning in humans. The PSSa was introduced to enable an overall uniform evaluation of the cases, taking into account the most severe clinical features during the course of the poisoning and outcome (Persson et al, 1998). The author's system for grading the severity of acute poisoning was also used in this review (Table 2). The criteria are given in Table 3.

Table 2: Severity grades

Grade	Score	Severity
None	0	No symptoms or signs related to poisoning
Minor	1	Mild, transient, and spontaneously resolving symptoms or signs
Moderate	2	Pronounced or prolonged symptoms or signs
Severe	3	Severe or life-threatening symptoms or signs
Fatal	4	Death

The most severe signs or symptoms in any of the organs systems affected determine the overall severity of the case.

Table 3: Scheme for describing the severity of poisoning (Persson et al, 1998)

Organ	None (0)	Minor (1)	Moderate (2)	Severe (3)	Fatal (4)
			Severity (score) [a]		
GI-tract		• Vomiting, diarrhoea, pain	• Pronounced or prolonged vomiting, diarrhoea, pain, ileus	• Massive haemorrhage, perforation	
		• Irritation, first degree burns, minimal ulcerations in the mouth	• First degree burns of critical localisation or second and third degree burns in restricted areas	• More widespread second and third degree burns	
			• Dysphagia	• Severe dysphagia	
		• Endoscopy: Erythema, oedema	• Endoscopy: Ulcerative transmucosal lesions	• Endoscopy: Ulcerative transmural lesions, circumferential lesions, perforation	
Respiratory system		• Irritation, coughing, breathlessness, mild dyspnoea, mild bronchospasm	• Prolonged coughing, bronchospasm, dyspnoea, stridor, hypoxemia requiring extra oxygen	• Manifest respiratory insufficiency (e.g. severe bronchospasm, airway obstruction, glottal oedema, pulmonary oedema, ARDS [b], pneumonitis, pneumonia, pneumothorax)	
		• Chest X-ray: Abnormal with minor or no symptoms	• Chest X-ray: Abnormal with moderate symptoms	• Chest X-ray: Abnormal with severe symptoms	
Nervous system		• Drowsiness, vertigo, tinnitus, ataxia	• Unconsciousness with appropriate response to pain	• Deep coma with inappropriate response to pain or unresponsive to pain	
			• Brief apnoea, bradypnoea	• Respiratory depression with insufficiency	
		• Restlessness	• Confusion, agitation, hallucinations, delirium	• Extreme agitation	
			• Infrequent, generalised or local seizures	• Frequent, generalised seizures, status epilepticus, opisthtotonus	
		• Mild extrapyramidal symptoms	• Pronounced extrapyramidal symptoms		
		• Mild cholinergic / anticholinergic symptoms	• Pronounced cholinergic / anticholinergic symptoms		
		• Paraesthesia	• Localised paralysis not affecting vital functions	• Generalised paralysis or paralysis affecting vital functions	
		• Mild visual or auditory disturbances	• Visual and auditory disturbances	• Blindness, deafness	

Table 3: Scheme for describing the severity of poisoning (Persson et al, 1998) (cont'd)

Organ	None (0)	Minor (1)	Moderate (2)	Severe (3)	Fatal (4)
			Severity (score) [a]		
Cardio-vascular system			• Sinus bradycardia (HR [c] 40 - 50 in adults, 60 - 80 in infants and children, 80 - 90 in neonates)	• Severe sinus bradycardia (HR < 40 in adults, < 60 in infants, < 80 in neonates)	
			• Sinus tachycardia (HR 140 - 180 in adults, 160 - 190 in infants and children, 160 - 200 in neonates)	• Severe sinus tachycardia (HR > 180 in adults, > 190 in infants and children, > 200 in neonates)	
		• Isolated extrasystoles	• Frequent extrasystoles, atrial fibrillation / flutter, AV [d]-block I-II, prolonged QRS [e] and QT_c [f]-time, repolarisation abnormalities	• Life-threatening ventricular dysrhythmias, AV-block III, asystole	
			• Myocardial ischaemia	• Myocardial infarction	
		• Mild and transient hypo / hypertension	• More pronounced hypo / hypertension	• Shock, hypertensive crisis	
Metabolic balance		• Mild acid-base disturbances (HCO_3^- 15 - 20 or 30 - 40 mmol/l, pH 7.25 - 7.32 or 7.50 - 7.59)	• More pronounced acid-base disturbances (HCO_3^- 10 - 14 or > 40 mmol/l, pH 7.15 - 7.24 or 7.60 - 7.69)	• Severe acid-base disturbances (HCO_3^- < 10 mmol/l, pH < 7.15 or > 7.7)	
		• Mild electrolyte and fluid disturbances (K^+ 3.0 - 3.4 or 5.2 - 5.9 mmol/l)	• More pronounced electrolyte and fluid disturbances (K^+ 2.5 - 2.9 or 6.0 - 6.9 mmol/l)	• Severe electrolyte and fluid disturbances (K^+ < 2.5 or > 7.0 mmol/l)	
		• Mild hypoglycaemia (50 - 70 mg/dl or 2.8 - 3.9 mmol/l in adults)	• More pronounced hypoglycaemia (30 - 50 mg/dl or 1.7 - 2.8 mmol/l in adults)	• Severe hypoglycaemia (< 30 mg/dl or 1.7 mmol/l in adults)	
		• Hyperthermia of short duration	• Hyperthermia of longer duration	• Dangerous hyper- or hypothermia	
Blood		• Mild methaemoglobinaemia (MetHb 10 - 30%)	• More pronounced methaemoglobinaemia (MetHb 30 - 50%)	• Severe methaemoglobinaemia (MetHb > 50%)	
		• Mild haemolysis	• Haemolysis	• Massive haemolysis	
			• Coagulation disturbances without bleeding	• Coagulation disturbances with bleeding	
			• Anaemia, leukopoenia, thrombocytopoenia	• Severe anaemia, leukopoenia, thrombocytopoenia	
Liver		• Minimal rise in serum enzymes (AST, ALT 2 - 5 x normal)	• Rise in serum enzymes (AST, ALT 5 - 50 x normal) but no diagnostic biochemical (e.g. ammonia, clotting factors) or clinical evidence of liver dysfunction	• Rise in serum enzymes (> 50 x normal) or biochemical (e.g. ammonia, clotting factors) or clinical evidence of liver failure	

Table 3: *Scheme for describing the severity of poisoning* (Persson et al, 1998) (cont'd)

Organ	None (0)	Minor (1)	Moderate (2)	Severe (3)	Fatal (4)
			Severity (score) [a]		
Kidney		• Minimal proteinuria or haematuria	• Massive proteinuria or haematuria		
			• Renal dysfunction (e.g. oliguria, polyuria, serum creatinine of 200 - 500 µmol/l)	• Renal failure (e.g. anuria, serum creatinine of > 500 µmol/l	
Muscular system		• Mild pain, tenderness	• Pain, rigidity, cramping and fasciculations	• Intense pain, extreme rigidity, extensive cramping and fasciculations	
		• CPK [g] 250 - 1,500 IU [h]/l	• Rhabdomyolysis CPK 1,500 – 10,000 IU/l	• Rhabdomyolysis with complications CPK > 10,000 IU/l	
				• Compartment syndrome	
Local effects on skin, eye, from bites and stings		Not applicable [i]	Not applicable [i]	Not applicable [i]	

[a] Described in Table 2
[b] Acute respiratory distress syndrome
[c] Heart rate
[d] Atrioventricular
[e] QRS complex (on electrocardiogram)
[f] QT interval corrected for heart rate
[g] Creatine phosphokinase
[h] International unit
[i] Therefore not reproduced here

This PSSa was adapted by the Task Force to cyanide poisoning symptoms, including organ dysfunction and failure induced by cyanide(s). Some parameters were omitted as not relevant for cyanide poisoning and other relevant parameters were added (Table 4). Efficacy and safety were addressed separately, using criteria established for the treatment of snakebites (Dart et al, 2001). In the case of safety assessment, blood methaemoglobin (MetHb) was included.

With its range from 0 (asymptomatic) to 4 (fatal), the 'PSSa adapted to cyanide poisoning' (PSSa) was used to assess the severity of cyanide poisoning at the time of presentation, after antidote administration (or after each antidote if administered in a sequence) and at discharge. For each sequence, the PSSa was based upon the parameter resulting in the highest score.

Table 4: Scheme for describing the severity of cyanide poisoning[a] using an adapted PSS (PSSa)

Organ	None (0)	Minor (1)	Moderate (2)	Severe (3)	Fatal (4)
			Severity (score)[b]		
GI-tract		• Vomiting, diarrhoea, pain	• Vomiting, diarrhoea, pain, ileus • Dysphagia		
Respiratory system		• Irritation, coughing, breathlessness, mild dyspnoea, mild bronchospasm	• Coughing, dyspnoea, bronchospasm, stridor, *tachypnoea* • Hypoxaemia requiring extra oxygen	• Manifest respiratory insufficiency, *bradypnoea, apnoea, cyanosis*	
		Chest X-ray: Abnormal with minor or no symptoms	• Chest X-ray: Abnormal with moderate symptoms	• Chest X-ray: Abnormal with severe symptoms	
Nervous system		• Drowsiness, vertigo, tinnitus, ataxia	• Unconsciousness with appropriate response to pain, *GSC[c] > 8*	• Deep coma with inappropriate response to pain or unresponsive to pain, *GSC ≤ 8* • Tendon reflexes absent	
		• Restlessness	• Confusion, agitation, *restlessness*, hallucinations, delirium	• Extreme agitation	
			• Infrequent, generalised or local seizures	• Frequent generalised seizures, status epilepticus, opisthtonos	
		• Paraesthesia	• Localised paralysis not affecting vital functions	• Generalised paralysis or paralysis affecting vital functions	
			• Brief apnoea	• Respiratory depression with insufficiency	
		• Visual or auditory disturbances	• Visual or auditory disturbances	• Blindness, deafness	
		• Extrapyramidal symptoms	• Extrapyramidal symptoms		
		• Cholinergic / anticholinergic symptoms	• Cholinergic / anticholinergic symptoms		

Table 4: Scheme for describing the severity of cyanide poisoning[a] using an adapted PSS (PSSa) (cont'd)

Organ	None (0)	Minor (1)	Moderate (2)	Severe (3)	Fatal (4)
			Severity (score) [b]		
Cardio-vascular system			• Sinus bradycardia (*HR [d] 40 - 50 in adults, 60 - 80 in children*)	• Sinus bradycardia (*HR ~ < 40 in adults, < 60 in children*)	
			• Sinus tachycardia (*HR 100 - 140 / minute*)	• Sinus tachycardia (*HR ~ > 140 in adults*)	
		• Isolated extrasystoles	• Frequent extrasystoles, atrial fibrillation / flutter, AV [e]-block I-II, prolonged QRS [f] and QT$_c$ [g]-time, repolarisation abnormalities	• Life-threatening ventricular dysrhythmias, AV-block III, asystole	
		• Hypotension: *SBP [h] ≤105 or 115 mm Hg and > 100 mm Hg*	• Hypotension: *SBP 90 - 100 mm Hg*	• Shock: *SBP < 90 mm Hg + other signs of shock*	
		• Hypertension: SBP > 140 - < 180	• Hypertension: *SBP 140 - 180*	• Hypertensive crisis	
			• Myocardial ischaemia	• Myocardial infarction	
Metabolic balance		• HCO$_3^{3-}$: 15 - 20 mmol/l, pH 7.25 - 7.32	• HCO$_3^{3-}$: 10 - 14 mmol/l, pH 7.15 - 7.24	• HCO$_3^{3-}$: < 10 mmol/l, pH < 7.15	
		• Electrolyte and fluid disturbances	• Electrolyte and fluid disturbances	• Electrolyte and fluid disturbances	
		• Hypoglycaemia	• Hypoglycaemia	• Hypoglycaemia	
		• Hyperthermia, sweating	• Hyperthermia	• Dangerous hyper- or hypothermia	
Blood		• MetHb [i] 10 - 30%	• MetHb 30 - 50%	• MetHb > 50%	
		• Haemolysis	• Haemolysis	• Haemolysis	
			• Coagulation disturbances	• Coagulation disturbances with bleeding	
			• Anaemia, leukopoenia, thrombocytopaenia	• Anaemia, leukopaenia, thrombocytopaenia	
Liver		• Serum enzymes 2 - 5 x VU [j]	• Serum enzymes 5 - 50 x VU	• Serum enzymes > 50 x VU + clinical or biochemical (ammonia, clotting factors) evidence of failure	
Kidney		• Minimal proteinuria or haematuria	• Massive proteinuria or haematuria		
			• Renal dysfunction: Oliguria, serum creatinine 200 - 500 µmol/l	• Renal failure: Anuria, creatinine > 500 µmol/l	

Table 4: Scheme for describing the severity of cyanide poisoning[a] using an adapted PSS (PSSa) (cont'd)

Organ	None (0)	Minor (1)	Moderate (2)	Severe (3)	Fatal (4)
			Severity (score) [b]		
Muscular system		• Mild pain, tenderness • CPK [k] 250 – 1,500 IU [l]/l	• Pain, rigidity, cramping and fasciculation • Rhabdomyolysis CPK 1,500 – 10,000 IU/l	• Intense pain • Rhabdomyolysis with complications CPK > 10,000 IU/l • Compartment syndrome	
Others		• *Pupils* • *Skin: Colour, burns* • *Corporal temperature*			

[a] Includes significant items from Table 3 with additions (made by the Task Force) marked in *italics*. For metabolic balance, blood, liver, kidney and muscular system, there are no real changes compared to Table 3. The parameters of these aspects were simplified and values were left out.
[b] Described in Table 2
[c] Gloucester severity score
[d] Heart rate
[e] Atrioventricular
[f] QRS complex on electrocardiogram
[g] QT interval corrected for heart rate
[h] Systolic blood pressure
[i] MetHb is considered relevant only for safety, not for efficacy. It is regarded as a potentially treatment-induced risk factor, not as a factor of severity of the intoxication
[j] Viscosimetric unit
[k] Creatine phosphokinase
[l] International unit

To assess efficacy / effectiveness and safety of antidotes the cases were analysed as to which antidote or antidote combination had been applied. In many cases where where more than one antidote or antidote combination had been given with intervals between successive treatments, each separate antidote or combination used, called 'a sequence', was analysed separately by using the PSSa. In this way a 'sequential PSSa' was determined.

To facilitate this analysis, clinical information had to be available before administration of the antidote(s) as well as during and/or just after the administration. Furthermore, the exact nature of the treatment, i.e. whether this was specific or just supportive had to be indicated.

Antidotes were used alone as well as in combination. In case of a combination of treatments being administered during a short period of time (minutes), the effectiveness of the therapy was assessed as a whole for the combination. This includes injection of a second antidote during one application or sequence, e.g. SN followed by STS. Particular attention was paid to oxygen and supportive treatment as both have been reported by a number of authors as being efficient without any additional antidotes. To address this major issue, the effectiveness of oxygen applied alone or in combination with supportive treatment, but without any additional antidotes, was assessed first. PSSa analysis of the data presently available shows that normobaric or hyperbaric oxygen, when applied alone or in combination, was not effective in cases of moderate and severe cyanide poisonings. Thus, oxygen was not further considered as a particular antidote to cyanide in the assessments of combinations of antidotes. However, we cannot completely exclude any antidotal effectiveness of (hyperbaric) oxygen, or the potentiation of the action of other antidotes (i.e. pharmacological synergism) as one limitation of our study was that we found only a very small number of cyanide poisoning cases reported in the medical literature in which hyperbaric oxygen was administered. Conversely, particular attention was paid to the efficacy of each antidote used either alone or in combination in patients not receiving oxygen.

Non-severely-poisoned patients cannot be considered for assessing antidote efficacy. Consequently, a PSSa above 0 before antidotal treatment was regarded as mandatory for assessing antidote efficacy. A mention is added regarding assessment of oxygen and supportive treatment without any additional antidotes. In assessment of combinations, the combination of antidotes and oxygen was not assessed, as oxygen was not regarded as an antidote any more after assessing cases treated with oxygen alone. Conversely, particular attention was paid to the efficiency of each antidotes and combination of antidotes used without oxygen.

The use of the sequential PSSa is based on the assumption that the effects of both supportive treatment and antidotes in cyanide poisoning are expected to occur very quickly, i.e. within minutes. A temporal correlation between antidote administration and the onset of improvement, side-effect, or deterioration, should be clearly indicated in the case report. This means that improvements as well as deterioration occurring several hours after the start of antidote were not

considered (by the Task Force) to be the result from of the antidote but rather from the spontaneous course of the poisoning. For cases of cyanogenic compound poisoning, attention was paid to data according to the expected time-course of the toxic events directly related to cyanide and the effects of treatments. Often in these cases antidotes were used to prevent expected symptoms (PSSa above 0 before antidotal treatment).

The sequential PSSa database is presented in Appendix B.

The database was analysed as follows. At the time the PSSa database was frozen, there were 400 cases in total (published and otherwise available cases till the 14th of March 2010). The flow chart of inclusion and non-inclusion of patients is shown in Table 4. Those found dead at the scene (11 cases), who received an unknown treatment (59 cases) or who were not treated (41 cases) were not included. Thus, 289 cases were retained for analysis. In the database, treatment used in cyanide poisoned could be described in no more than 5 sequences regarding the use of antidotes to cyanide. The distribution of the patients in the 5 sequences is presented, as follows.

Sequence 1

Comprising 287 cases. PSSa before treatment was unknown in 2 cases.

- No antidote: 103 (35.9%)
- Antidote unknown: 3 (1.0%)
- Antidotes: 181 (63.1%)

Sequence 2

Comprising 109 cases. PSSa before treatment was known in all cases.

- No antidote: 37 (33.9%)
- Antidote unknown: 4 (3.7%)
- Antidotes: 68 (62.4%)

Sequence 3

Comprising 46 cases. PSSa before treatment was known in all cases.

- No antidote: 13 (28.3%)

- Antidote unknown: 1 (2.1%)
- Antidotes: 32 (69.6 %)

Sequence 4

Comprising 16 cases. PSSa before treatment was known in all cases.

- No antidote: 6 (37%)
- Antidote unknown: none
- Antidotes: 10 (63%)

Sequence 5

Comprising 3 cases. PSSa before treatment was known in all cases.

- No antidote: none
- Antidote unknown: none
- Antidotes: 3 (100%)

Sequential overall efficacy was judged from the PSSa scores as follows:

- Complete: Decrease of PSSa from 1, 2, or 3 to 0
- Partial: decrease by at least one level but not returning to 0
- Lack of improvement: no change after antidotal treatment
- Deterioration: increase of at least one level

3.4.5   Additional cases for dicobalt edetate

After the PSSa database was frozen, additional case reports were received dealing with Co-EDTA. Eight publications reported data valuable for PSSa calculation in 15 cases having received Co-EDTA. Analysis of used antidotes in the 15 additional cases showed that Co-EDTA was used alone in 29 sequences and in combination with other antidotes in 18 sequences, including STS (2 cases), AN (3), SN (0), HOCO (3), and DMAP (1). The Task Force decided to analyse the overall efficacy of Co-EDTA, frequency and severity of toxicity and adverse events including these 15 additional cases. However, these few cases were not included in the PSSa data base that had already been closed as including them would have had no impact in the overall assessment of treatments and their outcomes.

4. ANTIDOTES

4.1 Oxygen

4.1.1 Introduction

The toxicity of cyanide is initiated by the potent inhibition of mitochondrial respiration with binding to the ferric ion of the cytochrome aa_3 within the mitochondria (Ballantyne, 1987). Furthermore, cyanide more avidly combines with the reduced rather than oxidised cytochrome oxidase, thus oxygen competes with cyanide to bind to reduced cytochrome oxidase. This suggested a promising role for oxygen in the treatment of cyanide poisoning, especially as the binding of cyanide with the ferric ion is fully reversible.

While the cellular and molecular mechanisms supported the potential efficacy of oxygen in cyanide poisoning; experimental studies resulted in somewhat conflicting reports, and only recently experimental results provided new insight on the role of oxygen in the treatment of cyanide poisoning.

In humans, evidence supporting the efficacy of oxygen as an antidote to cyanide remains limited. In the fifties and sixties, at a time when oxygen was not available at the scene while cyanide poisonings in plants were frequent, excellent results were obtained using antidotes without oxygen. Artificial respiration was advocated by the back pressure-arm lift as reported by De Forest and Potthoff (1951) and was a standard of care at the scene allowing the inhalation of AN sprayed on a handkerchief. There was no mention for oxygen (Chen and Rose, 1952). In the later revised version (Chen and Rose, 1956) oxygen was not recommended. However, it should be pointed out that one out of the two reported cyanide casualties by Chen and Rose in 1956 was given oxygen and carbon dioxide inhalation supplemented with AN inhalation. Goodman and Gilman (1956) did not mention oxygen therapy in cyanide poisoning except in connection with the treatment of excess nitrite-induced methaemoglobinaemia. Levine (1959) noted that in most textbooks oxygen was not mentioned or was advised only when respiration had ceased and artificial ventilation was required.

Presently, oxygen is a treatment used in both basic and advanced life support situations. As cyanide frequently and rapidly induces severe organ failures, including lactic acidosis, coma, respiratory arrest, and cardiovascular shock followed by cardiac arrest, oxygen is widely used on a symptom-to-treat basis. The apparent success of supportive treatment, including oxygen, resulted in a number of authors recommending oxygen as an antidote to cyanide. However, there have never been any controlled studies supporting such an assumption. So the question of whether oxygen is an antidote to cyanide is still unanswered. This is of utmost importance as in a number of developing countries; oxygen is not freely or immediately available. Furthermore, in a

chemical disaster potentially involving exposure of large numbers of individuals to cyanide it seems rather likely that oxygen supply will be limited.

4.1.2 Identity

IUPAC name:	Oxygen
CAS registry number:	7782-44-7
Formula:	O_2
Molecular mass:	31.9988
Chemical structure:	O=O

4.1.3 Practicality

Oxygen 93% is readily available as a compressed gas or liquid in appropriate cylindesr or pressurised storage tanks (USP 31) and therefore has no obvious restrictions on grounds other than cost and availability.

4.1.4 Pharmacokinetics and metabolism

The pharmacokinetics of oxygen in cyanide poisoning has not been addressed.

4.1.5 Mode of action, dosage and time of onset

Mitochondrial cytochrome oxidase activity and partial pressure of oxygen

Cyanide may combine with reduced cytochrome oxidase thereby competing with oxygen (Wainio, 1956; van Buuren et al (1972) studied the reaction of cyanide with the fully oxidised and reduced states of cytochrome aa_3. In both states a single molecule of cyanide is bound reversibly per molecule of aa_3 with the equilibrium constant for complex formation with the oxidised form (ferric aa_3 (KD)) being about 1 µM compared with about 100 µM for the reduced form (ferrous aa_3 (KD)). Therefore the reduced form of cytochrome oxidase, which predominates under conditions of low oxygen tension, is more readily inhibited by cyanide. In contrast, under conditions of high oxygen tension the oxidised form of cytochrome oxidase may be protected from the effect of cyanide. At a normal, physiological, intracellular oxygen tension of 40 mm Hg, the cytochrome oxidase activity may have only 60% of its potential activity with maximal activity not being reached until the intracellular oxygen tension excedes 70 mm Hg (Way, 1984). In other words, at least 30 mm Hg of extra oxygen tension may be required to saturate a cell from a respiratory standpoint (Cope, 1961).

While *in vitro* data such as that produced by Warburg (1927) and Isom and Way (1984) tend not to support a mechanism of action involving directly the interaction of oxygen with cytochrome oxidase, *in vivo* data by Gordh and Norberg (1947) in rabbits show that low S_VO_2 in advanced cyanide poisoning does.

Experimental studies

Rabbits were exposed to hydrocyanic acid vapour to the point of failing cardiac activity / respiratory arrest before administering either room air or oxygen (Gordh and Norberg, 1947). Blood was collected by cardiac puncture and saturated blood oxygen level determined as 32.5 $\pm$ 18.0% (mean $\pm$ s.d., range 5 - 64%, n=16). Administration of oxygen considerably increased the tolerance to cyanide. While animal breathing oxygen inhaled a larger amount of HCN until cardiorespiratory failure set in, blood HCN and thiocyanate concentrations were not significantly increased, suggesting that oxygen fascilitated the detoxification of HCN by a route other than though formation of thiocyanate.

In rats poisoned with KCN administered i.v., slightly more cyanide was required to induce brain damage if oxygen was administered than in the animals than if the experiment was conducted in air. Studies in mice showed that the LD_{50} of KCN was increased by 10 to 20% by administration of oxygen; survival time after fatal doses was also increased while data were not presented (Levine 1959)

In a dog model of cyanide poisoningoxygen increased the survival rate of poisoned animals. At the time of the onset of apnoea, the saturation of arterial blood in oxygen was very high but then rapidly decreased. There was a delayed onset of tissue hypoxemia related to cardiovascular failure. The administration of oxygen at the time of onset of cardiac failure prevented the decrease in oxygen saturation of arterial blood and promoted the transfer and delivery of oxygen to cells (Paulet 1955),

A prompt improvement in EKG abnormalities was observed in dogs intravenously (i.v.) poisoned with cyanide after 100% oxygen intermittently, as opposed to air. Also, cyanide-poisoned goldfish survived longer when placed in fresh water with bubbled oxygen (Cope, 1961)

In mice administered a lethal dose of KCN i.p., 100% oxygen protected against the lethal effect of KCN. However, similar studies, in sheep provided only modest, if any, protection against the lethal effect of NaCN (Burrows and Way, 1977).

Treatment with 95% of oxygen failed to prevent the onset and magnitude of inhibition of liver cytochrome oxidase induced by the intraperitoneal administration of 5 mg/kg KCN although the time course of inhibition was shortened by about 10 minutes when compared to mice breathing air In brain tissues, the EC_5 (concentration required to produce 5% inhibition of cytochrome

oxidase) was increased from 24 mg/kg (range 16.6 to 34.8) to 55 mg/kg (range 39.3 to 77.0) (Isom et al., 1982).

The efficiency of various antidotes was studied in anaesthetised, mechanically ventilated, dogs that had been poisoned with KCN. Administration of normobaric oxygen resulted in a moderate, but statistically significant increase in the observed lethal dose of KCN from 2.4 ± 0.2 to 3.2 ± 0.4 mg/kg (Ivankovich et al., 1980).

Oxygen in blood: the two phases of cyanide poisoning

During the course of near-lethal hydrocyanic acid poisoning in rabbits, the oxygen saturation in the blood sank to values that were incompatible with life (Gordh and Norberg, 1947).

The biphasic time-course of the venous oxygen saturation in anaesthetised dogs poisoned with a lethal dose of KCN started after the initiation of cyanide infusion with a progressive increase in oxygen saturation in both the mixed venous blood and the coronary sinus blood. In the late phase, shortly before death, parallel to the blood pressure decreasing rapidly, oxygen saturation in both the mixed venous blood and the coronary sinus blood decreased (Mercker et al. 1958).

Paulet (1984) outlined that. The role of oxygen and its efficacy seems to depend on the stage of poisoning with the main factor for success being the speed of application, preferably being administered at the earliest possible moment (Cope, 1961). In the late phase of cyanide poisoning venous oxygen desaturation, resulting from cardiac failure and respiratory arrest, may account for the observed lack of efficacy of oxygen during this period. Oxygen should be given as soon as possible in suspected cyanide poisoning (Paulet 1984).

These studies allow for the conclusions:

-   If blood pressure and breathing are adequate, the subject will probably recover without any further treatment. It is interesting to note that in a normal person, cyanide-induced hyperventilation can raise the arterial oxygen tension up to 110 to 120 mm Hg; this rise in oxygen tension most certainly helps to block the cyanide reaction (Cope, 1961).
-   If the pulse is present but breathing has stopped, immediate artificial respiration with oxygen would be life-saving. The additional use of vasopressor agents to maintain blood pressure will allow a good chance of recovery.
-   If there is apnoea and the heart has just stopped beating, immediate artificial respiration with oxygen in combination with cardiac massage should be tried in an attempt to restore heart action and blood pressure.

A study of cyanide poisoning in dogs showed two phases of injury. The first compensated phase was consistent with a traditional global oxygen consumption defect. The second, decompensated, phase had a mechanism consistent with cardiac failure. This was due to bradycardia as stroke volume remained unchanged. These data suggest that, at least in the model, cardiac contractility was preserved (Pham et al. 2007).

The effects of carbon monoxide and cyanide were studied in anaesthetised, endotracheally intubated, mechanically ventilated dogs. Carbon monoxide exposure resulting in COHb of 45.9 $\pm$ 6.4% had neither effect on any haemodynamic parameters nor on any metabolic, including PvO_2, or acid-base variables. In contrast, cyanide infusion resulting in peak blood cyanide concentration of 162 $\pm$ 27.1 µmol/l produced a significant decrease in cardiac output from baseline value of 6.4 l/min to 3.1 $\pm$ 0.5 l/min at the peak effect. There were significant increases in pulmonary artery pressures and resistance, and pulmonary wedge pressure increased from 8.1 $\pm$ 2.7 mm Hg to 15.2 $\pm$ 5 mm Hg. During cyanide infusion $PvCO_2$ was significantly decreased, while plasma lactate significantly increased from 1.2 $\pm$ 0.7 mmol/l to 4.7 $\pm$ 2.5 mmol/l. During CO inhalation arterial content in oxygen (CaO_2) significantly decreased from 15.7 $\pm$ 4.2 ml% to 10.0 $\pm$ 2.5 ml% and then increased during cyanide infusion to 13.0 $\pm$ 3.5 ml%. The changes in mixed CvO_2 paralleled those of CaO_2. After stopping CN infusion the blood cyanide half-lives followed a two-compartment model with an initial half-life of 62.3 minutes followed by a late elimination half-life of 129 minutes ($r^2 = 0.999$). The decrease of COHb during air ventilation followed a mono-compartment model with a half-life of 114 minutes (Breen et al. 1995a).

Other supposed mechanisms of action of oxygen have been proposed including the possibility that supplemental oxygen may bypass the intra-mitochondrial cytochrome activity and supply oxygen needed to continue metabolic functions through some alternative cyanide-resistant pathway (Litowitz 1987).

In experimental conditions, oxygen was shown to synergistically increase the antidotal potency of the combination of SN and STS with minor (thiosulphate) and no effect (nitrite) administered separately (Way, 1984). It should be noted that the enzymatic conversion of cyanide to thiocyanate in the presence of Rhodanese can occur without oxygen (Way, 1984).

4.1.6 Safety

Toxicology

Although Bond (1962) suggested that formation of oxidised degradation products (oxygen reactive species) may augment cyanide toxicity this has not been reported elsewhere.

Human safety considerations

The duration of effective oxygen therapy in cyanide poisonings has not been determined. However, considering the relatively short duration of oxygen administration compared to the risk to health posed by cyanide poisoning precludes any concern regarding oxygen.

When treating individuals with a special sensitivity to oxygen toxicity, such as neonates and individuals with chronic obstructive lung disease, the risk / benefit ratio should be assessed in the context of the severity of cyanide poisoning.

The adverse events related to hyperbaric oxygen are non-specific and may equally occur in cases of cyanide poisoning treated with, or without, hyperbaric oxygen.

4.1.7 Case series and casuistics of cyanide / cyanogenic poisonings treated with or without oxygen

The review of the literature showed two consecutive periods regarding the use of oxygen in cyanide poisoning. During the fifties and sixties, oxygen was not considered as a first-line treatment; the priority was the administration of antidotes to cyanide (Chen and Rose, 1952 & 1956; Wolfsie, 1951). In latter years oxygen has been included on the list of antidotes to cyanide poisoning. This resulted from experimental data as well as the infrequent use of the other antidotes, the onset of potentially severe antidote-related adverse events, and case reports. Simultaneously, the systematic use of oxygen in basic as well as advanced life supported recommendations in organ failure following cyanide poisoning. Consequently, oxygen has been used as early phase treatment for cyanide poisoning.

According to literature cyanide poisoning victims can recover without oxygen and without antidotes, using only withdrawal from the source of cyanide, decontamination and supportive treatment not including oxygen. However, there are neither retrospective case-control studies nor prospective randomized studies supporting the assumption that oxygen either alone or in combination with supportive treatment results in recovery. It can be assumed that only successes and not failures of oxygen and supportive treatment have been reported. Rrecovery was associated with the administration of antidotes more frequently with rather than without oxygen.

Cyanide poisonings treated without oxygen

Wolfsie et al (1951) reported on 12 cases of known CN poisonings occurring as a consequence of occupational exposure.for which oxygen was not a first-line treatment. The use of oxygen was not mentioned in 11 out of the 12 cases while the remaining case received oxygen during 1 minute only. Regarding the severity of poisoning according to the PSS, 6 were moderate and six were severe. The patient treated with 1 minute oxygen was classified as severe. All the 11 patients not receiving oxygen but treated with other antidotes to cyanide recovered without obvious sequelae.

Chen et al (1952 & 1956) reported a total of 18 cases of known CN poisonings occurring as a consequence of occupational exposure. The use of oxygen was not mentioned in 8 out of the 18 cases. Of the patients that were not treated with oxygen, regarding the severity of poisoning according to the PSS, the PSSa cannot be assessed for one patient, while one was minor, and not considered for further evaluation (both fully recovered). Poisoning was moderate in 4 cases, 3 fully recovered while one experienced sequelae. Poisoning was severe in 3 cases, one fully recovered while the others recovered with sequelae.

Lurie et al (1953) reported a case of severe CN poisoning who completely recovered while not receiving oxygen.

Patients treated with oxygen alone or in combination with supportive treatment, including cardiopulmonary resuscitation but without additional antidotes

There were a number of single case reports and limited series describing the use of oxygen in cyanide poisoning. One major problem was the authors focusing on the effect of oxygen while the patients received a combination of treatment. It should be noted that the greater the severity, the larger the number of associated treatment.

A retrospective study of 25 cases of cyanide poisoning indicated: cardiorespiratory arrests are frequent (7/25); in severe intoxications (7/25) deep metabolic acidosis is the rule, and cyanide poisoning should always be suspected in cases of coma with severe acidosis; mild intoxications are frequently symptomless. Anxiety and agitation should not be considered as evidence of cyanide poisoning; they are merely due to fear in most cases. The present treatment of acute cyanide poisoning relies basically on symptomatic measures including sodium bicarbonate, circulatory support and, above all, assisted ventilation with 100% oxygen. The author's experience did not support the concept of a lethal cyanide blood level when patients can rapidly be transferred by a medical team to an intensive care unit. Survival depends more on prompt medical care than on the accessibility to sophisticated antidotes (Bismuth et al, 1984a&b).

4.1.8   Assessment of efficacy and effectiveness in clinical use

This denotes patients receiving oxygen administered either alone by nasal prongs, facial mask, without manual ventilation or with an Ambu-bag as well as combination of mechanical intubation or mechanical ventilation, other supportive treatment, and even cardiopulmonary resuscitation with oxygen.

Regarding treatment in cyanide poisonings with a particular attention to oxygen and other antidotes, it is important to identify and compare the efficacy of the treatment for each separate sequence (sequential antidote treatment, or combination of treatments) in the different sequences for groups of patients that received different combinations of treatment:

Group	Treatment
1	Neither oxygen nor antidotes
2	No oxygen but received antidotes.
3	Oxygen without antidotes
4	Oxygen and antidotes
5	Hyperbaric oxygen with/without antidotes

Analysis was conducted following the flow chart in Figure 4.

Figure 4: Flow chart of patients receiving oxygen
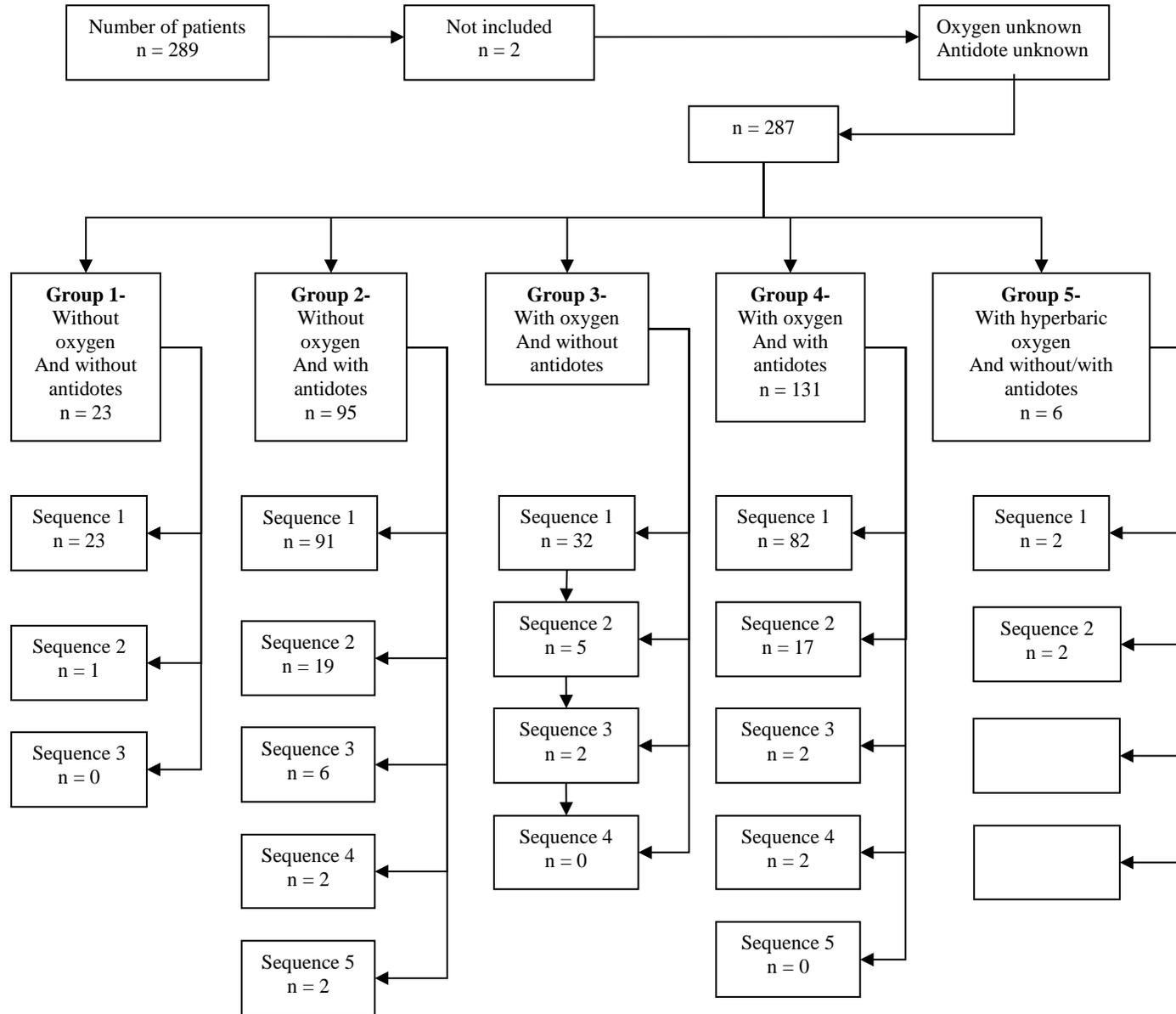
Each patient is counted only once in the flow chart. Classification is made on the basis of the sequence reporting either oxygen (Yes/No) and/or antidotes (Yes/No).

Sequences in patients having received neither oxygen nor antidotes (Group1)

In the medical literature, 23 cases of cyanide poisoning were reported while not having received normobaric oxygen and not having received any other antidotes. All twenty-three cases were assessed in sequence 1. The median PSSa before treatment was 2. They spontaneously completely recovered as evidenced by a PSSa of 0following treatment. The improvement was statistically significant ($p = 0.0011$).

> *i.e. Sequence 1* n = 23 $p = 0.0011$ ** Median PSSa before: 2 – PSSa after: 0

The majority of treatment included gastro-intestinal decontamination and supportive treatment. In 17 cases there was a complete recovery, in 3 there was a lack of improvement, while in 3 cases there was a worsening of the clinical status.

Sequences in patients having not received oxygen but received antidotes (Group2)

The medical literature reported 91 cases in which the patient received one or more antidote(s) but did not receive normobaric oxygen. Nineteen of these received 2 sequential treatments, 6 received 3 treatments, 2 received 4 and 2 received 5. Hence, 91 cases were assessed in sequence 1, 19 in sequence 2, 6 in sequence 3, 2 in sequence 4, and 2 in sequence 5.

In sequence 1, 91 patients were studied. The median PSSa before treatment was 2. They completely recovered as evidenced by a PSSa following treatment of 0. The improvement was highly statistically significant ($p < 0.0001$).

> i.e. Sequence 1 n = 91 $p = < 0.0001$ ***   Median PSSa before: 2 – PSSa after: 0

There was a complete recovery in 67 of these cases; a partial recovery in 4 cases; there was a lack of improvement in 14 cases, while there was a worsening of the clinical status in 6 cases.

The specific antidotes used are addressed in the respective antidote chapters.

This can be represented as follows:
    One antidote
    STS   = 23 $p = 0.0018$** Median PSSa before 2 - PSSa after 0
    AN   = 14 $p = 0.027$*   Median PSSa before 2.5 - PSSa after 1

Two antidotes
STS + DMAP   = 11   p = 0.0126*   Median PSSa before: 1 - PSSa after: 0
STS + SN   = 10   p = 0.0341*   Median PSSa before: 2 - PSSa after: 0

Three antidotes
AN + STS + SN   = 15   p = 0.0009***   Median PSSa before: 2 - PSSa after: 0

In sequence 2, 19 patients were studied. The median PSSa before treatment was 2. They completely recovered as evidenced by a PSSa after treatment of 0. The improvement was statistically significant (p = 0.0229).

i.e. Sequence 2 n = 19 p = 0.0229* Median PSSa before: 2 – PSSa after: 0

There was a complete recovery in 11 cases; partial recovery in 5; there was a lack improvement in 2 cases, while there was a worsening of the clinical status in 1 case.

Sequences with 2 antidotes
STS + SN   = 8   p = 0.053 ns   Median PSSa before: 2   - PSSa after: 0

Sequences in patients having received oxygen without antidote (group 3)

The medical literature reported 32 cases in which the patient received oxygen without any other antidotes, all in sequence 1. Sequence 2 was reported in 5 cases, and sequence 3 in 2 cases. Therefore, no statistical analysis was made on sequences 2 and 3. In the 32 patients in sequence 1 the median PSSa before treatment was 3. They partially recovered as evidenced by a PSSa after treatment of 1.5. The improvement was not statistically significant (p = 0.13). In sequence 1, there was a complete recovery in 13 cases; there was a lack improvement in 3 cases, while there was a worsening of the clinical status in 12 cases.

i.e. Sequence 1   n = 32   p = 0.13   Median PSSa before: 3 – PSSa After: 1.5

Sequences in patients having received oxygen and antidotes (group 4)

The medical literature reported 82 cases in which the patient received normobaric oxygen with any other antidotes. 82 cases were assessed in sequence 1. In sequence 2, 17 cases were assessed. In sequences 3 and 4, only two patients were assessed and statistical analysis was not performed.

In sequence 1 in 82 patients the median PSSa before treatment was 3. They completely recovered as evidenced by a PSSa after treatment of 0. The improvement was highly statistically significant ($p < 0.0001$).

> i.e. Sequence 1 n = 82 $p \leq 0.0001$*** Median PSSa before: 3 – PSSa after: 0

There was a complete recovery in 54 cases; there was partial improvement in 8 cases, there was a lack improvement in 14 cases, while there was a worsening of the clinical status in 6 cases.

This can be represented as follows regarding the antidotes:

Combination with one antidote

HOCO	= 16	$p = 0.0012$**	Median PSSa before 3 - PSSa after 0
AN	= 7	$p = 0.0206$*	Median PSSa before 2 - PSSa after 0

Combination with two antidotes

STS + SN	= 15	$p = 0.0022$**	Median PSSa before 3 - PSSa after 0
STS + DMAP	= 11	$p = 0.0371$*	Median PSSa before 3 - PSSa after 2

Combination with three antidotes

STS + AN + SN	= 9	$p = 0.05$ ns	Median PSSa before 3 - PSSa after 0

In sequence 2, in the 17 patients having received normobaric oxygen with any other antidotes, the median PSSa before treatment was 3. They partially recovered as evidenced by a PSSa after treatment of 2. The improvement was statistically significant, however ($p = 0.0295$).

> i.e. Sequence 2 n = 17 $p = 0.0295$*   Median PSSa before: 3 – PSSa after: 2

There was a complete recovery in 7 cases; there was a lack improvement in 5 cases, while there was a worsening of the clinical status in 2 cases.

Sequences in patients having received hyperbaric oxygen with/without antidotes (Group 5)

Hyperbaric oxygen was reported in 6 patients.

Hyperbaric oxygen was used in 3 patients without any other antidote. None in sequence 1, 1 in sequence 2, 1 in sequence 3; and 1 in sequence 4.

Hyperbaric oxygen was used in 3 patients with any other antidote. Two in sequence 1, and 1 in sequence 2.

Owing to the limited number of patients (6 in total) statistical analysis was not done.

Hyperbaric oxygen was used in 3 patients without any other antidote. None in sequence 1, and one in each of sequences 2 to 4. It was used with any other antidote twice in sequence 1 and once in sequence 2.

Table 5: Overview of cases treated without oxygen and with antidotes

Total	Sequence	STS	AN	Co-EDTA	HOCO	DMAP	SN	STS + DMAP	STS + SN	AN + SN	DMAP + Co-EDTA	STS + Co-EDTA	AN + STS + SN	HOCO + STS + AN + Co-EDTA	AN, SN alone and with obsolete antidotes
91	S1	23	14	4	3	2	1	11	10	2	1	1	15	1	3
19	S2	6	0	0	0	3	1	0	8	1	0	0	0	0	0
6	S3	3	0	0	0	0	0	0	3	0	0	0	0	0	0
2	S4	1	0	0	0	0	0	0	1	0	0	0	0	0	0
2	S5	2	0	0	0	0	0	0	0	0	0	0	0	0	0

Table 6: Overview of cases treated with oxygen and with antidotes

Total	Sequence	STS	AN	HOCO	Co-EDTA	STS + AN	STS + HOCO	STS + DMAP	STS + SN	STS + Co-EDTA	HOCO + Co-EDTA	STS + AN + SN	AN + SN	STS + HOCO + SN + Co-EDTA	HOCO, AN, SN, STS and obsolete antidotes
82	S1	4	7	16	2	4	4	11	15	0	3	9	1	2	4
17	S2	1	2	5	1	0	2	0	2	1	0	2	0	0	1
2	S3	1	0	0	0	0	0	0	1	0	0	0	0	0	0
2	S4	0	0	0	0	0	0	0	0	0	0	1	0	0	1
0	S5	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Comparing the successes of antidote therapy with and without oxygen application, no appreciable differences were seen:

Table 7: Recovery from cyanide poisoning with and without treatment with oxygen

	Antidotes without oxygen - %	Antidotes with oxygen - %
Fully effective	73	66
Partially effective	4	10
Lack of improvement	16	17
Worsening	7	7

4.1.9 Gaps in knowledge

The mechanisms of action of oxygen, either normobaric or hyperbaric, in cyanide poisonings remains to be fully clarified.

High flow of oxygen administered by mask and 100% of oxygen in endotracheally, intubated cyanide poisonings should be considered. However, the duration of oxygen therapy and the best dosage regimen to use in the various forms of cyanide poisoning remains to be determined.

The efficiency of hyperbaric oxygen in cyanide poisoning remains an open question; the rarity of cases reported in the medical literature precludes any conclusion.

4.1.10 Conclusion

Experimental data showing efficacy of normobaric oxygen treatment in overt cyanide poisoning is limited and controversial.

The mechanism of action of oxygen is quite complex and poorly understood. But displacement of oxygen by cyanide from the reduced ferric ion of the mitochondrial cytochrome aa_3 appears credible. Efficacy of oxygen as a treatment for cyanide might be limited by new insight on the pathophysiology of cyanide poisoning suggesting it is not limited to inhibition of cytochrome oxidase. Experimental data regarding oxygen transfer in the blood have indicated two phases in cyanide poisoning: (a) An early phase without oxygen depletion in the blood where oxygen would be a considered as a true antidote; (b) A late phase with oxygen depletion in the blood where the supportive effect of oxygen is mixed with its antidotal effect.

The available data clearly show that cyanide poisonings can recover without oxygen, and antidotes administered without oxygen are highly efficient. This is of major importance when

considering the concern of cyanide-induced mass disaster where oxygen availability would not be adequate to treat all symptomatic victims. However, the preceding analysis marginally supports the conclusion that the best treatment regimen is based on the combination of oxygen and supportive treatment with additional antidotes to cyanide.

In case of cyanide poiosning, normobaric oxygen should be administered as high flow by mask or at 100% after endotracheal intubation. Owing to the frequency of bradypnoea and apnoea, mechanical ventilation should be considered as a mode of administration of oxygen. The duration of oxygen therapy has not been determined. However, the required duration is limited to hours or days at max. The short duration of oxygen administration precludes any concern regarding oxygen toxicity as a treatment of cyanide poisoning.

4.2 Sodium thiosulphate

4.2.1 Introduction

Chemically defined as sodium thiosulphate pentahydrate, sodium thiosulphate (as it is colloquially named) is abbreviated as STS forthwith. The anhydrous form is also used (Section 4.2.3). The use of STS as an antidote in cyanide poisoning has been established for decades and was first described by Lang in 1895.

STS is an antidote to cyanide that works by supporting the body's natural mechanism for detoxification of cyanide by transforming the toxic cyanide ion into the far less toxic thiocyanate, which is further eliminated by the kidneys. STS does not act by itself but is the substrate of a ubiquitous enzyme thiosulphate sulphurtransferase, also entitled Lang's Rhodanese[a]. The efficacy of STS is well documented. However, the number of case reports or clinical trials describing the use of STS alone is limited because it is predominantly used in combination with other antidotes.

STS has been thoroughly discussed in the IPCS/CEC Evaluation of Antidotes Series, from which most of the background information has been taken (IPCS, 1993).

4.2.2 Identity

IUPAC name:	Sodium thiosulphate, pentahydrate (or anhydrous)
CAS Registry number:	10102-17-7 (pentahydrate), 7772-98-7 (anhydrous);
Formula:	$Na_2S_2O_3.5H_2O$ (pentahydrate), $Na_2S_2O_3$ (anhydrous)
Molecular weight:	248.2 (pentahydrate), 158.1 (anhydrous)
Chemical structure:	
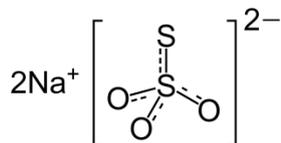
* 5 H_2O (Wikimedia Commons, 2010)

STS is a transparent, colourless crystalline, odourless compound. It is efflorescent in dry air and highly soluble in water.

4.2.3 Practicality

STS is stored at room temperature between 15 and 25°C (Köhler Chemie, 2013), packaged in air-tight containers (Ph Eur 6.0; USP 31). The shelf life has been cited as 2 years (Köhler Chemie, 2013), while other indicate 3 years (Kirk-Othmer, 1969; Windholz, 1983; Martindale, 1989, National Corporation of Swedish Pharmacies, in IPCS, 2009) in intact ampouls.

STS is incompatible with iodine, acids, lead, mercury, silver salts, salts of heavy metals, and oxidizing agents. STS mixed with hydroxocobalamin results in the production of thiocobalamins (Evans 1964). The compatibility of STS with another cobalt donor, dicobalt edetate, has not been addressed.

STS should be administered by parenteral route. The recommended route is the intravenous. In patients with a poor hemodynamic status, intra-osseous administration should be also considered. The modes of administration of STS recommended presently require medical supervision.

4.2.4 Pharmacokinetics and metabolism

Pharmacokinetics in non-poisoned animals/healthy volunteers

Absorption

Following (recommended) intravenous administration of STS, the bioavailability is 100%. The absorption by the oral route is reported to be low (Martindale, 1989), but has not been quantified.

Distribution

Experiments in man show, as observed in animals, that after intravenous injection, STS is rapidly distributed in the extracellular space (Bucht, 1847; Foulks et al, 1952; Gilman et al, 1946).

The volume of distribution of STS was examined in 6 healthy male humans (ages 18 – 30 years) after infusion of 12g/100ml in sterile water over 10-12 minutes. The average was 12.2 litres or 16.6% of body weight. The apparent V_d was found to be 150 ml/kg (Cardozo and Edelman, 1952).

After injection of 150 mg/kgbw in 5 male humans the plasma concentration after 5 minutes was 1012 mg/l. The half-life of the distribution phase was 23 minutes (Ivankovich et al, 1983).

Metabolism

Rhodanese is an intra-mitochondrial enzyme. It is assumed that STS permeates slowly through cell and mitochondrial membranes (Himwich and Saunders, 1948; Cardozo and Edelman, 1952; Sörbo, 1962). The dicarboxylate carrier is involved in the penetration across the mitochondrial membrane (Crompton et al, 1974). Since normal endogenous production of thiosulphate is relatively small (see next section; Ivankovich et al, 1983) a therapeutic dose of 150 mg/kgbw would elevate plasma concentrations about 100 fold. Such high concentrations are required to increase the intracellular concentration and enable rhodanese to detoxify cyanide at the mitochondrial membrane, which is the site of action of thiosulphate (IPCS, 1986).

Elimination

In a volunteer study in healthy male humans a physiological plasma concentration of STS of 11.3 mg/l and in urine of 2.8 mg/l were recorded without any exogenous non-dietary supply of STS. Excretion amounted to circa 3 mg per day (Ivankovich et al, 1983). The elimination half-life was found to be approximately 80 minutes using a one-compartment kinetic model (Shea et al, 1984). After injection of 150 mg/kg bw in 5 male humans the plasma concentrations after 5 minutes was

1012 mg/l, the elimination half-life was 182 min. Urine concentration and excretion rate of STS increased significantly after injection. 180 min post-injection, the excretion was 42.6% of the administered dose (Ivankovich et al, 1983).

Studies in man and animals showed that STS is both secreted and reabsorbed in the kidney (Bucht, 1949; Foulks et al, 1952). The clearance of STS is low, but at high levels of STS, secretion Tm (transfer maximum) is similar to reabsorption Tm, whereas at low plasma levels both filtered and secreted thiosulphate are reabsorbed resulting in a diminished clearance value for thiosulphate.

Most of an administered dose will be eliminated unchanged, while the rest is oxidised to sulphate in the liver (Himwich and Saunders, 1948; Sörbo, 1972).

Metabolism and elimination in poisoned patients

The serum half-life for STS was found to be 15 minutes in man during SNP therapy (Schulz et al, 1982; Schulz, 1984) and about 40 minutes in children aged 4 months to 14 years (Gladke, 1966).

Alteration of the kinetics of cyanide induced by the antidote

The therapeutic efficiency of STS in acute cyanide poisoning was studied in rats (Renard et al, 2005). STS (225 mg/kg i.p.) administered 10 minutes post-cyanide injection i.e. at the time at which peak blood cyanide concentration was measured, and resulted in a significant decrease in peak (0.48 + 0.04 vs 1.67 + 0.29 mg/l) as well as approximately halfing the AUC of the blood cyanide concentrations (45.8 + 1.29 vs 82.2 + 12.25 mg/min/l).

Plasma, red cell cyanide, and plasma sodium thiocyanate concentrations were measured in 30 patients undergoing elective SNP-induced hypotension (Cole and Vesey, 1987). One randomly selected group (n = 15), who received 0.21-0.70 mg/kg SNP over periods of 50-160 minutes was given a bolus of STS (10.6-38.5 mg/kg) immediately on cessation of the SNP administration. The other group (n = 15), who received infusions of 0.11- 0.85 mg/kg SNP for periods of 59-197 minutes, received no antidote. Cyanide concentrations, expressed as a percentage of the immediate post-infusion values, were significantly lower in the treated group in all subsequent blood samples (at 10, 30, and 60 minutes; plasma cyanide $P < 0.05$; red cell cyanide $P < 0.001$). Improved cyanide metabolism was further demonstrated by sharp increase in mean plasma thiocyanate concentration ($P < 0.05$) in the group receiving the antidote.

In 1982 cyanide toxicity resulting from therapeutic use of SNP was studied with and without STS treatment. Fifty one patients received SNP for short periods. A further 19 patients received SNP over longer periods of up to two weeks. In seven of these 19 patients, 1 g STS was given i.v. as a

bolus injection during SNP treatment. STS given by infusion stopped the accumulation of cyanide, elevated cyanide levels declined, and SCN levels increased. The simultaneous infusion of STS in SNP therapy prevented accumulation of cyanide. (Schulz et al, 1982)

4.2.5 Mode of action, dosage and time of onset

STS is given as a specific antidote to augment the systemic clearance of cyanide via sulphurtransferases mediated thiocyanate formation. Generally thiosulphate is present in the body but only in small quantities, being derived mainly from the metabolism of cysteine and other mercapto compounds. Iit is the main source of sulphur for rhodanese, an enzyme that converts trace amounts of cyanide arising during normal metabolism into thiocyanate. The physiological reserves of STS available for detoxifying cyanide are therefore limited and depend on nutritional status (Schulz et al, 1979; Krapez et al, 1981, Schulz et al 1982).

Inter-individual variation in serum rhodanese activity can vary by a factor of 6 (Nawata et al, 1991) or 3 to 8 (Narendranathan et al, 1989), but rhodanese is present in all body tissues in considerable excess (Schulz et al, 1982), and the highest concentrations are found in the liver and also in muscles. Within the cells, rhodanese is located mainly in the matrix of mitochondria (Westley et al, 1983). In a pharmacokinetic study on cyanide distribution and metabolism with and without STS conducted in mongrel dogs, STS produced a greater than 30-fold increase of the trans-sulphuration reaction (Sylvester et al, 1983). The liver metabolises nearly all cyanide at subtoxic dose levels via the rhodanese. After oral cyanide exposure a first pass effect has to be taken into account. In the absence of sulphur donating agents in the human body, the maximum detoxification rate was claimed to be as low as 0.9 µg CN^-/kgbw/min (Schulz et al, 1982). However, a re-analysis of the data suggest a rate of 3.0 µg CN^-/kgbw/min (80 min mean infusion duration) based on the dose rate at which no clinical symptoms occurred (ECETOC, 2007).

Thiocyanate, metabolite of the detoxification of cyanides

Cyanide is detoxified in the body through the formation of thiocyanate which is subsequently eliminated in the urine (Goldfrank, 1994). In healthy humans receiving single doses of 1.2 to 1.5 g Sodium thiocyanate, 96 to 99% of the dose was excreted in urine within 5 to 14 days; 3 to 7 mg/d were excreted in faeces. After prolonged exposure to thiocyanate urinary excretion was increased and serum levels decreased (Weuffen, 1982). The elimination half-life was prolonged in patients with renal failure (Schulz et al, 1979). Human experience suggests that serum levels of 20 to 40 µg SCN^-/ml would not lead to adverse effects in healthy humans (Barker, 1936; Barker et al, 1941), but it should be taken into account that thiocyanate is a competitive inhibitor of iodine uptake by the thyroid (Barrere et al, 2000). In humans, the apparent volume of

distribution for thiocyanate was 0.25 l/kgbw and the elimination half-life was 2.7 days in healthy individuals and up to 9 days in subjects with impaired renal function (Schulz, 1984).

Thiocyanate concentrations are normally between 1-4 mg/l in the plasma of non-smokers and 3-12 mg/l in smokers. The plasma half-life of thiocyanate in patients with normal renal function is 4 h (Blaschle and Melmon, 1980) but in those with renal insufficiency it is markedly prolonged and these patients are therefore an increased risk of toxicity (Schulz et al, 1978). Thiocyanate levels exceeding 100 mg/l are thought to be associated with toxicity characterised by weakness, muscle spam, nausea, disorientation, psychosis, and hyper-reflexia; and lethality at concentration greater than 180 mg/l (IPCS, 2009). Haemodialysis is recommended as an effective means of removing thiocyanate (Pahl and Vaziri, 1982, Nessim and Richardson 2006).

Since the renal clearance of thiosulphate and the metabolite sulphate is at least ten times higher, and their toxicity at least ten times lower, than for the metabolite thiocyanate (see 8, 15, 23, 37 in Schulz, 1982) thiosulphate toxicity does not need to be monitored when given theraputically.

Stoichiometry – implications for dosage

One mole of STS (MW: 158.1) is expected to bind one mole of the cyanide ion (MW: 26).

In experiments in animals and humans it could be shown that there is a positive correlation between the available amount of STS and the enhancement of the detoxification of cyanide. The recommended initial dose for adults is 8 to 12.5 g or 0.1 to 0.2 g thiosulphate/kg body weight i.v. (Chen, 1944; Chen, 1952; Baskin, 1992; Goldfrank, 1994).

A dose of approximately 410 mg/kg body weight is suggested for children with normal Hb (Berlin, 1970) and a paediatric dose of 1.65 ml/kgbw is reported in the ECETOC JACC.

Onset of the effect

STS is able to shorten the time of recovery remarkably after sublethal or lethal doses of cyanide to 4-10 minutes in animal experiments (Schubert and Brill, 1968; Friedberg, 1968; Schwarzkopf, 1971).

In acute SNP intoxication, the simultaneous bolus injection of STS (SNP/antidote molar ratio of 1:5) was effective in reducing the early signs and severity of the metabolic acidosis in rabbits and is converted to thiocyanate as quickly as it is released from the SNP when STS is given simultaneously (Pill et al, 1980).

STS acts more rapidly than had been considered previously (Krapez et al, 1981; Sylvester, 1983). Complete cyanide detoxification was achieved with prophylactic thiosulphate alone (Krapez et al, 1981) and then given both prophylactically and post-exposure in dogs (Ivankovich et al, 1980)).

4.2.6 Safety

Toxicology of STS:

The oral toxicity in man is alsovery low and single doses of 15 – 18 g have only laxative effects. Nausea, headache and vomiting have also been reported (Sörbo, 1972; Poisindex, 1987), especially in case of rapid i.v. injection (Ivankovich, 1983). Hypotension due to the formation of thiocyanate may also occur (Done, 1961; Sylvester, 1983).

A dose of 2 g STS/m^2 (body surface area) per hour given over 12 hours was without side-effects (Howell et al, 1982). The authors found that 12 g/m^2 can be given safely to humans over 6 hours provided that cardiac and renal functions are normal.

No data are available about mutagenicity, carcinogenicity and reproduction. Also there is no classification and labelling for STS required by GHS.

Contraindications

There are no specific contraindications. STS should not be administered in cases of known sodium metabisulphite hypersensitivity due to the content of sodium metabisulphite (0.01 g) (Essential Medicines 17th ed. WHO Model List, 2011).

The toxicity of STS is low and toxic effects should not be expected unless doses far exceed those recommended.

In patients with renal insufficiency, dialysis can be considered for the more rapid elimination of thiocyanate (NIOSH, Pahl and Vaziri, 1982, Nessim and Richardson 2006).

No other special risk groups could be identified.

Adverse effects related to antidotes in cyanide-poisoned humans

The safety of 5 g of HOCO given i.v., alone or in combination with 12.5 g of STS, was evaluated in healthy adult men who were heavy smokers. STS administered alone caused nausea, vomiting, and localised burning, muscle cramping, or twitching at the infusion site (Forsyth and Hall, 1993).

The excipient content of maximum 6.7% (as SO_2) sodium metabisulphite in the pharmaceutical preparation can in very rare cases cause allergic reactions (Information Dr. Köhler Chemie, as producer).

4.2.7 Case series and casuistics of cyanide / cyanogenic poisonings treated with STS

An overview of cases in which STS was used and included in the PSSa study is presented in Table 8. This is classified into STS alone and in combinations with other antidotes in 4.2.8.1 to 4.2.8.3, if the number of cases is sufficiently high for statistical analysis.

The individual cases are described in Appendix C and the details were compiled into the sequential PSSa database (Appendix B) (Section 3.4.4). Relevant details are summarised in Table 9.

*Table 8: Overview of STS use alone and with other antidote(s) **

	Total	Sequence	STS	AN	SN	HOCO	Co-EDTA	DMAP	AN + obsolete antidote	BAL	Coramine + AN	AN + SN	AN + SN + Co-EDTA	HOCO + SN + Co-EDTA	HOCO + SN	HOCO + Co-EDTA + AN	HOCO + BAL	SN + DMAP	SN + outmoded antidote
STS	119	S1	**29**	5	**25**	4	1	**23**	1	0	1	**25**	1	2	0	2	0	0	0
	44	S2	**14**	1	**15**	3	1	1	0	0	0	5	0	0	1	0	1	1	1
	21	S3	**7**	0	**12**	1	0	0	0	0	0	0	0	1	0	0	0	0	0
	7	S4	3	0	3	0	0	0	0	0	0	1	0	0	0	0	0	0	0
	2	S5	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

* Data in bold are used for analysis (n ≥ 7)

Table 9: Cases treated with STS alone

Case			Antidote treatment				Reference
			Cumulative dose of antidote (g)	**Number of treatment steps**	**PSSa**		
Age	**Sex**	**Type of poisoning**			**Before**	**After**	
28	M	Acetonitrile	NS	1	3	0	Amdur, 1955
28	M	Acetonitrile	NS	1	2	0	Amdur, 1955
NS	M	Adipic nitrile	3.75	1	2	0	Ghiringhelli, 1955
14	M	SNP	> 5.4 [a]	1	3	0	Perschau, 1977
2.5	M	Cassava	12.5	1	1	0	Cheok, 1978
1.5	F	Cassava	12.5	1	3	0	Cheok, 1978
27	F	KCN	NS	1	3	4	Favarel, 1982
58	M	SNP	> 0.1	2	2	0	Schulz and Roth, 1982
NS	NS	HCN	NS	2	3	0	Bonsall, 1984
72	F	SNP	18.75	1	3	0	Patel, 1986
71	F	SNP	0.1 or 0.2	1	2	0	Patel, 1986
65	M	SNP	0.1 or 0.2	1	2	3	Patel, 1986
61	M	SNP	0.1 or 0.2	1	2	2	Patel, 1986
71	M	SNP	0.1 or 0.2	1	2	0	Patel, 1986
58	M	SNP	0.1 or 0.2	1	2	0	Patel, 1986
NS	M	SNP	0.1 or 0.2	1	3	4	Patel, 1986
31	M	Acetonitrile	8.75	1	2	0	Geller, 1991
25	M	KCN	7.8 [b]	1	3	0	Lundquist, 1992
51	M	Laetrile	25	1	0	0	Yeh, 1992
32	M	Acrylonitrile	1	1	0	0	Steffens, 1998
28	M	Acrylonitrile	2	1	2	0	Steffens, 1998
52	M	Acetonitrile	1	1	0	0	Steffens, 1998
52	M	Propionitrile	2	1	2	0	Steffens, 1998
29	M	Acetone cyanohydrin	10	1	0	0	Roedelsperger, 2009
NS	M	Acetone cyanohydrin	2	1	0	0	Roedelsperger, 2009
27	M	Acetone cyanohydrin	10	1	0	0	Roedelsperger, 2009
52	M	Acetone cyanohydrin	10	1	0	0	Roedelsperger, 2009
53	M	Acetone cyanohydrin	10	1	0	0	Roedelsperger, 2009
NS	M	Methacrylonitrile	1	1	2	0	Roedelsperger, 2009
38	M	Methacrylonitrile	1	1	2	0	Roedelsperger, 2009
56	M	Acrylonitrile	6	2	1	0	Steffens, 2002
18	M	Acrylonitrile	3	2	1	0	Steffens, 2002
83	M	KCN	NS	1	3	4	Mutlu, 2002

Case studies are detailed Appendix C
[a] Reported as 150 mg + 75 mg/kgbw every 3 h
[bc] Reported as 120 mmol
NS, not stated

Table 10: Cases treated with STS and other antidotes

Case			Antidote treatment									Reference
			Cumulative dose of antidote (g)						Number of treatment steps	PSSa		
Age	Sex	Type of poisoning	AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO		Before	After	
With Co-EDTA												
22	F	KCN			20		0.6		1	3	0	Hoang The Dan, 1981
28	M	CN salt			24 [a]		0.3		1	3	0	Lundquist, 1992
32	M	KCN			8		4		1	ND	4	Jourdan, 1993
STS, nitrites and DMAP												
21	M	NaCN	-	0.3	20	> 0.25			2	3	4	Van Dijk, van Heijst, 1987
STS, nitrites and Co-EDTA												
48	M	NS	2	0.3	12.5		0.3		2	2	0	Davis and Ewer, 1988
STS, nitrites and HOCO												
23	M	KCN	-	0.5	75			0.03	1	3	4	Buchanan, 1976
STS, DMAP and Co-EDTA												
26	F	KCN			6	0.25	0.3		2	2	0	Daunderer, 1974
STS, Co-EDTA and HOCO												
23	M	KCN	-	-	NS		0.3	4	1	3	0	Lutier, 1971
NS	M	KCN			NS		0.3	4	2	2	0	Lutier, 1971
26	M	Acetonitrile			NS		NS	NS	1	3	0	Jaeger, 1992
30	F	CN salt			16		NS	15	2	3	4	Baud, 2001
STS, nitrites, Co-EDTA and HOCO												
NS	M	KCN	-	NS	NS		0.6	4	1	2	0	Lutier, 1971
NS	NS	KCN	NS	-	NS		0.6	4	1	2	0	Lutier, 1971

Case studies are detailed Appendix C
[a] Reported as 98 mmol
NS, not stated; -, not applied

4.2.8   Assessment of efficacy and effectiveness in clinical use

- STS was used either alone, or in combination with other cyanide antidotes, in 191 sequences allowing determination of a PSSa before and after its administration.
- STS was shown to be efficient when used alone in 50 sequences resulting in full recovery of moderate poisoning, independent of the sequence and even late in the course of treatment. There is no clinical experience of STS alone in severe cyanide poisoning.
- STS was fully efficient when combined with SNin severe cyanide poisonings resulting in full recovery, both early and late in the course of treatment. It was fully efficient in moderate poisonings if given with AN/SN or DMAP. Available PSSa data do not allow for statistical analyses of the combinations with Co-EDTA or HOCO.

Assessment of efficacy using sequential PSSa

STS alone

| Sequence 1 | STS = 23 | $p = 0.0018**$ | Median PSSa before 2 - PSSa after 0 |
| Sequence 2 | STS + SN = 8 | $p = 0.053$ ns | Median PSSa before 2 - PSSa after 0 |

STS alone with oxygen and/or supportive treatment

Sequence 1	$n = 29$	$p = 0.0002$ ***	Median PSSa before: 2 - PSSa after: 0
Sequence 2	$n = 14$	$p = 0.08$ ns	Median PSSa before: 2 - PSSa after: 0
Sequence 3	$n = 7$	$p = 0.13$ ns	Median PSSa before: 2 - PSSa after: 1

STS in combination with any other antidote with oxygen and/or supportive treatment

Sequence 1	$n = 119$	$p = < 0.0001$ ***	Median PSSa before: 2 - PSSa after: 0
Sequence 2	$n = 44$	$p = 0.0003$ ***	Median PSSa before: 3 - PSSa after: 0
Sequence 3	$n = 21$	$p = 0.0022$ **	Median PSSa before: 2 - PSSa after: 1
Sequence 4	$n = 7$	$p = 0.3387$ ns	Median PSSa before: 2 - PSSa after: 0

STS and SN

Sequence 1	$n = 25$	$p = 0.0002$ ***	Median PSSa before: 3 - PSSa after: 0
Sequence 2	$n = 15$	$p = 0.0634$ ns	Median PSSa before: 3 - PSSa after: 0
Sequence 3	$n = 12$	$p = 0.0097$ **	Median PSSa before: 3 - PSSa after: 0

STS and AN and SN

| Sequence 1 | $n = 25$ | $p = 0.0003$ *** | Median PSSa before: 2 - PSSa after: 0 |

STS and 4-DMAP

Sequence 1   n = 23   p = 0.0009 ***   Median PSSa before: 2 - PSSa after: 0

STS and Co-EDTA - No statistical analysis.

STS and HOCO - No statistical analysis.

Data from the use if STS either alone, or in combination with other antidotes to cyanide, in 191 sequences allowing determination of PSSa before and after its administration and support the following assumption:

- STS was efficient when used alone in 50 sequences. STS alone was completely efficient and resulted in full recovery of moderate poisoning. It has to be recognised, however, that there is no clinical experience of STS alone in severe cyanide poisoning. The effectiveness of STS reported in the first four sequences suggested that it is efficient when used alone even if administered late during the course of the poisoning.
- STS was shown to be efficient when combined with other antidotes (sequences 1 to 4), in severe cyanide poisoning. In these cases, both early and late recovery was full. The other antidotes that were shown to be efficient when combined with STS included SN (severe cyanide poisonings), AN combined with SN (moderate poisoning) and DMAP (moderate poisoning). The available PSSa data for combinations of STS with Co-EDTA or HOCO are insufficient for statistical analyses.

Global assessment of efficacy and effectiveness of STS alone and in combination

Efficacy

STS alone is capable of providing complete protection against both cyanide (KCN) and cyanide-forming compounds especially when administered simultaneously with these compounds as a continuous infusion. When high plasma concentrations of STS are present, the detoxification mechanism is fast enough to provide adequate protection (Ivankovich, 1980).

The combinations of AN, SN and DMAP with STS have also been found to be fully effective, even in severe poisonings.

Gaps in knowledge

The effectiveness of STS would strongly depend on the activity of the rhodanese. While in 1980 monomorphism was described in all populations tested except for 2 Indian populations (Scott and Wright, 1980), a genetic polymorphism of rhodanese has been seen recently

(Billaut-Laden et al, 2006) but its clinical implications, if any, are not known. Inducers or inhibitors of rhodanese activity could not be identified.

Enzymatic detoxification means that a saturation of the detoxification process may occur even without any limitation of STS. The liver metabolises nearly all cyanide at subtoxic dose levels via the rhodanese pathway. A re-analysis of human data suggests a rate of 3.0 µg CN$^-$/kgbw/min (80 minutes mean infusion duration) based on the dose rate at which no clinical symptoms occurred (ECETOC, 2007). More information about the actual effectiveness should be provided. Recent data suggest that the onset of the antidotal effectiveness of STS occurs sooner than previously reported. More data should be gathered to confirm this.

4.2.9 Conclusion

STS is an antidote to cyanide whose principal mechanism of action is known. It acts by modifying the toxicokinetics of cyanide by increasing its elimination by an established enzymatic pathway.

The pharmacokinetics of STS in humans is known for a dose up to 150 mg/kg, including healthy volunteers as well as cyanide poisoned humans while the pharmacokinetics for greater doses is unknown. However, higher doses have been applied without problems. The delay in onset of the antidotal effectiveness is still a pending question, but, according to recent data it seems to be shorter than previously supposed.

STS is safe at the doses used for antidotal treatment of cyanide poisonings. Only mild and infrequent adverse effects have been reported, both in volunteers and poisoned subjects. The presence of sodium metabisulphite in the pharmaceutical preparation can result in very rare cases of allergic reactions.

Although there is no clinical experience of STS alone in severe cyanide poisoning STS alone was completely efficient and resulted in full recovery of moderate poisoning. STS was efficient when used alone even administered late during the course of the poisoning.

STS was fully effective when combined with SN (severe cyanide poisonings), AN plus SN (moderate cyanide poisonings), and DMAP (moderate cyanide poisonings).

The recommended initial dose is 8 to 12.5 g in adults. A paediatric dose recommendation does exist of about 410 mg/kg body weight.

Regarding practicality, STS is relatively inexpensive and stable. Need for i.v. administration limits somewhat its application in the industrial and other non-clinical settings.

4.3 Nitrites

4.3.1 Introduction

Sodium nitrite (SN) has been in use as a cyanide antidote since at least the 1950s. Commercially developed in the U.S. by Eli Lilly and Company, the drug became part of a kit widely known as the 'Lilly Kit' containing AN pearls together with SN (10 ml containing 300 mg) and STS (25 ml containing 25g), both for injection. A similar kit remains for sale in the US known as the 'Taylor Kit' or 'Pasadena Kit'. It continues to be used in other countries as well, including much of Central and South America. Last producer was Akorn Inc., but the current Akorn kits expired in September 2012 and there were no plans for resuming production. This kit is being replaced by a kit called 'Nithiodote', which contains SN and STS, but no AN. It was approved by the FDA in 2011 (the Lilly Kit has never been an approved drug!). Other comparable kits which include AN are still available, such as the Cyanide Antidote kit marketed by ABO Pharmaceuticals and the Cyan SOS™ kit marketed by Troikaa Pharmaceuticals Ltd.

For many years the presumed mechanism of action of SN was the production of MetHb, which has a higher binding affinity for cyanide than does cytochrome oxidase. SN has been reported to induce approximately 20% methaemoglobinaemia when given as a dose of 300 mg in an adult (Chen and Rose, 1952). In recent years, the formation of MetHb as a sole or predominant mechanism of action has been questioned. Way (1984) suggested that the antidotal mechanism of action of nitrites might be vasodilatation. This seems to be even more convincing, as at the beginning and in the middle of the last century SN was developed for angina patients dilating the small cardiac vessels and leading to a fall in the blood pressure. New evidence suggests that the formation of nitric oxide by SN may play a direct critical role in protecting cytochrome oxidase against the toxic effects of cyanide independent of the vasodilatation it induces (Pearce, 2003). This mechanism will be discussed further below.

4.3.2 Identity

Sodium nitrite

IUPAC name:	Sodium nitrite
CAS Registry number:	7632-00-0
EC (EINECS) number:	231-555-9
Formula:	$NaNO_2$
Molecular weight:	69.00
Chemical structure:	

(Wikimedia Commons, 2007)
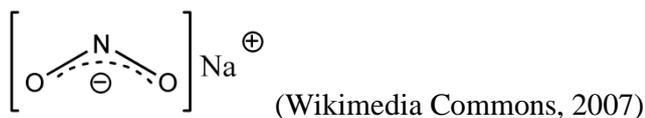
Amyl nitrite

IUPAC name:	3-Methylbutyl nitrite, 2-methylbutyl nitrite
CAS Registry number:	8017-89-8 (mixture with 2-methylbutyl nitrite)
	110-46-3 (3-methylbutyl nitrite)
EC (EINECS) number:	203-770-8 (mixed isomers)
Formula:	$C_5H_{11}NO_2$
Molecular weight:	117.15
Chemical structure:	
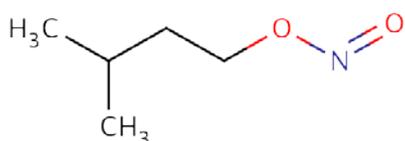
(3-methylbutyl nitrite) (DrugBank, 2012)

4.3.3 Physico-chemical and pharmaceutical properties

Sodium nitrite

Sodium nitrite forms colourless crystals or mass, or yellowish rods. It is freely soluble in water but not in alcohol. Since SN will oxidise to nitric oxides upon exposure to air (slowly) and acids (faster) it should be kept in air-tight containers (Abdata/Pharm. Stoffliste/Martindale 31).

AN

AN occurs as a clear yellowish, volatile, flammable liquid having a peculiar, ethereal, fruity odour and pungent aromatic taste. It is practically insoluble in water but it is miscible with alcohol and ether. AN is a mixture of the nitrite esters of 3-methylbutan-1-ol and 2-methylbutan-l-ol. Isoamyl nitrite is not miscible with water, but can be dissolved in alcohol, diethylether and chloroform (Abdata).

4.3.4 Practicality

SN

SN should be packaged in air-tight containers and stored at 25°C, with excursions permitted between 15° and 30°C. SN injection is preserved in single-dose containers of Type I glass. The shelf life of the drug is about 3 years if stored at 25°C (IPCS, 1993).

SN is administered i.v. (10 ml solution containing 300 mg of SN), ideally over at least 5 to 10 minutes. Monitoring of the vital parameters, especially blood pressure, heart rate and oxygenation is essential / mandatory. The administration of the antidote may be repeated in half or the same dosage (Anderson BD from Brent et al, 2005). Possible side effects are a decrease in blood pressure (that might require the use of catecholamines or similar drugs for circulatory support) and an excessive rise in the MetHb-level of the patient (Baskin and Brewer, 1997). Therefore it should only be administered by medically trained staff. If overdosed, or if a severe methaemoglobinaemia already occurs under normal dosing, Toluidine or Methylene-Blue might be administered as antidotes for SN. It kmust be kept in mind, that this will again liberate cyanide from the methaemoglobin (van Heijst et al, 1987) Fear of The potential risk of these severe side effects may keep prevent some physicians from using SN (Lam and Lau, 2000).

AN

AN is packaged in air-tight containers and must be stored in a cool place at 10°C, protected from light (Abdata/Martindale 28). AN is highly flammable and forms an explosive mixture in air or oxygen at room temperature. Caution is therefore advised when handling AN and sources of ignition should be avoided.

AN inhalant

Within the Taylor-Antidote-Kit one ampoule contains 0.3 ml of the antidote (JACC 53/Vol.1).

Stability: The drug shelf live is up to 2 years below 15°C (JACC 53/Vol. 1) or at the lowest possible temperature allowing for immediate access, while refrigeration is not required (Beasley et al, 1978).

A stabiliser such as diphenylamine or epoxolol is added to commercially available products. The stability of amyl nitrite ampoules might be potentially increased by much as tenfold over that of present commercial products by the addition of chemical agents that act as sinks for nitrogen dioxide and/or nitric acid produced during chemical breakdown. Possible agents include potassium carbonate (solid), trisodium phosphate (solid), magnesium oxide (solid), pyridine (dissolved), and diphenylamine (dissolved) (Yunker and Higuchi, 1958), AN is used as an inhalant over the mouth and nose of the patient. This makes it the perfect antidote for being used by paramedics or first responders. The ampoules can be broken over the tubus in the intubated and ventilated patient as well. One ampoule should be broken and the patient should breathe it for about 15 seconds, then an interval of about the same time should be made. This procedure can be repeated several times (5-6 times per ampoule and 1-4 ampoules might be administered,

that means up to 20 turns of inhalation, without causing severe methaemoglobinaemia i.e. less than 5% (ECETOC 2007).

4.3.5 Pharmacokinetics and metabolism

Pharmacokinetics of nitrite in animals

Little is known aboit the ADME of AN in animals other than nitrites are mostly excreted via by the kidneys (Poisindex).

Pharmacokinetics of nitrite in humans

Absorption

SN should be administered i.v. and is thereby bioavailable within minutes. The first effect will be vasodilatation (Poisindex, Anderson BD from Brent 2005). AN should be administered by inhalation. It will be absorbed rapidly and is bioavailable even faster than SN given i.v. It causes vasodilatation with the most common side effect being temporary headache (Anderson BD from Brent, 2005).

Distribution

SN as well as AN is distributed to all organs rapidly by the circulation. The first effects on MetHb formation in erythrocytes occur within a few minutes (Anderson BD from Brent, 2005). One 300 mg ampoule of SN results in a MetHb-level of 10 to 20% which means it does not cause severe hypoxia (Anderson BD from Brent, 2005, JACC 53/Vol. 1).

De Beer et al measured nitrite levels in a fatal case of oral SN intoxication (cited by PoisIndex). The MetHb level was 49% and the nitrite levels were 0.5 mg/l in the blood, 0.0 mg/kg in the liver, 0.3 mg/kg in the kidneys, 8.7 mg/l in the urine and 3.9 mg/l in the stomach. This shows that nitrites remain at the place of first contact for some time and a significant portion is excreted unchanged in the urine. In the body the greatest amount of the drug can be found in the blood compartment, followed by organs with high arterial blood flow. It is hardly detectable in the liver. This is consistent with the most common clinical symptoms (MetHb formation and vasodilatation).

Metabolism

Nitrites could be oxidised to nitrate within about one hour in the body and subsequently excreted via the urine (Anderson BD from Brent, 2005). In contrast, Sweetman found out that 60% of the nitrite was metabolised to ammonia. (Poisindex, citing Sweetman, 2000). Blood or serum nitrite

and nitrate levels are usually not monitored during the treatment of cyanide poisoning since MetHb formation develops more or less immediately after nitrite administration, remains high for longer than nitrite and can be controlled more easily (Poisindex). As nitrite metabolism shows considerable differences between individuals, Goldfrank's does not recommend using a target-MetHb-level for controlling the effectiveness of nitrite therapy (Goldfrank, 1984).

Elimination

Nitrites, as well as their metabolite nitrate, are excreted predominently via the urinary tract. Between 30 and 40% of the drug will be excreted unchanged (Baselt, 2000; Sweetman, 2000; cited by Poisindex).

4.3.6 Mode of action, dosage and time of onset

Major mode of action: Methaemoglobin formation

The first proponents of the cyanide antidote kit proposed that SN works via the formation of MetHb, which can then bind with cyanide to form cyanomethaemoglobin. While cyanomethaemoglobin, like MetHb, is unable to carry oxygen it shields the cytochromes from unbound cyanide. Cyanide is released slowly form methaemoglobin thereby giving the organism more time to detoxify it via the rhodanese pathway (Chen and Rose, 1956).

Because SN induces MetHb formation and MetHb cannot carry oxygen, most authors caution against using SN in the setting of smoke inhalation-related cyanide poisoning (Hall et al, 1989; Baskin and Brewer, 1997). Other authors concluded the use of SN and STS in seven fire victims was 'relatively safe', discharging 5 of 7 from the hospital neurologically intact (Kirk et al, 1993). Of the remaining patients, one died of severe hypoxic encephalopathy while the other survived with neurologic sequelae. It is unclear, however, whether SN may have aggravated their conditions.

Vasodilatation

Way proposed that another mechanism of protection by the nitrites might be through vasodilation (Way, 1984). SN results in the production of nitric oxide, which is responsible for vasomotor control of blood pressure and induction of vasodilatation.

Nitric oxide

Pearce and colleagues more recently proposed that nitric oxide may act as another of the organism's pathways of cyanide detoxification / protection by competing directly with cyanide at the level of the cytochrome to reversibly bind it, thereby preventing the attachment of cyanide to the molecule (Pearce, 2003).

Onset of the effect

The therapeutic effect of SN after intravenous injection occurs within minutes, of AN after inhalation within seconds. The AN effect is faster because it is directly absorbed to the arterial blood which contains more oxygenated Hb that can be transformed to MetHb. Adverse effects occur also within the same time-span. The result of nitrite administration should be monitored by the MetHb level (note: Goldfrank's Toxicologic Emergencies 8th ed. 2006 does not recommend a target MetHb-level). This parameter can be checked easily and the MetHb occurs within a very short time after the nitrite is administered. The blood gases are also very informative in monitoring the patient.

4.3.7 Safety

SN alone

SN causes vasodilatation, which may lead to a drop in blood pressure, headache, and dizziness (Way, 1984). Severe methaemoglobinaemia which interferes with oxygen-carrying capacity of the blood may also be produced. Excessive (overdose) administration of SN has likely been responsible in the deaths of two children (Berlin, 1970; Lasch and El Shawa, 1981), and was the presumed cause of death in an elderly fire victim (Hall et al, 1989). Overdose with SN has been reported by van Heijst and colleagues in 3 additional cyanide poisoned patients (Douze and van Heijst, 1973; van Heijst et al, 1987).

No further data are available on SN combined with other antidotes.

AN alone or combined with other antidotes.

No data are available.
Toxicology: List of toxic effects already reported in non-cyanide poisoned humans.

Animal data

There is considerable difference between species regarding the sensitivity against nitrite treatment in animals not poisoned by cyanide but given (sodium) nitrite. In mice the LD_{50} was 158 mg/kgbw after i.p. administration and 175 mg/kgbw after oral exposition. In the rat an oral LD_{50} of 180 mg/kgbw and s.c. LD_{50} of 96.6 mg/kgbw was found (RTCES, 2000; cited by Poisindex). Humphreys reported 150 to 170 mg nitrite/kgbw being a lethal dose for cattle (Humphreys, 1988; cited by Poisindex). For swine this will be 90 mg/kgbw (Beasley et al, 1989; cited by Poisindex). In the rabbit death occurs after 80 to 90 mg nitrite/kgbw (Beasley et al, 1989; cited by Poisindex). Furthermore, he found that 170 mg nitrite/kgbw in sheep was a lethal dose, and that foetuses and neonates are more sensitive to the drug.

Human data

Toxic (side) effects of SN and AN (in normal and excessive doses).

SN leads to MetHb formation and can cause excessive methaemoglobinaemia if overdosed, or even sometimes under normal dosage. Indeed, there exist considerable inter-individual differences in the reaction to nitrite treatment. Often levels between 20 to 25% of MetHb are recommended in patients poisoned by cyanide, however, this may be a figure already too high and in some cases the normal dose of 300 mg SN can result in higher MetHb levels (Anderson BD in Brent, 2005). As a result the patient will have problems with the oxygen transportation and possibly a severe central and peripheral hypoxia / cyanosis. The hypoxia might even cause syncopes (Anderson BD in Brent, 2005). It is especially difficult to predict the development of MetHb after SN infusion in children. Several authors reported on fatal outcomes of cyanide poisonings in children that were most probably caused by excessive MetHb production after nitrite treatment (Berlin, 1970; Ellenhorn and Barceloux, 1988; Hall et al, 1989; Lasch and El Shawa, 1981). Also, five elderly fire victims may have died from the same cause (Hall et al, 1989). AN, if used for the same purpose and in the recommended dosage is not capable of forming such high levels of MetHb so is unlikely to cause similar adverse effects (Klimmek et al, 1988). As the high CO-levels in cases of smoke inhalation may cause CO-Hb, that would be additive to the MetHb formation caused by SN, thereby further reducing the oxygen-carrying-capacity of the erythrocytes, it is not generally recommended under these circumstances (Anderson BD in Brent, 2005; citing Kulig, 1991). Some authors, however, believe this effect is not of clinical relevance (Anderson BD in Brent, 2005; citing literature not given in the list of references).

SN and AN also may lead to hypotension quite regularly (especially after rapid injection), requiring supportive care means, e.g. administration of cardio-active drugs (which are normally able to compete this adverse effect) or even resuscitation (PoisIndex, Anderson BD in Brent,

2005). The drop in blood pressure already occurs with normal doses of SN, especially in predisposed patients (Anderson BD in Brent, 2005). AN and SN should not be administered to patients suffering from current cerebral bleedings (Bain, Knowles, 1987). The hypotension causes a sympathetic reaction of the body: tachycardia occurs. Other symptoms are dizziness, headache or fainting / collapse / syncope (Anderson BD in Brent, 2005).

As nitrites can cause anaemia they should not be used during pregnancy and in paediatric patients. In patients suffering from anaemia caution must be taken and the nitrite dose has to be adjusted in respect to the Hb-levels (Anderson BD in Brent, 2005).

Patients with pre-existing low blood pressure (e.g. women or children) showed a higher sensitivity to nitrite treatment and therefore suffered from methaemoglobinaemia and hypotension more frequently.   Pregnancy in itself and malignancy can also raise the individual nitrite sensitivity (Metcalf 1961, cited by Poisindex). In patients suffering from glucose-6-phosphate dehydrogenase deficiency a greater sensitivity for nitrite induced methaemoglobinaemia was mentioned by Calabrese et al, 1980 (cited by Poisindex).

In a patient not poisoned by cyanide the orally lethal dose of SN was 2.6 g (Ten Brink et al, 1982; cited by Poisindex). Another patient ingested up to 700 mg of AN and survived despite the MetHb level reaching 49% (Bradberry et al, 1993; cited by Poisindex).

Adverse effects in cyanide poisoned humans

So far there is no evidence for any allergic potential of both nitrite antidotes. Hypotension has been discussed above.

4.3.8   Case series and casuistics of cyanide / cyanogenic poisonings treated with nitrites

There are 3 MetHb forming agents used in cyanide poisoning; AN, SN and 4-DMAP. As all MetHb forming agents do not lead to an elimination of the poison but only to a redistribution of the cyanide, they are usually applied in combination with sodium-thiosulphate (STS), which will capture liberated cyanide.

The total number of cases in which SN was used is in the range of a few dozens, which means that experience with the drug is relatively limited. More unpublished cases, treated successfully or not, undoubtedly exist, but are not accessible. Among the reported cases, details of poisoning and confirmation of poisoning are often lacking. Many of the cases reported by Chen and Rose were second hand case reports. In none of their cases reported in 1944 or 1952 as well as in the cases of Wolsie (1951) and Würzburg (1996) was a blood cyanide concentration documented. In most cases, however, the cyanide poisoning clinically was without doubt and in some cases the

victims were critically ill, suggesting that the drug is indeed efficacious when appropriately administered in a timely fashion.

Table 11: Overview of combination of AN without and with other antidote(s)

	Total	Sequence	STS	AN	SN	HOCO	Co-EDTA	DMAP	Methylene blue	BAL	Coramine	STS + SN	STS + SN + Kelocyanor	HOCO + STS + Co-EDTA	STS + coramine
AN	61	S1	5	**22**	3	0	0	0	0	0	2	**25**	1	2	1
	11	S2	1	3	1	0	1	0	0	0	0	5	0	0	0
	2	S3	0	2	0	0	0	0	0	0	0	0	0	0	0
	1	S4	0	0	0	0	0	0	0	0	0	1	0	0	0
	0	S5	0	0	0	0	0	0	0	0	0	0	0	0	0

* Data used for analysis (n ≥ 7) are in bold

Table 12: Overview of combination of SN without and with other antidote(s)

	Total	Sequence	STS	AN	SN	HOCO	Co-EDTA	DMAP	Methylene blue	BAL	Coramine	AN + STS	STS + AN + Co-EDTA	Hydroxo + STS + keylocyanor	hydroxo + STS	STS + methylene blue	STS + DMAP
SN	57	S1	**25**	3	1	0	0	0	0	0	0	**25**	1	2	0	0	0
	26	S2	**15**	1	2	0	0	0	0	0	0	5	0	0	1	1	1
	14	S3	**12**	0	1	0	0	0	0	0	0	0	0	1	0	0	0
	5	S4	3	0	0	0	0	0	1	0	0	1	0	0	0	0	0
	0	S5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

* Data used for analysis (n ≥ 7) are in bold

Table 13: Cases treated with nitrites alone

Case			Antidote treatment					Reference
			Cumulative dose of antidote		**Number of treatment steps**	**PSSa**		
Age	**Sex**	**Type of poisoning**	**AN (pearls)**	**SN (g)**		**Before**	**After**	**Reference**
26	M	HCN	NS	-	1	2	0	Wolfsie, 1951
40	M	HCN + CN salts	> 3	-	3	3	0	Wolfsie, 1951
33	M	HCN	NS	-	1	3	0	Wolfsie, 1951
44	M	HCN	NS	-	1	3	0	Wolfsie, 1951
29	M	HCN + CaCN	NS	0.3	2	3	0	Wolfsie, 1951
NS	M	HCN	NS	-	1	NS	0	Chen and Rose, 1952
67	M	HCN	NS	-	1	NS	0	Chen, 1952
44	M	HCN	NS	-	1	NS	0	Chen, 1952
NS	NS	Acrylonitrile	NS	-	1	1	3	Van Luijt, 1963
NS	NS	SNP	NS	-	1	2	0	Cetnarowski, 1986
NS	NS	HCN	NS	-	1	1	0	Würzburg, 1996
NS	NS	HCN	NS	-	1	3	0	Würzburg, 1996
NS	NS	HCN	NS	-	1	3	0	Würzburg, 1996
NS	NS	HCN	NS	-	1	2	0	Würzburg, 1996
NS	NS	HCN	NS	-	1	2	0	Würzburg, 1996
NS	NS	HCN	NS	-	1	2	0	Würzburg, 1996
NS	NS	HCN	NS	-	1	3	0	Würzburg, 1996

Case studies are detailed Appendix C
NS, not stated; -, not applied

For cases treated with nitrites and STS, see Table 14. Combinations of nitrites with other antidotes are summarised in Table 15 excerpted from Table 14.

Table 14: Cases treated with nitrites and STS (Lilly Kit): 92 cases

Case			Antidote treatment						Reference
			Cumulative dose of antidote			**No. of treatment steps**	**PSSa**		
Age	**Sex**	**Type of poisoning**	**AN (pearls)**	**SN (g)**	**STS (g)**		**Before**	**After**	
41	M	HCN	3	0.3	25	3	3	0	Potter, 1950
51	M	NS	2	0.3	25	1	3	0	Potter, 1950
32	M	KCN	4	-	NS	2	3	0	Miller and Toops, 1951
37	M	CaCN	NS	-	NS	3	2	0	Wolfsie, 1951
29	M	HCN	NS	0.3	7.5	4	3	0	Wolfsie, 1951
40	M	HCN	NS	0.3	12.5	2	3	0	Wolfsie, 1951
45	M	HCN	NS	0.3	12.5	2	3	0	Wolfsie, 1951
39	M	HCN	NS	-	5	2	2	0	Wolfsie, 1951
44	M	HCN	NS	0.3	12.5	2	3	0	Wolfsie, 1951

Case			Antidote treatment						Reference
			Cumulative dose of antidote			No. of treatment steps	PSSa		
Age	Sex	Type of poisoning	AN (pearls)	SN (g)	STS (g)		Before	After	
24	M	AgCN, NaCN	-	0.3	6.25	1	1	0	Chen and Rose,1952
NS	M	HCN	-	0.6	2	1	3	2	Chen and Rose, 1952
64	M	HCN	NS	0.3	12.5	1	3	0	Chen and Rose, 1952
22	F	CNCl	20	0.6	12.5	2	3	0	Chen and Rose, 1952
61	M	HCN	NS	0.3	12.5	2	3	0	Chen and Rose, 1952
NS	M	HCN	12.5	0.3	12.5	1	1	0	Chen and Rose, 1952
22	M	HCN	NS	0.3	12.5	1	1	0	Chen and Rose, 1952
24	M	HCN	NS	0.3	12.5	1	1	0	Chen and Rose, 1952
25	M	HCN	NS	0.3	12.5	1	1	0	Chen and Rose, 1952
26	M	HCN	NS	0.3	12.5	1	1	0	Chen and Rose, 1952
32	M	HCN	NS	0.3	12.5	1	1	0	Chen and Rose, 1952
39	M	HCN	NS	0.3	12.5	1	1	0	Chen and Rose, 1952
47	M	HCN	NS	NS	NS	1	1	2	Lurie, 1953
39	M	NS	NS	0.3	12.5	1	3	0	Chen and Rose, 1956
41	F	HCN	NS	0.5	8.1	3	3	3	Chen and Rose, 1956
6	M	Cassava	-	1.5	1.8	2	2	0	Gonsalves et al, 1956
6	M	Cassava	-	NS	NS	1	2	0	Gonsalves et al, 1956
5	F	Cassava	-	NS	NS	1	1	0	Gonsalves et al, 1956
9	M	Cassava	-	NS	NS	1	2	0	Gonsalves et al, 1956
6	M	Cassava	4	0.5	6	1	3	0	Gonsalves et al, 1956
4	F	Cassava	-	0.5	3	1	1	0	Gonsalves et al, 1956
24	M	NaCN	NS	0.3	25	2	3	0	Gonsalves, 1969
31	F	KCN	-	0.3	17.5	1	3	0	De Busk and Segal, 1969
23	M	Acetone cyanohydrin	NS	6.6	20	2	3	0	Thiess and Hey, 1969
49	M	Isobutyro-nitrile	NS	3	10	1	3	3	Thiess and Hey, 1969
1.4	M	KCN	NS	0.45	25	2	0	3	Berlin, 1970
35	M	CN salt	-	0.3	12.5	1	3	4	Lee-Jones, 1970
25	M	KCN	1	NS	>12.5	1	3	0	Lee-Jones, 1970
14	F	CN salt	1	NS	>12.5	1	3	0	Lee-Jones, 1970
30	M	KCN, AgCN	NS	NS	NS	1	2	0	Trapp, 1970
0.9	F	Laetrile	-	NS	NS	2	3	4	Humbert et al, 1977 + Braico, 1979
49	F	Apricot seeds	NS	0.3	12.5	1	2	0	Rubino and Davidoff, 1979
NS	NS	Apricot kernel	-	NS	NS	1	1	4	Lasch and El Shawa, 1981
32	F	Laetrile	NS	0.3	0.05	1	2	0	Moss, 1981
60	M	KCN	-	NS	NS	1	3	0	Feihl, 1982
58	F	SNP	-	0.3	12.5	1	3	0	Marbury, 1982
31	M	KCN	-	0.9	225	1	3	0	Peters, 1982

Case			Antidote treatment						Reference
			Cumulative dose of antidote			No. of treatment steps	PSSa		
Age	Sex	Type of poisoning	AN (pearls)	SN (g)	STS (g)		Before	After	
67	F	Bitter almonds	NS	0.3	12.5	1	3	0	Shragg, 1982
23	M	Laetrile	NS	0.6	12.5	2	3	2	Beamer, 1983
23	F	KCN	NS	0.3	12.5	1	2	0	Litovitz, 1983
25	F	NS	NS	0.3	12.5	2	3	4	Litovitz, 1983
24	M	Acrylonitrile	NS	39	162.5	1	2	0	Vogel, 1984
18	M	KCN	-	NS	NS	1	3	2	Uitti, 1985
59	M	KCN	-	0.3	12.5	1	3	0	Wesson, 1985
4	M	Laetrile	NS	0.15	6.25	2	3	0	Hall, 1986
56	F	Amygdalin	-	0.6	NS	1	3	0	Leor, 1986
34	M	KCN	-	0.3	12.5	3	2	0	Hall, 1987
4	NS	Laetrile	NS,	-	NS	1	3	0	Hall, 1987
23	M	CN salt	1	0.45	18.75	1	3	0	Krieg and Saxena, 1987
2.5	F	CN salt	1	0.076	2.5	1	3	0	Krieg, 1987
30	M	NaCN	2	0.3	12.5	1	2	0	Johnson and Mellors, 1988
29	M	KCN + arsenic CN	-	0.3	12.5	1	3	0	Di Napoli, 1989
24	F	KCN	-	0.3	12.5	1	3	0	Johnson, 1989
46	M	KCN	-	NS	NS	1	3	2	Rosenberg, 1989
24	M	KCN	-	NS	NS	1	3	0	Feldman, 1990
31	M	KCN	NS	0.6	25	1	3	0	Selden, 1990
2	F	Acetonitrile	NS	0.099	42.5	1	3	0	Kurt, 1991
2	M	Acetonitrile	1	-	8.25	2	2	0	Losek, 1991
39	F	Acetonitrile	-	0.6	50	4	3	0	Turchen et al, 1991
31	M	KCN	NS	-	NS	1	3	0	Kasamo, 1993
28	M	Propionitrile	-	0.3	12.5	1	3	1	Scolnick, 1993
34	M	Propionitrile	-	0.3	12.5	1	2	0	Scolnick, 1993
54	M	KCN	NS	0.6	25	1	2	3	Goodhart, 1994
39	F	Acetonitrile	-	> 1.5	100	2	3	0	Mueller and Borland, 1997
NS	NS	HCN	-	NS	NS	1	3	4	Würzburg, 1996
NS	NS	NaCN	-	NS	NS	1	1	0	Würzburg, 1996
19	F	NS	-	NS	NS	1	3	0	Martin-Bermudez, 1997
41	F	Apricot kernel	NS	0.45	> 25	3	3	0	Suchard, 1998
4	F	Cassava	-	0.12	6.75	1	2	0	Ruangkanchanasetr, 1999
19	F	CN salt	-	0.45	33.25	2	3	0	Chin and Calderon, 2000
19	F	HCN	1	-	12.5	1	3	3	Lam and Lau, 2000
NS	M	HCN	1	0.3	12.5	1	1	0	Lam and Lau, 2000
35	M	NS	-	2	17	1	3	1	Muraki, 2001

Case			Antidote treatment						Reference
			Cumulative dose of antidote			No. of treatment steps	PSSa		
Age	Sex	Type of poisoning	AN (pearls)	SN (g)	STS (g)		Before	After	
78	F	SNP	-	0.6	25	1	2	4	Sipe, 2001
52	M	Unknown	0.3	0.3	12.5	1	3	0	Zavotsky, 2004
38	M	Pentene nitrile	NS	NS	NS	1	2	0	Fernández et al, 2008
14	F	SNP	-	0.3	18.5	1	0	0	Quinlan, 2008
31	M	KCN	3	-	10	1	3	0	Nakatani, 1992
NS	NS	Cassava	Check: 1, 0.2 ml/kg	0.42 [a]	17.5 [b]	1	3	0	Espinoza et al, 1992
NS	NS	Cassava	Check: 1, 0.2 ml/kg	0.42 [a]	17.5 [b]	1	3	0	Espinoza et al, 1992
NS	NS	Cassava	Check: 1, 0.2 ml/kg	0.42 [ab]	17.5 [b]	1	3	0	Espinoza et al, 1992
NS	NS	Cassava	Check: 1, 0.2 ml/kg	0.42 [a]	17.5 [b]	1	3	0	Espinoza et al, 1992

Case studies are detailed Appendix C

NS, not stated; -, not applied

[a] Reported as SN 3% (0.2 ml/kgbw)

[b] Reported as STS 25% (1 ml/kgbw).

Cases treated with nitrites in combination with STS and other antidotes: see Table 15.

Table 15: Cases treated with nitrites combined with other antidotes

Case			Antidote treatment						Number of treatment steps	PSSa		Reference
Age	Sex	Type of poisoning	Cumulative dose of antidote (g)									
			AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO		Before	After	
With DMAP												
19	M	KCN	-	0.15	-	0.125	-	-	2	2	1	Werner, 1979
With STS and DMAP												
21	M	NaCN	-	0.3	20	> 1.75	-	-	2	3	4	Van Dijk and van Heijst, 1971
With Co-EDTA												
35	M	CN salt	11	-	-	-	0.6	-	1	3	0	Naughton, 1974
With STS and Co-EDTA												
48	M	NS	2	0.3	12.5	-	0.3	-	2	2	0	Davis and Ewer, 1988
With STS and Co-EDTA												
80	M	KCN	-	0.6	3	-	-	4	1	3	0	Mannaioni, 2002
With STS, Co-EDTA and HOCO												
NS	M	KCN	-	NS	NS	-	0.6	4	1	2	0	Lutier, 1971
NS	NS	KCN	NS	-	NS	-	0.6	4	1	2	0	Lutier, 1971
23	M	KCN	-	0.5	75	-	0.3	3	1	3	4	Buchanan, 1976

Case studies are detailed Appendix C
NS, not stated; -, not applied

4.3.9   Occurrence of adverse effects in clinical use

SN is not without significant side effects. Several descriptions of overdose with SN have been reported, with severe degrees of methaemoglobinaemia, hypotension, and some deaths that may be ascribed to its overzealous use. Most experts agree that SN should be used with extreme caution if at all in the setting of smoke intoxication. This is important, as smoke inhalation constitutes the most common source of hydrogen cyanide poisoning. There appears to be hesitation on the part of many physicians to administer SN when any doubt exists as to the identity of the poison. This indecision may be warranted, as misidentification is not uncommon in the initial hours after many chemical incidents. For example, in the 1998 mass poisoning in Japan, where 42 patients were hospitalised and 21 treated for cyanide poisoning and released after eating a curry distributed at a local festival. It was later discovered that the poison was not cyanide at all, but rather arsenic. It remains unknown if any of the 4 victims' deaths might have been accelerated by the use of inappropriate antidotes. In short, antidotes should be ideally safe enough to allow errors in toxicant identification. This may not be the case for SN, which should prompt its use only in definite cyanide poisoning (clinically or cyanide analytics).

4.3.10 Assessment of efficacy and effectiveness in clinical use

AN alone

Sequence 1   n = 22   p = 0.0008 *** Median PSSa before: 2.5 – PSSa After : 0

AN in combination with any other antidote

Sequence 1   n = 61   p = < 0.0001 ***   Median PSSa before: 3 – PSSa After: 0
Sequence 2   n = 11   p = 0.0199 *   Median PSSa before: 3 – PSSa After: 2

Amyl Nitrite and Sodium Nitrite

Sequence 1   n = 3   No statistical analysis
Sequence 2   n = 1   No statistical analysis

Amyl Nitrite and Co-EDTA

Sequence 2   n = 1   No statistical analysis

SN alone: No statistical analysis

SN in combination with any other antidote

Sequence 1   n = 57   p = < 0.0001 ***   Median PSSa before: 3 – PSSa After: 0

Sequence 2   n = 26   p = 0.0042 **   Median PSSa before: 3 – PSSa After: 1.5

Sequence 3   n = 14   p = 0.0105 *   Median PSSa before: 3 – PSSa After: 0.5

Note: See Section 4.2.10 for nitrites and STS, other combinations not analysed (n < 7).

Overall assessment of efficacy and effectiveness of nitrites alone and in combination

Data from the sequential PSSa a study support the following assumptions.

Amyl Nitrite

AN was used alone or in combination with other antidotes to cyanide in 99 sequences allowing determination of PSSa before and after its administration.

AN was shown efficient when used alone in 22 cases in sequence 1. AN was highly efficient resulting in full recovery (median PSSa after 0) in moderate to severe poisoning (median PSSa before 2.5) (p = 0.0008).

AN was shown efficient when used in combination with other antidotes in 72 cases.

- In sequence 1, AN was used in 61 cases.
  - AN has highly efficient resulting in full recovery (median PSSa after 0) in severe poisoning (PSSa before 3) (p < 0.0001).
- In sequence 2, AN was used in 11 cases.
  - AN was partially efficient resulting in moderate poisoning (median PSSa after 2) when administered in severe poisoning (PSSa before 3) (p = 0.0199).

Antidotes were combined with AN.

The antidotes combined with AN were STS (n = 6), SN (n = 4), STS + SN (n = 31).

Sodium nitrite

SN was used alone in 1 case only in sequence 1 only precluding any further statistical analysis.

SN was used in combination.

- Sequence 1
  - SN was used in 57 cases.

- o SN was highly efficient resulting in full recovery (median PSSa after 0) in severe poisoning (median PSSa before 3) (p<0.0001).
- Sequence 2
  - SN was used in 26 cases.
    - o SN was partially efficient resulting in partial recovery (median PSSa after 1.5) in severe poisoning (median PSSa before 3) (p = 0.0042).
- Sequence 3
  - SN was used in 14 cases.
    - o SN was partially efficient resulting in partial recovery (median PSSa after 0.5) in severe poisoning (median PSSa before 3) (p = 0.0105).

Antidotes were combined with SN.

The antidotes combined with SN were STS (n = 55), AN (n = 4); AN + STS (n = 31).

4.3.11 Gaps in knowledge

The main point concerning nitrites as cyanide antidotes is the lack of knowledge regarding the exact pharmacodynamics. Further research should clarify how nitrites can minimise cyanide toxicity apart from MetHb formation (as MetHb formation was modest in many cases).

Another issue is the lack of information regarding the interaction of SN and AN with other nitrites, e.g. pharmaceutical used organic nitrates and PDE5-inhibitors. It would be interesting to know if an additional effect occurs in patients treated with these substances as far as hypotension / vasodilation are concerned. If pre-treatment is known, certain safety precautions (e.g. preparedness for shock therapy) would have to be taken. If nitrates and PDE5-inhibitors are combined an additive effect takes place so it would be reasonable to expect the same in nitrite-nitrate or nitrite-PDE5-inhibitor combinations as well.

To date no randomised studies have been undertaken in respect to the possible additive negative effect of nitrite-induced MetHb formation on the oxygenation in CO-poisoned patients, e.g. smoke inhalation. Severe CO-intoxication after smoke-inhalation is mostly combined with higher cyanide levels (Baud, 2007). If more data were collected about the exact way of action of nitrite-antidotes and the outcome of more cases of smoke-inhalation treated with SN and AN, perhaps better recommendations could be made about the treatment of this frequently occurring form of intoxication. Due to theoretical considerations MetHb forming agents are harmful in smoke inhalation because they further reduce oxygen transport capacity. There are studies in animal in which MetHb was effective in protection against smoke intoxication. For ethical reasons it would not be possible to undertake further studies in humans.

4.3.12 Conclusion

There are some advantages of SN as an antidote. First, it is relatively inexpensive and relatively stable in solution when kept under reasonable conditions of temperature and light. One disadvantage is that it must be injected i.v. or intraosseously (i.o.), something which is not always practical in the non-clinical, e.g. industrial environment. The i.v. injection of SN can also be managed quickly as the amount of 300 mg SN is dissolved in only 10 ml of fluid.

Experimental and limited clinical data suggest that SN is an effective antidote against cyanide poisoning. Because it has most often been administered with STS, it is difficult to discern its efficacy in human poisoning alone. However, effectiveness was seen before STS application, which is indicated to capture liberate cyanide during MetHb re-reduction Relative certainty in the diagnosis is required before administering SN due to its reduction of oxygen carrying capacity and vasodilatative effect, which could aggravate other poisonings or medication effects. Specifically, it should likely not be employed in the setting of cyanide poisoning associated with smoke inhalation which limits its applicability. SN is relatively inexpensive and stable. Requirement for i.v. administration limits its application somewhat in the non-clinical setting.

AN can be administered immediately only breaking the ampoules. In industrial settings there may be a role for AN alone.

Both SN and AN develop their antidotal effect very rapidly so they are efficient in severe intoxications. As they might have a mode of action apart from MetHb formation they might also be effective in relatively low doses with consequently low MetHb-levels and so less effect on oxygen supply. But the antidote kit can also be administered twice if the first dose does not result in an adequate clinical improvement.

4.4   4-Dimethylaminophenol

4.4.1   Introduction

4-DMAP was developed and studied in the laboratories of the Walther Straub Institute of the Ludwig Maximilian University Munich. Its development was supported by the German army because at that time it was thought that cyanide gas could be used as chemical warfare agent.

4-dimethylaminophenol was developed as it met the criteria of rapidly creating sufficient MetHb with little adverse effects. In 1969 aminophenols were tested for their formation of ferri-Hb in humans by Kiese and Weger (1965, 1969). It was first used in human cyanide poisoning successfully by Daunderer (1972). As severe cyanide poisoning has become a very rare incident, only single case reports have been published since. In Munich, a series of

13 clinical case reports involving use of 4-DMAP between 1973 and 1979 were documented in a thesis (Werner, 1979). A further series of 9 cases from between 1981 and 1991 were also recorded (Stickel, 2008). 4-DMAP was also used in the Netherlands (van Dijk et al, 1987; van Heijst et al, 1987). 4-DMAP is registered as a pharmaceutical drug by the German authorities (Bundesinstitut für Arzneimittel und Medizinprodukte, BfArM). Its use permit was renewed in 1991 and the current manufacturer is Dr. Franz Koehler Chemie, 64665 Alsbach-Hähnlein Neue Bergstr. 3-7, Alsbach-Hähnlein, Germany.

4.4.2 Identity

IUPAC name:	4-Dimethylparaaminophenol (4-DMAP)
CAS Registry number:	619-60-3 (4-dimethylaminophenol), 5882-48-4
	(4-dimethylaminophenol hydrochloride)
Formula:	$C_8H_{11}ON.HCl$
Molecular weight:	173.5, 173.6
Chemical structure	
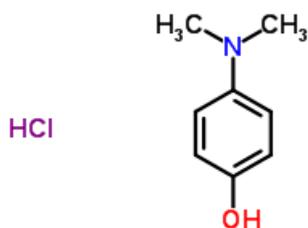
(ChemSpider, 2012)

4-DMAP (hydrochloride) is a white, crystal powder without odour or colour. Solutions in water and ethanol are clear. If exposed to air, the colour changes to a reddish-brown by auto-oxidation.

4.4.3 Practicality

Shelf-life, storage condition, stability

As 4-DMAP is readily oxidised, turning black-brown., itmust be stored in sealed opaque containers and cannot be stored, if opened. 4-DMAP stored under the conditions recommended by the supplier has a shelf life of up to 3 years. 4-DMAP is administered i.v. (5 ml solution containing 250 mg of 4-DMAP) within 1min. Monitoring of the vital parameters is done as a matter of routine when 4-DMAP is being administered. The oxygen saturation on pulsoximetry will seemingly within minutes of administration due to the formation of methaemoglobin. This is an intended effect, being the mode of action, although it should not exceed 30%. The amount of

MetHb formed can easily be measured by modern equipment (haemoglobin analysers). One package of 4-DMAP contains 5 ampoules. This amount is sufficient to treat several patients.

Pharmacokinetics and metabolism

Absorption

The i.m. injection of 3.25 mg/kg 4-DMAP leads to a Methaemoglobin level of 35% after 15 minutes, and a maximum of 41.6% after 30 minutes. The administration of 15 mg/kg 4-DMAP orally creates 35% of MetHb within 30 minutes, but bioavailability by the oral route is uncertain (Klimmek et al, 1983).

Distribution

In humans and dogs, 4-DMAP (3.25 mg/kg i.v.) is cleared rapidly from the blood with a half-life of less than 1 minute (Eyer et al, 1971). This rapid clearance is due to various first-pass effects (Klimmek et al, 1983). Experiments using C_{14}-labelled 4-DMAP in dogs showed that approximately one third of the 4-DMAP was found in red blood cells, and two thirds in plasma and the extracellular space (apparent volume of distribution 0.17 l/kg) (Eyer et al, 1978). To understand the particular pharmacokinetics of 4-DMAP, one must differentiate between the metabolism in erythrocytes and elsewhere, mainly in the liver. The distribution of 4-DMAP between plasma and erythrocytes is not known because of the extremelyshort half-life of 4-DMAP within red blood cells.

Metabolism and elimination of 4-DMAP via Erythrocytes

4-DMAP is co-oxidised rapidly with oxy-Hb to form MetHb and a phenoxyl radical. The phenoxyl radical oxidises deoxy-Hb, sustaining the catalytic cycle of MetHb formation (Eyer et al, 1984). Alternatively, the phenoxyl radical disproportionates to form 4-DMAP and a quinone-imine that is bound covalently to Hb SH groups (Eyer et al, 1983). In the presence of high concentrations of glutathione, as occurs in erythrocytes, the quinone-imine undergoes sequential addition / oxidation reactions with formation of mono-glutathione, bis-glutathione, and tris-glutathione adducts of 4-DMAP (Ludwig et al, 1995). The tri-substituted conjugate is not oxidised further but is actively excreted from the erythrocyte into plasma (Eckert et al, 1986). Studies in dogs have shown that this conjugate has a half-life of about 1 hour in plasma and is processed further by the kidneys, being excreted mainly as a tris-cysteinyl derivative of DMAP (Jansco et al, 1981). It has been calculated that probably all 4-DMAP thioethers excreted (15% of the dose) originate from the metabolism of 4-DMAP within the red blood cells. About the same amount of 4-DMAP is bound covalently to the Hb SH groups.

Metabolism and elimination of 4-DMAP via the liver and kidney

About 50% of the 4-DMAP administered i.v. to humans is transformed in the liver to the glucuronide and sulphate conjugate. In urine, 41% 4-DMAP glucuronide and 12% 4-DMAP sulphate were detected (Klimmek et al, 1983). Conjugation appears to occur rapidly since covalent binding to liver proteins or formation of glutathione conjugates in isolated Hb-free perfused rat liver were of no importance (Eyer et al, 1978). First-pass metabolism in the liver may explain why a much higher dose of 4-DMAP is required by the oral route compared with parenteral administration.

4.4.4   Mode of action, dosage and time of onset

In experiments on seven volunteers an i.v. bolus of 3.25 mg 4-DMAP/kg resulted in 15% MetHb after 1 minute. After 10 minutes the concentration was 26% and the peak value of 31% was reached after 30 minutes (Kiese and Weger, 1969; Klimmek et al, 1983). The spontaneous half-life of MetHb was 117 minutes (Zilker et al, 2005). After 4 hours the MetHb concentration had dropped back to 10% (figure 5).

Figure 5: Course of MetHb-level in cyanide poisoned patients with different doses and human volunteers after 4-DMAP i.v.
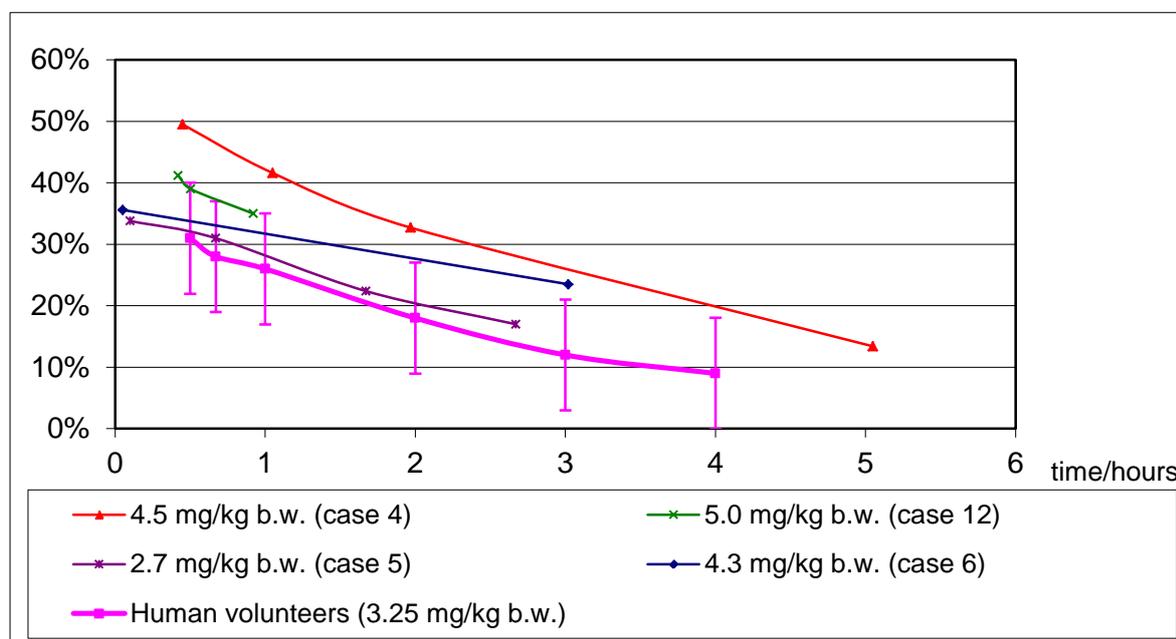
The rate of spontaneous reduction of MetHb to ferro-Hb was found to be 8%/hour. *In vivo* 1 mol 4-DMAP yields 15 equivalents of MetHb, which means that in practice one molecules of

4-DMAP converted 15 molecules of Hb to MetHb (Klimmek et al, 1983). The catalytic formation of MetHb by 4-DMAP takes place very rapidly, but due to the very short (<1 minute) half-life of 4-DMAP within the erythrocytes, the effect is not long lasting with MetHb formation ceasing just10 minutes after a bolus injection (Eyer et al, 1975), this being (Eyer et al, 1973). 4-DMAP (3.25 mg/kg) given i.v. to dogs 1 minute after being poisoned with a lethal dose of KCN (4 mg/kg) resulted in the survival of all dogs (Klimmek et al, 1979). The peak concentration of MetHb was 38.8% ± 1.7% (Eyer et al, 1975).

Dosage

The recommended dose is 3.5 mg/kgbw, which is equivalent to the contents of a single ampoule, for a 70 kg person. As a rule of thumb a normal weight person receives one ampoule (5ml), an overweight individual, one and a half (7.5 ml); and an underweight person, half an ampoule (2.5 ml). However, single cases have shown a sufficient and immediate effect (PSSa 2 and 3 decreasing tro 0) of application of 100 and 150 mg in patients of normal weight, resulting in Met-Hb levels of 5.5 and 16.3% (Steffens, 2003). Overdosing with 4-DMAP must be avoided. It is possible to administer 4-DMAP in the same dose i.m. in mass poisoning events.

Methaemoglobin formation in patients -

MetHb formation after 250 mg of 4-DMAP amounts to 33% and 49.5% with a half-life around 140 minutes as shown in Figure 5.

500 mg of 4-DMAP, which is an overdose, can lead to a long-lasting MetHb formation. It is likely that most of the MetHb found in overdosed cases stemmed from extracellular MetHb due to haemolysis. In an overdosed patient 1,000 mg of 4-DMAP produced a life-threatening MetHb level over 70%.

Repeated 4-DMAP administrations are not necessary if STS administration follows.

4.4.5 Safety

Before considering administering 4-DMAP to a patient one should be quite certain that he/she has been poisoned by cyanide. It should only be used if the patient is in coma. It should not be used in cases of smoke inhalation, even if the patient is comatose.

There is no experience of use of 4-DMAP in neonates or children. Glucose-6-phosphate dehydrogenase (G6PDH) deficiency is thought to be a contraindication for the use of 4-DMAP

since they are unable to reduce MetHb by the pentose phosphate shunt and may develop a long lasting methaemoglobinaemia and severe haemolysis. Although this is a very rare disease in Caucasians in Europe it may be more prevalent in the US with a larger Latin-American and Afro-American population. As far as can be seen from literature, the drug has never been used in such a case.

There are two major adverse effects which are closely related to the desired action of 4-DMAP - first, that too much MetHb is produced; and second, that haemolysis is induced. Taking published experiences into consideration, 4-DMAP did not induce significant haemolysis up to a dose of 5 mg/kgbw (Zilker and Eyer, 2005; Werner, 1979; Stickel, 2008). According to these experiences the recommended dose of 3.5 mg/kgbw did never create an excessive MetHb level. In one fatal case of cyanide poisoning a too high MetHb level was observed after the recommended dose of 4-DMAP but nitrite had also been given previously and significantly contributed to the met-Hb level (van Dijk et al, 1987). In vitro experiments have shown that the MetHb production rate at atmospheric oxygen pressure was only 60% of that at venous blood oxygen pressure (40 mm Hg). This may be of some importance in hypoxic patients when cardiopulmonary insufficiency is present (Eyer et al, 1975). This should not, however, prompt reducing the dose of 4-DMAP in such circumstances as long as the patient is ventilated with a fraction of inspired oxygen (FiO$_2$) of 1.0.

There are some minor adverse effects which are of little relevance in severe poisoned patients. Phlebitis was observed 6-7 days after 4-DMAP infusion via the ante-cubital vein. Following an i.m. injection of 4-DMAP a slight pressure was felt after 5 to 10 minutes at the site of injection and this slowly grew in intensity, finally resulting in severe pain. Approximately 10 h after the injection shivering, sweating, and fever developed. In test persons receiving i.v. injection of 4-DMAP (3.25 mg/kg) the total bilirubin concentration increased by 140%, that of conjugated bilirubin by 180% and that of iron by 200%. Within 24 hours of an i.m. injection of 4-DMAP (3.5 mg/kg) the total bilirubin increased by 270% and then declined rapidly, while the bilirubin concentration rose by 120% and that of iron by 50% (Klimmek et al, 1983).

Treatment of adverse effects

Excess methaemoglobinaemia may be corrected by 2 mg/kgbw toluidine blue or by 1 mg/kgbw methylene blue i.v. This should be only done if within 1 hour after the administration of 4-DMAP the MetHb level exceeds 60%. It should be kept in mind that doing so will release cyanide again thus thiosulphate infusion is mandatory. Exchange transfusion is needed if the MetHb level remains high for a long period since this MetHb comes from haemolysis and cannot be reversed again.

Adverse effects related to DMAP

No other adverse effects have been reported in the relatively low number of cases treated with DMAP.

4.4.6 Case series and casuistics of cyanide / cyanogenic poisonings treated with DMAP

There are limited data available for the use of 4-DMAP in cyanide poisoning. This is due to cyanide poisonings being a relatively rare event and that most individuals are not found alive. Some single case reports are published (Daunderer et al, 1974; van Dijk et al, 1987; van Heijst et al, 1987; Jakobs, 1984; Weger, 1975). It should be recognised that there may be bias towards publication of successful outcomes.

Two cases were treated with DMAP alone, see Table 16.

Table 16: Cases treated with DMAP alone

Case			Antidote treatment				Reference
Age	Sex	Type of poisoning	Cumulative dose of DMAP (g)	Number of treatment steps	PSSa Before	After	
26	F	KCN	0.25	1	1	0	Werner, 1979
50	M	Benzyl-CN	0.25	1	1	0	Werner, 1979

Case studies are detailed Appendix C.

The 25 cases treated with DMAP and STS are reported in Table 17.

Table 17: Cases treated with DMAP and STS

Case			Antidote treatment						
			Cumulative dose of antidote (g)		Number of treatment steps	PSSa			
Age	Sex	Type of poisoning	DMAP	STS		Before	After		Reference
27	M	KCN	0.25	10	1	1	0		Werner, 1979
43	M	NaCN	0.25	16	2	3	3		Werner, 1979
29	M	KCN	0.25	12	1	1	0		Werner, 1979
20	F	HCN	0.25	8	1	1	0		Werner, 1979
19	M	KCN	0.25	5	1	2	0		Werner, 1979
25	M	KCN	0.375	3	2	3	3		Werner, 1979
27	F	KCN	0.25	25	1	0	0		Werner, 1979
30	F	Bitter almonds	0.25	10	1	1	0		Werner, 1979
66	M	KCN	0.5	10	1	2	0		Werner, 1979
35	F	Bitter almonds	0.5	15	1	1	1		Werner, 1979
59	M	SNP	0.25	12.5	1	3	0		Ram, 1989
30	M	Acetonitrile	0.25	> 1	2	1	0		Michaelis, 1991
29	M	$KAu(CN)_2$	0.25	NS	1	3	0		Kampe, 2000
24	M	KCN	0.5	10	2	2	0		Zilker and Stickel, 2008
29	M	NaCN 1 g	0.5	12.5	1	3	0		Zilker and Stickel, 2008
28	F	KCN	0.5	12.5	1	2	2		Zilker and Stickel, 2008
39	M	KCN	0.25	2	2	3	2		Zilker and Stickel, 2008
56	M	NaCN	1	12.5	1	2	2		Zilker and Stickel, 2008
17	M	KCN	0.3	17	1	3	4		Zilker and Stickel, 2008
36	F	KCN	0.25	1	1	3	2		Zilker and Stickel, 2008
40	F	CN salt	0.25	1	1	3	3		Zilker and Stickel, 2008
37	F	KCN	NS	NS	1	3	2		Zaknum (2005) & Rachinger (2002, 2005)
NS	M	HCN	0.15	4	1	3	0		Steffens, 2003
NS	M	HCN	0.1	3	1	2	0		Steffens, 2003
64	F	KCN	0.25	10	1	2	0		Zilker and Solingen, 2010

Case studies are detailed Appendix C

In one case, DMAP was used preceded by SN, see Table 17 excerpted in Table 18. DMAP was not used in conjunction with the other antidotes of interest.

Table 18: Case treated with nitrites and DMAP (Werner, 1979)

Case			Antidote treatment						
			Cumulative dose of antidote (g)			Number of treatment steps	PSSa		
Age	Sex	Type of poisoning	AN (pearls)	SN	DMAP		Before	After	
19	M	KCN	-	0.125	0.15	2	2	1	

Case studies are detailed Appendix C
Note: See Section 4.2.10 for DMAP and STS, other combinations not analysed (n < 7)

4.4.7   Occurrence of adverse effects in clinical use

In a series of 23 patients receiving 4-DMAP evaluated for this report the eldest person was 66 years old. This patient received 9.3 mg/kg 4-DMAP. He developed a mild haemolysis with a rise in LDH to 378 U/l and Hb falling from 13.8 g/dl to 10.0 g/dl within 5 days. The bilirubin rose to 3.0 mg/l. As he was given twice the recommended dose, which in younger patients led to similar effects, it would appear that there is no difference between elder and younger people in reacting to this dose.

4.4.8   Assessment of efficacy and effectiveness clinical use

Table 19: Overview of combination of DMAP without and with other antidote(s)

Total	Sequence	STS	AN	SN	HOCO	Co-EDTA	DMAP	Methylene blue	BAL	Coramine	STS + SN
26	S1	**23**	0	0	0	1	2	0	0	0	0
6	S2	1	0	0	0	0	4	0	0	0	1
1	S3	0	0	0	0	0	1	0	0	0	0
0	S4	0	0	0	0	0	0	0	0	0	0
0	S5	0	0	0	0	0	0	0	0	0	0

* Emboldened are data used for analysis (n ≥ 7)

Assessment of efficacy using sequential PSS

Data from the sequential PSSa a study support the following assumptions.

DMAP was used alone or in combination with the other antidotes to cyanide in 33 cases.

DMAP was used alone in 7 cases, namely 2 cases in sequence 1, 4 in sequence 2; 1 in sequence 3, precluding any statistical analysis.

DMAP was used in combination with other antidotes in 26 cases in sequence 1.

- DMAP combined with STS was highly efficient resulting in full recovery in moderate poisoning (PSSa 2 → 0) (p = 0.0004).
- DMAP was used in combination with other antidotes in 2 cases in sequence 2 precluding any statistical analysis.
- The antidote combined with DMAP were STS (n = 23), Co-EDTA (n = 1).

Note: See Section 4.2.10 for DMAP and STS, other combinations not analysed (n < 7).

Global assessment of efficacy and effectiveness of each antidote alone and in combination

In animal and human studies and from experiences in human cyanide poisoned cases 4-DMAP is efficacious. The rate of survival in severe cases is 50%. It is likely that these cases would not have survived without that treatment. On the available experience it appears equal in efficacy to nitrites. The costs are relatively low and storage is not problematic. It can also be used for mass poisonings since it can be given i.m. in desperate situaitons.

Currently 4-DMAP is only available in the pharmaceutical market in Germany and some adjacent countries, but it was used in the Netherlands and, according to the poison control centre of Munich, in Austria.

4.4.9 Gaps in knowledge

It is not known if 4-DMAP acts via any other mechanism of action other than the formation of methaemoglobin. It may be that it also acts through a protection of cytochrome aa_3 by creating NO, as has been postulated for SN (see above). Nothing is known about its effectiveness and safety of 4-DMAP in smoke inhalation. On theoretical reasons it should not be used in smoke inhalation because the methaemoglobinaemia formed DMAP will be additional to any COHb from CO exposure and this may critically impair oxygen transport in the already compromised patient. Studies in this direction are therefore not to be expected.

4.4.10 Conclusion

Animal experiments and limited clinical data suggest that 4-DMAP is an effective antidote against cyanide poisoning. Although in most cases it was used in combination with STS and therefore it is difficult to distinguish the efficacy of the two drugs, Single cases have received DMAP only successfully. There was, in most cases, an improvement shortly after its administration and before the use of STS, suggesting that 4-DMAP was effective. In cases of severe cyanide poisoning 4-DMAP is probably life-saving, if administered in time. 4-DMAP produces more MetHb more rapidly than nitrites, although it has to be acknowledged that since both treatments have an equal chance of successful outcome, the speed of forming MetHb might not be of utmost importance. It should not be used in the case of smoke inhalation since the induced methaemoglobinaemia may be additive to CO poisoning. However, there is no evidence that it has ever been tested in such circumstances. The possibility to administer 4-DMAP i.m., might make it an appropriate antidote for very severe cases in the industrial environment and for use in treatment of mass casualties, for e.g. in terror attacks.

4.5 *Dicobalt edetate*

4.5.1 Introduction

Cobalt ions can bind cyanide and reactivate the cyanide-inhibited cytochrome oxidase (Tarkowski, 1966). However, the use of cobalt as a cyanide antidote has not received general support, mainly because of the toxicity of cobalt ion (Way, 1984).

Dicobalt ethylenediaminetetraacetate or dicobalt edetate (DCE) is an organic salt containing cobalt. It is a cyanide antidote utilised in Europe where it is authorised for use in the treatment of severe cyanide poisoning in the United Kingdom and in France (EMEA, 2003). DCE is the antidote of choice in the United Kingdom with the condition that cyanide poisoning is known for certain (Meredith et al, 1992).

4.5.2 Identity

IUPAC name:	Dicobalt edetate (hexahydrate)
CAS Registry number:	36499-65-7
Formula:	$C_{10}H_{12}Co_2N_2O_8.6H_2O$
Molecular weight:	514.2

Chemical structure:

 (Wikipedia, 2012)
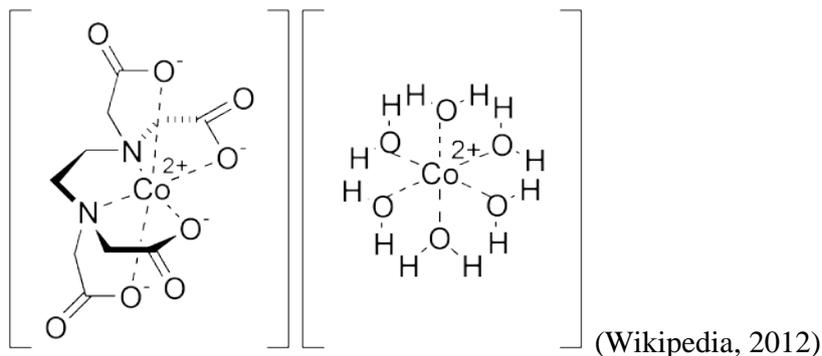
The usual preparation of DCE is Kelocyanor®.

4.5.3 Practicality

DCE is commercialised in Europe as Kelocyanor®, 300 mg ampoules, a trademark of Laroche Navarron Laboratories. Injectable Kelocyanor® is a clear violet solution, produced in clear glass 20 ml ampoules, of pH 4.0-7.5. It contains 0.196-0.240 g/100 ml free cobalt and 1.35-1.65 g/100 ml Co-EDTA, as well as 4 g glucose per ampoule.

The shelf-life is 3 years at 25°C and DCE should be stored in the dark. The drug has been stored for up to 3 years at room temperature 20-25, 30 and 37°C, the batches having been tested at regular intervals (12 months maximum) for compliance with the finished product specification with respect to appearance, extractable volume, pH and free cobalt and DCE content. The only deviation noted was that the colour of the solution became lighter at 30°C and above. All the other variables remained within the specification. If stored in the light, the drug bleaches in about one month.

The recommended dose of DCE is 300 mg i.v., which should be followed by 50ml of glucose 50% i.v.and can be repeated once or twice (Aaron, 1998; Borron and Baud, 1996; 1998; Dodds and McKnight, 1985; Hillman et al, 1974; Martindale, 1989b; Mégarbane et al, 2003; Rousselin and Garnier 1985).

4.5.4 Pharmacokinetics and metabolism

Pharmacokinetics in non-poisoned animals / healthy volunteers

There are no formal pharmacokinetics data available on the antidote in healthy volunteers, in poisoned patients or on the complex of detoxification in poisoned patients.

In mice excretion is mainly by the renal route (Frankenberg and Sörbo, 1975). According to data from the producer, after injection of a 20 mg/kg dose by i.v. route in rabbits, 52.7% of the administered drug is eliminated in the urine within the first 24 h and 60% by 48 hours.

4.5.5 Mode of action, dosage and time of onset

The use of cobalt derivatives is based on the experimental evidence that the presence of the cobalt ion can reactivate the cyanide-inhibited cytochrome oxidase (Tarkowski, 1966 cited by Way, 1984).

DCE acts by direct, non-enzymatic complexation of cyanide I, and the antidotal effect is dependent on the molar ratio of cobalt to cyanide (Way, 1984).

No additive effect of DCE and STS was found (Evans, 1964, Way 1984).

Stoichiometry – implications for dosage

Theoretically antagonising a LD_{50} of 50 mg of hydrocyanic acid (about 1,850 μmol) requires the administration of 930 μmol of DCE, or about 380 mg of cobalt (Evans, 1964).

The *in vivo* stoichiometry of the reaction using various doses of the cyanide and the DCE in mice and rabbits was assessed in order to calculate ratio of cyanide to cobalt until the antidote was no longer effective. One mole of Co^{++} can bind six moles of cyanide. A two-phased reaction has been proposed in which cobaltocyanides ($M_4Co(CN)_6$) and cobalticyanides ($M_3Co(CN)_6$) are first formed. However, DCE is only efficacious in a 2:1 ratio (Evans, 1964). The efficient dose of dicobalt-EDTA was found to be time dependant in cats and dogs, with an i.v. dose of 2 mg/kg and greater being efficient in animals with cyanide-induced primary apnoea. Later on during the course of cyanide poisoning, at the stage of return of breathing, a dose of 4 to 5 mg/kg was efficient. At the stage of secondary apnoea, a dose of 8 mg/kg and greater was efficient (Paulet, 1958).

Onset of the effect (animal and human studies)

An exact time for the onset of the effect cannot be given. In experimental studies, DCE was shown to be one of the most rapid and efficient antidotes to cyanide (Paulet, 1984). In cyanide-poisoned dogs, DCE (10 mg/kg) was the only antidote capable of saving the animals at the stage of secondary apnoea and in cardiac arrest for 2 minutes, if administered alongside oxygen therapy, artificial respiration, and cardiac massage (Paulet, 1958; Paulet and Le Bars, 1959).

4.5.6 Safety

Toxicology: List of toxic effects already reported

The LD_{50} of DCE ranged from 41 to 71 mg/kg in mice and rats by the i.v. or intraperitoneal routes (Evans, 1964).

Numerous adverse effects have been reported with DCE in non-poisoned and cyanide-poisoned patients. In line with the assumption of mutual antagonism of DCE and cyanide (Paulet et al, 1959), some authors reported that the effects were more frequent in non-cyanide poisoned rather than in cyanide-poisoned patients. However, later on the intrinsic effects of DCE were also reported in cyanide-poisoned humans.

The mechanism(s) of these adverse effects remain to be clarified. They occur frequently and it could be supposed that they are not of the truly allergic type but rather of the anaphylactoid type since certain cardiovascular manifestations can be experimentally reproduced in the dog (Riou et al, 1993). A relationship with the dose is not clearly apparent in the reported human cases. The existence of a protective effect of severe cyanide poisoning against the toxic effects of DCE, however, is not fully certain.

Free cobalt, in particular the cobalt (II) salts, are known to be toxic to the heart, liver, and kidneys (Speijers et al, 1982). The free cobalt in the preparation of Kelocyanor® has been measured; its concentration is on the order of 0.196 to 0.240 g/100 ml, while that of DCE is from 1.35-1.65 g/100 ml (Lipha). Clinical manifestations of cobalt toxicity include: gastrointestinal disturbances (nausea, vomiting at each administration when the dose is administered in divided doses); gastrointestinal haemorrhage, profuse diaphoresis; anginal pain/crushing chest pain; cardiac arrhythmias, including severe life-threatening ventricular arrhythmias; cardiovascular instability, hypotension as well as hypertension; dyspnoea, tachypnoea; decreased cerebral blood flow; nervousness, trembling convulsions; and anaphylactoid manifestations with a cutaneous rash, urticaria, angio-oedema affecting the face, the neck, and the larynx.

The experience of severe adverse effects related to DCE in the absence of cyanide poisoning was reported by a number of authors (Froneman, 1975; Nagler et al, 1978; McKiernan, 1980).

Comparisons of intrinsic effects of DCE compared to DMAP were assessed in non-cyanide poisoned anaesthetised dogs receiving either 10 mg/kg of DCE i.v. or a 3.25 mg/kg dose of DMAP hydrochloride (Klimmek et al, 1979b). Mean femoral arterial pressure increase by 20 mm Hg after DMAP while DCE induced a sharp decrease in mean arterial pressure to 80 mm Hg associated with a marked increase in heart rate. DMAP did not alter the venous central pressure (VCP) while DCE increased VCP of 90%. Blood flow in femoral vessels was mildly increased by DMAP while DCE induced a decrease of 40 to 50%. Both DMAP and DCE

increased the respiratory flow. DMAP did not induce any change in blood bicarbonate and base excess while DCE induced a marked decrease in both parameters. Cerebral blood flow increased of 30% following DMAP injection. In contrast, the injection of DCE was immediately followed by a decrease in cerebral blood flow.

In conclusion, a dose of DMAP resulting in at least 30% of MetHb levels did not cause cardiovascular alteration, while DCE induced significant and complex cardiovascular disturbances with a decrease in blood flow resulting from heart failure and metabolic acidosis.

Paulet observed that the acute toxicity of DCE in small animals is decreased by the presence of cyanide as well as glucose (Paulet, 1960a,b; 1961). The reduction of DCE toxicity by co-administration of glucose (mechanism uncertain) has also been advised by a number of authors (Beasley and Glass, 1998; Megarbane, 2003; Borron and Baud, 1996; Meredith, 1993) and subsequently led to its incorporation into the formulation. Meanwhile, hypoglycaemia was never reported as a toxic effect induced by or an adverse event of DCE. Therefore the administration of hypertonic solution was recommended to promote the transfer of the antidote to the heart (Paulet and Le Bars, 1959).

Contraindications

A few contraindications were pointed out by Borron and Lee (Toxicity of cyanide: www.emedicine.com/emerg/topic118.htm). These include known allergy to cobalt derivatives as absolute contraindication and an uncertainty regarding the definitive diagnosis of cyanide poisoning as a relative contraindication.

The indications for the administration of DCE was limited by Bryson as being a history of exposure and the presence of cyanide in the micro-environment as confirmed by Draeger tube, and the presence of features of severe cyanide poisoning, in particular, stupor, beginning and manifest coma (Bryson, 1987).

Adverse effects related to antidotes in cyanide poisoned humans

Cases of suspected cyanide poisoning were reported to the Health and Safety Executive (HSE) in the UK from 1970 to 1981. Of a total of 19 cases with a definite exposure to cyanide in 6 patients, 3 received DCE i.v. and presented with severe reactions with notable vomiting, hypotension, urticaria, angioneurotic oedema, and convulsions. Seven patients improved with AN or in the absence of antidotes. One improved with thiosulphate. From this experience the authors concluded that, in the occupational setting, the administration of DCE should only be considered in the minutes that follow the collapse of the patient (Aw and Bishop, 1981; Bryson, 1978; 1987).

4.5.7 Case series and casuistics of cyanide / cyanogenic poisonings treated with Dicobalt-EDTA

An overview of all cases in which DCE was used is presented in Table 23. This is further divided into cases that received DCE alone and cases that received DCE in combination with other antidotes in Tables 20 and 21 respectively, if the number of cases is high enough for statistical analysis.

The individual cases are described in Appendix C and the details were compiled into the sequential PSSa database (Appendix B) (Section 3.4.4). Relevant details are summarised in Table 21 below.

A total of 25 cases (19 male and 6 female) including suicidal (n=11) or accidental (n=13), intoxication with cyanide salts and chloroacylonitrile were reported (Bismuth et al, 1984). 7 patients were treated with DCE, while all with initially known cyanide poisoning received STS. The most common signs of the intoxication were respiratory arrest, coma, metabolic acidosis (n = 7), altered consciousness (n = 5), circulatory failure, cardiac arrest (n = 4) and convulsions (n = 3). Two victims were in a comatose state after cardio-respiratory arrest. Both were treated with DCE, HOCO and STS about 2 hours after admission (as DCE was mentioned at the first place and is generally considered being the most effective antidote of the 3 antidotes administered, it has probably been the 'main' antidote). Unfortunately the exact doses of the antidotes were not stated. CPR was performed in both cases an unknown time after intoxication so it is unclear whether the deaths occurred because of a delay in giving resuscitation and antidotal treatment, or because of a failure of antidote therapy itself. The cyanide-levels in the blood samples were 1.8mg/l and 6.8mg/l. Patient one could not be resuscitated, while patient two fell into a prolonged coma (after primarily successful CPR) and died six months later. A third patient was also found unconscious in respiratory arrest and with insufficient circulation. CPR was performed and the same antidotes were administered but without any improvement. No further assessment of the other cases treated with DCE is possible, so they could not be included in the tables.

Further cases came from case reports (see Appendix **X).**

Relevant details of cases treated with DCE alone or combined with STS and/or other antidotes are given in Table 20 and the following.

Table 20: Cases treated with Co-EDTA alone

Case			Antidotal Treatment	Number of treatment steps	PSSa		References
Age	Sex	Type of poisoning	Co-EDTA mg		Before	After	
68	M	NaCN	15x300	3+	3	4	Hillman et al. 1974
24	M	AgCN	1200	1	3	4	Brown et al. 1987
NS	M	CN salt	300	1	3	0	Naughton, 1974
7	F	Cassava	300	1	3	4	Brian, 1990
6	F	Cassava	300	1	2	0	Brian, 1990
7	F	Cassava	300	1	1	1	Brian, 1990
24	M	AgCN	600 + 300	2	3	4	Singh et al. 1989
61	M	Cyanide	2x 150	2	2	0	Bain and Knowles, 1967
42	M	HCN	2x300 mg	2	2	0	Nagler et at. 1978
NS	M	KCN	300 mg	1	1	1	McKiernan, 1980

Table 21: Cases treated with Co-EDTA and other antidotes

Case			Antidote treatment				Number of treatment steps	PSSa		References
Age	Sex	Type of poisoning	Cumulative dose of antidote (g)					Before	After	
			AN (pearls)	SN	Co-EDTA	HOCO				
With nitrites										
35	M	CN salt	11	-	0.6	-	1	3	0	Naughton, 1974
42	F	KAu(CN)$_2$	2	-	0.3	-	2	1	1	Wright and Vesey, 1986
19	M	Acetonitrile	-	-	0.6	4	2	3	4	Dequidt, 1974
NS	NS	CN	-	-	NS	NS	1	3	4	Bismuth, 1984
NS	NS	CN	-	-	NS	NS	1	3	3	Bismuth, 1984
NS	NS	CN	-	-	NS	NS	1	3	0	Bismuth, 1984

NS, not stated; -, not applied. *, coma worsened and patient died after 6 months.

Table 22: Cases treated ... .

Case			Antidote treatment						Number of treatment steps	PSSa initial and final		References
Age	Sex	Type of poisoning	Cumulative dose of antidote (g)									
			AN (pearls)	SN mg	DMAP mg	Co-EDTA mg	STS g	HOCO g		Before	After	
22	M	CN salt	2	300	-	600	12.5	-	2	2	0	Davis and Ewer, 1988
25	F	KCN	-	-	-	600	20	0.002	2	3	0	Hoang The Dan et al, 1981
43	M	CuCN	3	NS	-	600	NS	-	3	3	0	Dodds and McKnight, 1985
28	M	CN salt	-	-	-	300	23.6	-	2	3	0	Lundquist, 1992
42	F	KAu(CN)2	2	-	-	300	-	-	2	2	4	Wright and Vesey, 1986
50	M	KCN	2	-	-	300	2	4 in STS 8g	1	3	0	Yacoub et al, 1974
35	M	CN salt	11	-	-	600	-	-	2	2	0	Naughton, 1974
24	M	KCN	-	200	-	600	0.2	-	3	3	0	Paulet, 1965
32	M	KCN, Cu2CN	-	-	-	300	-	-	1	1	0	Paulet, 1965
NS	NS	NS	-	-	-	NS	-	NS	1?	3	4	Bismuth et al, 1984
NS	NS	NS	-	-	-	NS	-	NS	1?	3	3-4*	Bismuth et al, 1984
NS	NS	NS	-	-	-	NS	-	NS	1?	3	0	Bismuth et al, 1984
26	M	Acetonitrile	-	NS	-	NS	NS	NS	1	3	3-0**	Jaeger et al, 1977
19	M	Acetonitrile	-	-	-	600	-	4	2	3	4	Dequidt et al, 1974
28	M	KCN	-	-	-	600	-	4 in STS 8g	1	1	NS	Jourdan et al, 1993
26	F	KCN	-	-	250	300	6	-	2	2	0	Daunderer, 1974
		KCN										Lutier et al, 1971
		KCN										Lutier et al, 1971
43	M	HCN	1	-	-	300	-	-	2	1	0	Nagler et al, 1978
46	M	HCN	1	-	-	300	-	-	2	1	0	Nagler et al, 1978

NS, not stated; *, death after 6 months of coma; **, delayed recovery after 6 days of coma; ?, assumed since insufficient detail given in publication.

Occurrence of adverse effects in clinical use

No adverse effects have been recorded in six cases treated with Co-EDTA; but all patients had measurable levels of blood cyanide. The author concluded the intrinsic toxicity of Co-EDTA is neutralised by cyanide (Bryson, 1978). Other authors reported adverse effects of Co-EDTA in non-cyanide poisoned humans (McKiernan, 1980; Tyrer, 1981; Nagler and Provoost, 1978; Froneman, 1975). Later these findings and conclusions were challenged. In cyanide-poisoned individuals, too, mild to severe side-effects were seen, ranging from malaise and slight vomiting to facial, Quincke and laryngeal oedemas, anaphylaxis and crushing chest pain (Paulet, 1965; Bain and Knowles, 1967; Yacoub, 1974; Nagler, 1978; Dodds and McKnight, 1985; Wight and Vesey, 1986; Brian, 1990).

4.5.8 Assessment of efficacy and effectiveness in clinical use

Table 23: Overview of combination of Co-EDTA without and with other antidote(s) *

Total	Sequence	STS	AN	SN	HOCO	Co-EDTA	DMAP	AN + STS + SN	STS + HOCO	STS + SN	HOCO + STS + AN	HOCO + STS + SN
29	S1	1	2	0	3	**15**	1	1	1	1	2	2
10	S2	1	1	0	0	**8**	0	0	0	0	0	0
6	S3	0	0	0	0	5	0	0	0	0	0	1
1	S4	0	0	0	0	0	0	0	0	0	0	0
1	S5	0	0	0	0	1	0	0	0	0	0	0

* Emboldened data were used for analysis (n ≥ 7). One PSSa sequence in which Co-EDTA was used in combination with methylene blue did not figure in the table.

DCE alone: No statistical analysis possible.

DCE in combination with any antidotes:
Sequence 1   n = 16 p = 0.0528 ns   Median PSSa before: 3 – PSSa after: 1.5.

DCE and STS, other combinations not analysed (n < 7):
Sequence 1   n = 15 p = 0.94 ns   Median PSSa before: 2 – PSSa after: 2
Sequence 2   n = 8   Median PSSa before: 3 – PSSa after r: 3.

DCE in combination with any other antidote:

Sequence 1 n = 28   Median PSSa before: 2 – PSSa after: 2

Sequence 2 n = 10   Median PSSa before: 3 – PSSa after: 2.5.

Further evaluation was not possible due to low case numbers.

Global assessment of efficacy and effectiveness of antidote alone and in combination:

Data from the sequential PSSa study support the following assumptions:

DCE was used either alone or in combination with other antidotes to cyanide in 28 sequences allowing determination of PSSa before and after its administration. Twenty-six of the 28 sequences occurred from sequences one to three, suggesting Co-EDTA was used early during the course of cyanide poisonings.

Due to the limited data available it is not possible to draw any conclusions regarding the effectiveness of DCE other than it was never efficient when used alone in any of the sequences. It is possible, however, to conclude that when combined with other antidotes there was a trend towards the partial effectiveness of DCE in severe cyanide poisoning.

Although a mode of action for DCE has been hypothesised the chemical structure as well as the pharmacokinetics of the detoxification complex is lacking.

There is also no factual evidence for the effectiveness of DCE other than individual case reports.

The ideal dosing regimen of DCE in children is unknown, which makes its administration in children difficult (Borron and Lee: Toxicity of cyanide: www.emedicine.com/emerg/topic118.htm; Aaron, 1998).

4.5.9 Conclusion

While from a theoretical viewpoint, the advantages of DCE are the non-enzymatic mode of action, the potency of detoxification, rapid administration, and potential effectiveness in late stages of severe poisonings as observed in animal experiments, the drawbacks of DCE are the frequency and the severity of its side effects that occur in non-cyanide poisoned and cyanide-poisoned patients. These adverse events are qualitatively similar, occur frequently, and are of clinical relevance since they frequently resulted in the need for sustained clinical observation, medical intervention, and even supportive treatments.

Present clinical experience reported in the medical literature does not allow for a conclusion as to the efficacy of DCE when used alone in human cyanide poisonings of moderate grade (as assessed by the PSS). There is, however, a trend towards a partial effectiveness of DCE when used in combination with any other antidote in cyanide poisonings of severe grade (as assessed by the PSSa).

4.6 Hydroxocobalamin

4.6.1 Introduction

An antidotal effect of hydroxocobalamin (HOCO) against cyanide was first described in the 1950s and 1960s (Kaczka et al, 1950; Mushett et al, 1952; Friedberg et al, 1965) and later in combination with STS (Friedberg and Shukla, 1975). HOCO has been used for cyanide detoxification since the 1970s in France (Motin et al, 1970; Lutier et al, 1971, Jouglard et al, 1974; Yacoub et al, 1974). Initially HOCO was applied in combination with STS. More recently it was supplied dissolved in STS solution (4 g HOCO in 80 ml of 10% STS to be infused in at least 220 ml of 5% dextrose - Trousse Anticyanure, Laboratoire Anphar-Rolland) (Brouard et al, 1987; Hall and Rumack, 1987). Animal studies have shown an effect of the combination even if applied later in the course of cyanide poisoning (Paulet and Dassonville, 1985).

Efficacy of HOCO as cyanide antidote has been demonstrated *in vitro* (Tomoda, 1955; Delga et al, 1961a, 1961b; Rose et al, 1965; Mizoule 1966; Scheffer et al, 1970; Riou et al, 1989, 1990; Astier and Baud, 1996), and *in vivo* (Mushett et al, 1952; Friedberg and Shukla, 1975; Haguenoer et al, 1975; Posner, 1976a; Höbel et al, 1980; Mengel et al, 1989; Riou et al, 1991; Borron et al, 2006), though often in the context of SNP treatment. Studies, case series and casuistics in humans are addressed in detail below.

Low dosages of HOCO (500 mg every second day i.m.) have been advocated in humans for chronic cyanide poisonings like tobacco amblyopia (Bismuth et al, 1988).

Principally based on work of a French study group headed by Prof. FJ Baud in Paris, HOCO has been described and promoted as cyanide antidote worldwide since the 1990s, in particular for combined poisonings with CO in smoke inhalations (Bastigkeit, 2000; Becker, 1985; Borron and Baud, 1996; Brueske, 1997; Campbell, 2000; Dueras Laita and Nogue Xarau, 2000; Gonzales and Sabatini, 1989; Hall et al, 2007; Hantson et al, 1999; Hung et al, 2009; Imbert et al, 1993; Kulig, 1991; Kulling and Personne, 1997; Lipzak and Jensen, 1998; Montero Perez, 1998; O'Brien et al, 2011; Riou et al, 1992), but also as alternative antidote to STS for cyanide generated from nitroprusside or from nitriles, as well as for direct poisonings with HCN or cyanide salts (Meredith et al, 1993).

4.6.2   Identity

IUPAC name: Hydroxocobalamin
Chemical name: Coalpha-[alpha-(5,6-Dimethylbenzimidazolyl)]-cobeta-hydroxocobamide
CAS Registry number: 13422-51-0
Formula: $C_{62}H_{89}CoN_{13}O_{15}P$
Molecular weight: 1346.4
Chemical structure:
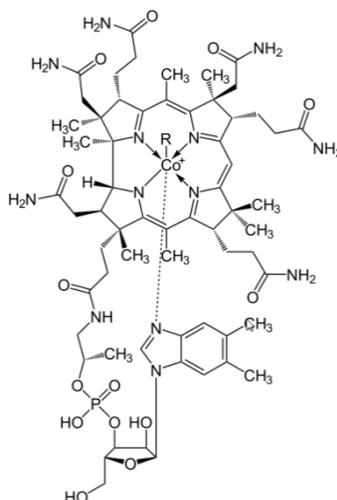
(Wikimedia, 2007)

Where R = -OH. Related compounds are- cyanocobalamin where R = $-CN$, methylcobalamin with R = $-CH_3$ and aquocobalamin where R = $-OH_2$.

4.6.3   Physico-chemical and pharmaceutical properties

Hydroxocobalamin, the hydroxylated active form of vitamin B_{12}, is a relatively large molecule in which a cobalt ion is coordinated in 4 positions by a tetrapyrrole ring. It is a hygroscopic, odourless, dark red crystalline powder that is freely soluble in water and ethanol, and practically insoluble in acetone and diethyl ether.

HOCO should be kept in tight, light-resistant containers and store in a cool place.

Cyanokit from Merck Sante (France) is a sterile, lyophilised powder for solution for i.v. infusion and contains 2.5 g of HOCO as active substance. Each vial of Cyanokit is recommended to be reconstituted with 100 ml of sterile saline (0.9% NaCl). Other diluents (i.e. Lactated Ringers Solution, 5% dextrose) have also been found to be compatible with HOCO. Cyanokit is currently available in a kit comprising two 2.5 g vials as the starting dose that totals 5 g HOCO. A new presentation contains the starting dose of 5 g HOCO in a single vial (Merck, 2010).

Incompatibility

Physical incompatibility (precipitate formation) was observed with the mixture of HOCO in solution and the following medicinal products: diazepam, dobutamine, dopamine, fentanyl, nitro-glycerine, pentobarbital, phenytoin sodium, propofol and thiopental. Chemical incompatibility was observed with STS, SN, and has been reported with ascorbic acid (EMA, 2013[a].

Consequently, these medicinal products must not be administered simultaneously through the same i.v. line as HOCO.

Simultaneous administration of HOCO and blood products (whole blood, packed red cells, platelet concentrate and fresh frozen plasma) through the same i.v. line is not recommended.

4.6.4 Practicality

Shelf life: 3 years.

Chemical and physical in-use stability of the reconstituted solution has been demonstrated for 6 hours at a temperature between 2°C and 40°C (104°F) (NIH, 2013). Any reconstituted product not used by 6 hours should be discarded.

Cyanokit should be stored below 25°C but not frozen.

Cyanokit may be exposed during short periods to the temperature variations of usual transport (15 days from 5°C to 40°C), transport in the desert (4 days from 5°C to 60°C) and freezing / defrosting cycles (15 days from -20°C to 40°C).

4.6.5   Pharmacokinetics and metabolism

Cobalamins, a complex group of molecules, are vitamins of the B_{12} series. Accordingly, physiological plasma concentrations of various cobalamins including HOCO and cyanocobalamin as well as their plasma transfer protein, trans-cobalamin, are always present.

Vitamin B_{12} depletion in plasma has been described from cyanide exposures e.g. from tobacco smoking, from cassava ingestion, but also in SNP application (Vesey et al, 1974).

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[a] EMA http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Product_Information/human/000806/WC500036429.pdf

However, the range of therapeutic dose used to treat pernicious anaemia, i.e. 1 mg (100-1,000 µg i.v. daily for two weeks, then weekly, and once Hb is stable monthly) (http://emedicine.medscape.com/article/204066-treatment#aw2aab6b6b2) is far below the range of dose of 5 to 10 g used to treat cyanide poisoning.

Studies have been done in naive animals, and human volunteers as well as patients poisoned by cyanide. Many studies refer to smoke inhalation cases (see below).

Pharmacokinetics in non-poisoned animals / healthy volunteers

Absorption

Owing to the recommended i.v. route of administration for antidotal use, the bioavailability is 100%.

Intraosseous application has been shown to be effective in goats with rapid onset of HOCO blood pressure effects (Borron et al, 2009).

Distribution

At low doses as used for vitamin B_{12} replacement HOCO is bound in plasma to trans-cobalamins. In particular, trans-cobalamin II is involved in rapid delivery to tissues. All cells have trans-cobalamin receptors and can internalise the transcobalamin-B_{12}-complex by active transport (Favier, 1992; Riou et al, 1992; Meredith et al, 1993; Herold, 2011). Only a fraction occurs as free HOCO (Linnell, 1987). HOCO allegedly also crosses the blood-brain barrier (Hall et al, 2007) although, considering the relatively large molecular weight and molecule size, this seems unlikely unless there is an active transport.

Storage is in the liver with a capacity of 2 mg total plus a further 2 mg in other tissues (Herold, 2011). Excretion from the liver is via the bile although there is some enterohepatic recycling. Part of the dose is excreted in urine, mostly within 8 hours (Reynold, ed, 1989).

No such data are available for high doses (4 to 5 g) as used in antidote (Riou et al, 1992; Riou et al, 1992).

Metabolism

Except for the substitution of hydroxyl-, H_2O-, methyl and adenosyl-groups attached to the cobalt-ion no metabolism has been described. Unless there is cyanide intoxication, high doses of HOCO are excreted unchanged (Favier, 1992).

Elimination

After parenteral injection most of HOCO in low doses is quickly excreted via the kidney in unchanged form (Reynold (ed.), 1989; Riou, 1992). However, data on the renal excretion are somewhat contradictory and range from 100% clearance within 24 hours to 42.1% and 32.5% retention at 3 and 28 days respectively (Hall and Rumack, 1986). Excretion of high antidotal doses is via the kidneys, obviously by glomerular filtration (Favier, 1992).

Pharmacokinetics in poisoned patients

The mechanism of detoxification is the immediate formation of a stable complex, cyanocobalamin, which results from the covalent binding of the cyanide ion to the Cobalt atom resulting in the release of a hydroxyl group (see below)

The formation of cyanocobalamin from HOCO is obviously a rapid process. Using rat cardiac papillary muscles exposed to 1 mM cyanide a complete recovery of mechanical parameters was reported after 10 minutes incubation in 1 mM HOCO in vitro (Astier and Baud, 1996).

Maximum measured plasma cyanocobalamin concentration reported ranged from 10 to 275 µmol/l (Astier and Baud, 1995). Data consistently suggested the greater the blood cyanide concentration, the greater the maximum measured plasma cyanocobalamin and the lower the maximum plasma HOCO (Astier and Baud, 1995; Houeto et al, 1995; Houeto et al, 1996).

The pharmacokinetics of cyanocobalamin at the doses corresponding to those resulting in neutralisation of cyanide has not been determined. The greatest reported cumulative amount of cyanocobalamin reported after the administration of a fixed 5 g dose of HOCO in humans was about 529 to 696 µmol corresponding to 0.717 and 0.944 g (Houeto, 1995a). The apparent volume of distribution, the total clearance as well as the renal clearance of cyanocobalamin at these doses and the corresponding plasma concentrations of cyanocobalamin still remain to be determined. The elimination half-lives of cyanocobalamin in plasma were reported to be 9.3 + 3.2 h (Astier and Baud, 1995) and 13.6 + 1.2 h (Houeto et al 1996).

In a patient severely poisoned by cyanide (blood level 128 µM) the elimination half-life was 19 hours, the apparent V_d was 0.15 l/kg (Astier and Baud, 1995). In heavy smokers the distribution half-life was found to be 2.83 hours (range 1.58-5.4) with an apparent V_d of 0.24 l/kg. The mean peak serum HOCO was 753 mg/l (range 359-1,286 mg/l) corresponding to 560 µmol/l (267 to 956 µmol/l) (Forsyth et al, 1993). In 11 smoke inhalation victims, 10 of whom had cyanide in blood, distribution half-life was 1.86 hours, elimination half-life was found to be 26.2 hours with an apparent V_d of 0.45 l/kg (Houeto et al, 1996). Other authors reported a short plasma half-life (5 min), and rapid redistribution and excretion (Cottrell et al, 1982; Vessey and Cole, 1981).

Total cobalt from HOCO and cyanocobalamin in 3 patients with suspected cyanide poisoning had an elimination half-life of 18.9 hours (mean) and a volume of distribution (V_d) of 0.15 l/kg (mean) (Chappey et al, undated).

In vitro a probable penetration of HOCO into heavily cyanide loaded cells has been reported in human fibroblasts after cyanide exposure (Astier and Baud, 1996). A study in healthy heavy smokers reported that in the group having received the combination of HOCO + STS, the amount of cyanide recovered as urinary cyanocobalamin was 25.5% while it reached 65.0% of the estimated total body cyanide burden in the volunteers having received HOCO alone (Forsyth, 1993).

4.6.6 Mode of action, dosage and time of onset

Primary mode of action

The mode of action is the non-enzymatic covalent binding of cyanide to HOCO resulting in the formation of cyanocobalamin, first described in the 1950s (Kaczka et al, 1950; Mushett et al, 1952). Therefore, its antidotal effect is merely dependent on the molar ratio of cobalt to cyanide (Way, 1984).

However, the capacity of a fixed dose of HOCO to detoxify cyanide is limited. Laboratory measurements on the detection of HOCO and cyanocobalamin in humans with cyanide intoxication report on 7 cases with cyanide levels of 0.4 to 6.76 mg/l (0.4 to 6.76 for ingestion of cyanide salts and 0.57 to 5.4 for smoke inhalations). The patients all received a fixed 5 g dose of HOCO and probably survived, as they are referred to as 'patients', not as 'victims' (Astier and Baud, 1995). Yet the data on cyanocobalamin show, that there is no significant difference between the cyanocobalamin levels for initial cyanide concentrations of 3.2 and 6.76 mg/l (cyanocobalamin 215 and 275 µM), which indicates that there is a limited capacity of HOCO to bind cyanide. HOCO has a high molecular weight (MW = 1,346.4). Owing to the stoichiometry of the reaction, HOCO will bind only 1 mol of HCN per mol of HOCO. Therefore, the detoxification (binding) of a single lethal dose of cyanide (1 mg/kgbw) requires at least 50 mg HOCO/kgbw (Eyer, 1994), as 1346.46 g/mole of HOCO is needed to bind 27 g/mole of HCN. Generally speaking, 5 g of HOCO can bind 100 mg of cyanide (Forsyth, 1993).

Thus, HOCO must be administered in a dose equimolar to cyanide (Evans, 1964; Friedberg et al, 1965). This recommendation was further supported by an *in vitro* study on a model of human fibroblast showing that after incubation with equimolar concentration of HOCO, the intracellular free cyanide pool was markedly decreased but not abolished since a mean residual amount of about 27% in comparison with controls was noted. However, this *in vitro* study was not designed to assess the reversal of cellular toxic effects (Astier and Baud, 1996).

Conversely, a model of nitroprusside-induced cyanide poisoning in dogs showed that the very small HOCO dose equivalent to one mole of HOCO per 25 moles of cyanide released by SNP, was unable to decrease red cell cyanide concentrations. There was no effect of this dose of HOCO on normalisation of blood pressure and base deficit. Surprisingly, even such a low dose partially reduced the increase in nitroprusside-induced increase in blood lactate. These findings at this dose of HOCO let the authors to conclude that STS is the antidote of choice in nitroprusside-induced cyanide poisoning while low dose of hydroxocobalamin should be considered in addition to STS when facing severe lactic acidosis induced by SNP (Krapez et al, 1981).

Consequently, it has been proposed that a single dose of 5 g of HOCO in humans can detoxify only 40 µmol/l or about 1 mg/l of cyanide in blood (Houeto et al, 1995a) although this has been challenged (Demedts et al, 1995) and the need for confirmation *in vivo* has been recognised (Houeto et al, 1995b).

The mixture of HOCO and STS was shown to be less effective than STS alone in rabbits, but not in mice (Evans, 1964). This might be due to the formation of thio-sulphato-cobalamin, which is unable to, or only less firmly, fix cyanide. Therefore, combined injection of HOCO and STS within the same vial must be avoided (Friedberg and Shukla, 1975). A further paper reported no synergistic effect of a therapeutic dose of STS in combination with a low dose of HOCO in a model of nitroprusside-induced cyanide poisoning in dogs (Krapez et al, 1981).

In contrast, other authors report a potentiation or synergism of HOCO-effect by 400 mg/kg STS in animals experiments. All animals died after 9 mg/kg cyanide and 225 mg/kg HOCO (half equimolar), while all survived with the addition of STS as above. This synergism was seen independent of whether HOCO and STS were applied together or separately (Delga et al, 1961a&b; Mizoule, 1966). This may be due to the fact that the antidotal effects of the two substances may appear at different points in time i.e.– initial and immediate effect of HOCO, followed by the somewhat delayed, but more persistent effect of STS (Riou, 1992).

It has been suggested that cyanide initially binds to HOCO and is released before being detoxified by rhodanese and excreted as thiocyanate. The regenerated HOCO is said to be available to rebind cyanide (Hall and Rumack, 1987). However, given that the reaction between HOCO and cyanide has been described as irreversible (Favier, 1992), and cyanobalamin is not known to break up spontaneously, this hypothesis remains doubtful, and is unsupported by the most recent data.

The exact role of cyanocobalamin in the metabolism of SNP is unclear (Drew, 1983). Cyanocobalamin has been postulated to function as a cofactor for detoxification by functioning as a rhodanese system cofactor (Tinker and Michenfelder, 1976). This was the basis for advocating

the use of HOCO only in combination with STS. However, recent cumulative experience with HOCO used alone in various forms of cyanide poisonings showed this hypothesis to be untrue.

In any case, it is suggested to separate the application of the two antidotes by at least a minute (Friedberg and Shukla, 1975).

Onset of the effect

In an *in vitro* study using a model of human fibroblasts showing that after incubation with equimolar concentration of HOCO, the mean intracellular to extracellular cyanocobalamin concentration ratio was about 158. Moreover, the authors were unable to find any measurable free intracellular HOCO (quantification threshold: 0.05 µM). These finding suggested, at least in this model of human fibroblasts, that the initial event was penetration of HOCO into cells followed by a rapid formation of non-toxic cyanocobalamin. These data support the hypothesis of an antidotal effect of HOCO appearing without any delay in this experimental model in which there is no barrier between extracellular HOCO and intracellular cyanide (Astier and Baud, 1996).

The rapidity of action of HOCO is further supported by an *in vitro* study on cyanide-poisoned rat cardiac papillary muscle. In rat cardiac papillary muscle poisoned with 1 mM of NaCN, an equimolar 1 mM of HOCO induced a beat to beat disappearance of cyanide-induced toxic effects resulting in an almost complete recovery of muscle functions 5 minutes after addition of HOCO (Riou et al, 1990).

A model of cyanide-poisoned guinea-pigs showed the immediate i.v. administration of an equimolar dose of HOCO after cyanide did not prevent the cyanide-induced alteration in respiratory pattern which, however, resumed to regular respiration within 3 minutes after HOCO injection (Posner et al, 1976).

A model of cyanide-poisoned dog showed that HOCO infusion was associated with rapid, dose-related cardiovascular recovery. Mean arterial blood pressure increased beginning within 1 to 3 minutes of initiation of HOCO infusion. The delay in onset of the therapeutic was dependent of the dose while the duration of infusion was unchanged (Borron et al, 2006).

The onset of effect has been described in humans to be rapid, too, which is in agreement with the above studies. Other case reports seem to indicate a somewhat delayed effect (Benaissa et al, 1995).

4.6.7 Safety

Toxicology: List of toxic effects already reported

An LD_{50} of HOCO has not been reported. Single i.v. doses of 540 mg/kg in dogs, 1,000 mg/kg in rats and guinea pigs have not prompted toxic effects (Posner et al, 1976); nor did the application of 1600 mg/kg cyanocobalamin to mice (Winter and Mushett, 1950).

Some histological alterations have been reported in liver, heart and kidney, though, in rabbits dosed with 177, 221 and 354 mg/kg/h relating to molar ratios SNP/HOCO of 1:4 to 1:8. In detail, toxic hepatosis with loss of trabecular structure, swollen hepatocytes, and hypoplastic Kupffer cells in the liver, myocardial swelling, some cell fragmentation and increased granulation of the cytoplasm in the heart were seen, while kidney changes were reported to be slight and non-specific only (Höbel, 1980).

While hemodynamic effects have only been seen at supra-therapeutic doses of 140 mg/kg, but not at therapeutic doses of 20 and 70 mg/kg, in the form of moderate negative inotropy associated with primary arterial vasoconstriction (Riou et al, 1992), a transient (30 minutes) trend of increasing blood pressure with concomitant heart rate decrease were seen after application of 5 g HOCO in heavy smokers (Forsyth et al, 1993). The slight increase in blood pressure, and also in systemic resistance, was confirmed by other authors in an experimental model of conscious dogs receiving a single i.v. dose of 70 mg/kg of HOCO (Riou et al, 1993). It has been hypothesised from animal experiments that scavenging of nitric oxide by HOCO may be causal for this (Gerth et al, 2006).

It has been discussed that in circulatory failure due to cyanide poisoning, this may actually be a desirable effect (Cescon and Juulink, 2009).

Toxicology: Toxic effects noticed in safety evaluation of cases

No further toxic effects were identified.

Adverse effects related to antidotes

Allergic reactions up to anaphylactic shock to HOCO given for vitamin substitution (dose of e.g. 1 mg) have been described (Hames and Warin, 1971; Auzepy et al, 1974; Dally and Gaultier, 1976), which have been due to impurities (Riou et al, 1992). In the human cases reported for antidotal use, there is one description of a type 1 allergy with urticarial and Quincke oedema. This patient, however, had received STS, AN, and Co-EDTA before and STS in parallel

to HOCO (Yacoub, 1974). In a randomised, double-blind, placebo-controlled study in 136 healthy volunteers with doses of 2.5, 5, 7.5 and 10 g HOCO vs. placebo over 7.5 to 30 minutes two allergic reactions were seen in the 102 volunteers in the groups (at 5 and at 10 g), in one case with itching, facial erythema, swelling and reddening of one eye and shivers, in the other with dyspnoea, facial oedema, exanthema and urticaria with weals. These could be managed with dexamethasone and/or dimethindene maleate (Uhl et al, 2006).

A transient pink discoloration of mucous membranes and urine was noted both in healthy volunteers (Uhl eet al. 2006) and in patients receiving a dose as low as 87.5 to 100 mg of HOCO; (Cottrell et al, 1976) whereas as dark red urine for was observed for up to 48 hours after application of 5 g HOCO in heavy smokers. No alterations of laboratory values of clinical significance were observed (Cottrell et al, 1976; Forsyth et al, 1993). The chromaturia and skin discoloration described repeatedly (Favier et al, 1993; Borron et al, 2001; Cescon and Juulink, 2009) do not have adverse consequences (Riou et al, 1992; Uhl et al, 2006) and disappear within 2 to 3 days (Borron, 2007a), or 7 days (Borron et al, 2001) although they may disturb some clinical laboratory measurement that rely on colorimetric analysis (Curry et al, 1994; Gourlain et al, 1994; 1996; Vest et al, 2002; Weng et al, 2004; Lee et al, 2007; Ednninghoff et al, 2008; Pamidi et al, 2009).

The plasma level of cyanide as measured by chrommetric assay with micro-diffusion was found to be higher than before HOCO application in one case. The effect was not seen in whole blood and must be regarded as artificial (Weng et al, 2004). In one other case a haemodialysis machine could not be made to run, indicating a 'blood leak'. This was due to a reaction of the photo-detector in the dialysate column reacting to the discoloration of body fluids by HOCO (Sutter et al, 2010). In the same year a pseudo-haematuria from HOCO was described in a severely traumatised patient having received HOCO for suspected additional cyanide poisoning. The authors point out that this side effect may prompt additional diagnostic measures to rule out urinary tract damage (Torremadé Barreda et al, 2010).

A randomised, double-blind, placebo-controlled study in136 healthy volunteers with doses of 2.5, 5, 7.5 and 10 g HOCO over 7.5 to 30 minutes found besides the effects mentioned above papular to pustular rash, headache, erythema at the injection site, decrease of lymphocyte percentage, nausea, pruritus, chest discomfort and dysphagia. Blood pressure increases was additionally seen in some volunteers with a systolic pressure rising by up to 27 mm Hg and returning to baseline within 4 hours (Uhl et al, 2006).

4.6.8 Case series and casuistics of cyanide / cyanogenic poisonings treated with hydroxocobalamin

Regarding acute poisonings there are several case series, which are difficult to assess, as they were published by a single centre in Paris. Since some cases seem to be overlapping (repeat citation) in several publications it is not possible to simply add up the cases reported. However, there is no overlap between cases reported by Bismuth (combination of HOCO and STS used) and by Baud (only HOCO used). Overlap for fire smoke cases of Baud (1991) and Borron (2006) was also limited. Regarding fire victims reported by the Paris Fire Brigade alone, blood cyanide levels were never reported (Fortin et al, 2006). Therefore, it is impossible to assess the degree of overlap with Baud and Borron's studies (Personal communication Baud).

Cyanide and cyanogen cases

Publications from Paris reported on 3 cases of ingestion and up to 7 cases of inhalation with cyanide levels ranging from 217 to 256 µmol/l (5.56 to 6.56 mg/l) that received 5 to 10 g of HOCO obviously survived. However, two of these inhalation patients did not have cyanide poisoning. More details are not given, as the aim of the papers was to show the formation and level of cyanocobalamin from HOCO in the presence of cyanide (Baud et al, 1990; Bismuth, 1992; Imbert et al, 1993).

In 1996, 36 cases of inhalation or ingestion of cyanide or cyanogens were reported, excluding smoke inhalations. Lactic acidosis was the most consistent symptom; bitter almond smell was often not detected. Of the 36 patients eight were asymptomatic, while the other 28 showed metabolic acidosis (n = 18), Coma (n = 13), cardiovascular collapse (n = 10), respiratory arrest (n = 8), convulsions (n = 6), psychomotor slowing (n = 6). Five died with post-anoxic coma. For HOCO it is stated by the authors that it had simplified considerably the resuscitation of such patients. To this goal, 18 patients are reported from literature having received HOCO at doses from 200 mg to 7 g, of which only one received HOCO alone, 17 the HOCO/STS mixture, 2 additional STS, 6 also Co-EDTA, and one each Co-EDTA, SN and STS, and Co-EDTA and SN. Doses for the other antidotes were not specified (Borron and Baud, 1996).

11 patients (8 males and 3 females; 14 – 63 y) with a history consistent with cyanide poisoning were treated in addition to supportive treatment including gastric lavage in cases of ingestion, with specific antidotal treatment. HOCO was the initial antidote used in these patients. STS was also administered by continuous intravenous infusion at the discretion of the attending physician. There was a significant correlation between plasma lactate and blood cyanide concentrations (r = 0.74, p = 0.017). During the course of cyanide poisonings, a plasma lactate concentration of ≤ 72 mg/dl (8 mmol) was sensitive (94%) and moderately specific (70%) for a toxic blood cyanide concentration (≥ 1.0 mg/l). The authors deemed the immediate and serial measurement

of plasma lactate concentrations to be useful in assessing the severity of cyanide poisoning. No evaluation of the efficacy of HOCO is possible (Baud et al, 1996).

Eleven cases hospitalised between 1998 and 1999, of which 10 were due to ingestion and 1 to inhalation, are described in another publication by this working group. Among 8 males and 3 females, mean age 38 years, 10 had cyanide and lactate levels measured before treatment. The blood cyanide levels ranged from 0.3 to 6.9 mg/l with a mean of 4.1 mg/l. Plasma lactate was in the range of 46 to 477 mg/dl. Five patients were apnoeic and 5 had a GCS of 3 upon arrival. All patients received between 5 and 20 g HOCO by infusion. Two patients received in addition 16 mg STS, 1 BAL 200 mg and DMSA 400 mg for associated mercury poisoning.

Seven out of 11 patients survived to hospital discharge. Four patients, who did not survive, all had a GCS of 3 and apnoea on arrival and had a trend toward longer treatment delay (Borron et al, 2006).

In 2007, a further publication reports on 14 hospitalised patients with cyanide poisoning between 1988 and 2003. The 11 cases as of 2006 are probably included in part. Now there were 12 male and 2 female cases with a median age of 35.2 years. In 10 cases the poisoning was due to KCN ingestion, and in one case each to mercuric cyanide ingestion, cyanogen bromide inhalation, acetonitrile ingestion, while in one case the source was unknown. Blood cyanide concentrations ranged from 0 to 260 µmol/l (about 6.7 mg/l), in 2 patients it was not measured and in 1 it was low. Eleven out of 12 patients with blood cyanide measurements were above the typically lethal threshold of 2.6 mg/l. Two patients were found in cardiac arrest, 4 in shock, 1 with respiratory arrest setting in, and 5 with severe neurological impairment (GCS at or below 8). Two more patients suffered cardiac arrest during transport. HOCO was given at 5 to 20 g with a mean of 3.1 hours after cyanide exposure. Four patients received STS in addition, and 1 STS and Co-EDTA. In the mercuric cyanide case BAL and DMSA were given. Oxygen (n = 11), resuscitation (n = 2), catecholamines (n = 8) and mechanical ventilation (n = 7) were applied as supportive therapy.

Ten out of 14 patients survived to discharge, 4 died in hospital (intensive care unit) as a result of cardiac or respiratory arrest. Mean time to death was 6.3 days. One surviving patient was found with cardiac arrest 12 hours after the onset of intoxication. He had post-anoxic encephalopathy. Seven out of 9 patients treated with HOCO alone survived. Cyanide level in these 7 survivors ranged from 13 to 216 µmol/l (ca. 0.3 to 5.5 mg/l). The 2 patients who did not survive had a level above 100 µmol/l. Eight patients had one or several adverse events – chromaturia (n = 5), pink to red skin discoloration (n = 3), increase in heart rate (n = 1), and elevated blood pressure (n = 1) (Borron et al, 2007a).

The correlation between the severity of the intoxication and the outcome can be described as follows: 8 mild (n=3, HOCO alone) to moderate (n = 5, 2 HOCO and STS, 3 HOCO alone) survived.

Five severe intoxications (cardiac or cardio-circulatory failure; n = 4) or low GCS (score 3; n = 1) were treated with HOCO/STS (n = 4), or HOCO/STS/Co-EDTA (n = 1). Four of these patients died, one survived with persistent encephalopathy. Thus in severe cases HOCO was not really effective.

Cyanide from smoke inhalation

A first paper from Paris in 1991 dealt with cyanide concentrations in blood of smoke inhalation victims. 109 cases, 50 women, and 59 men, aged 2 to 87 years, were included. 36 patients of these were found dead at the fire scene, 7 more died in hospital. The mean blood cyanide in all 109 patients was 59 µmol/l (ca. 1.5 mg/l), in the fatalities 116.4 µmol/l (ca. 3 mg/l) and in the survivors 21.6 µmol/l (ca. 0.55 mg/l). HOCO was introduced into treatment of fire smoke victims, but was restricted to cases with severe cardiovascular compromise. The dose was not specified. Obviously 5 cases with cardiac arrest were treated with "high doses of hydroxocobalamin", yet they all died. Also 11 out of 39 patients supposed to have had smoke inhalation had received HOCO. Whether these 2 HOCO groups overlap, is not obvious (Baud et al, 1991).

A report and a thesis related to 50 patients from 1987 to 1992 (28 women, 22 men, mean age 54 years), who received HOCO on site of smoke inhalation accidents. Cyanide poisoning was assumed on the basis of soot in mouth or expectoration plus disturbed consciousness. Five grams of HOCO (in case of cardiac arrest 10 g) were applied. Seventeen patients retrospectively without cyanide intoxication tolerated the HOCO well and without effect on blood pressure. In 33 patients with cyanide poisoning it led to a significant increase in blood pressure. However, 19 out of 33 died. The mean cyanide concentration for all 50 patients was reported to be 84.4 µmol/l (2.16 mg/l), with no significant difference between survivors (75.5 µmol/l – 1.94 mg/l) and fatalities (98.9 µmol/l - 2.54 mg/l). The GCS was not significantly different between start and end of HOCO infusion, but it was between start of infusion and 1 hour after end of infusions (5.6 and 7 respectively). The only adverse effect attributable to HOCO was pinkish skin discoloration in all patients (Favier, 1992; Favier et al, 1993).

From the same working group there is a report on 12 smoke inhalation victims, who all received HOCO within the first 30 minutes. Seven survived (cyanide levels 0 to 29 µmol/l [0 - 0.74 mg/l], age 24 – 62, mean 44.3 years), 2 with low cyanide levels died (cyanide levels 25 and 27 µmol/l [0.64 and 0.69 mg/l], age 78 and 94 years). Only 3 of these patients had cyanide levels above 40 µmol/l, age 83, 89 and 24 years, and these 3 all died. The data for all 5 fatalities are cyanide 25-135 µmol/l (0.64 – 3.5 mg/l; age 24-94, mean 72.6 years). Deaths were due to respiratory or

CNS complications (Houeto et al, 1995a). A possible conclusion would be that the surviving patients did not have critical cyanide poisonings, and that patients with critical cyanide levels died despite HOCO administration. In addition it could be said, that elder patients are at much higher risk.

Eleven of these 12 patients were presented again in a later paper. All received 5 g HOCO, one was excluded as cyanide was below limit of detection. Of the remaining ten 7 survived, and 3 died. No new data on these patients are presented (Houeto et al, 1996).

A congress abstract describes 63 patients, of which 42 were intoxicated by cyanide (CN level in blood 1.3 – 3.7 mg/l; median 2.5). All received HOCO at a dose of 5 g (10 g in case of cardiovascular collapse). Fourteen patients died, 9 from decerebration, 4 from septic shock, and 1 from hypoxemic pneumopathy. (Borron et al, 2001).

Six years later in a prospective study 69 patients (36 female, 33 male, mean age 44 years) with smoke inhalation and suspected cyanide poisoning (soot in face, mouth or expectorations and neurologic impairment) seen between 1987 and 1994 were described. Pre-hospital fatalities were excluded. In 63 patients pre-treatment blood cyanide levels could be measured, of which 42 had levels above 39 µmol/l (1 mg/l) and 21 had levels below that, thus no significant cyanide poisoning. Thirteen patients had cardiac arrest, and cyanide level median was 123 µmol/l (3.15 mg/l). Fifty-seven out of 69 patients had carbon monoxide poisoning (Borron et al., 2007). HOCO was given at a dose of 5 g (maximum 15 g) at the fire scene or in the hospital (intensive care unit) to all 69 patients. Additional treatments consisted in hyperbaric oxygen therapy in 57 of the 69 patients. Fifty out of 69 (72%) survived. The causes of death in the fatalities were brain death (n = 13, all initial cardiac arrest), and infectious complications (n = 6).

Twenty-eight out of 42 patients with confirmed cyanide poisoning survived, among these 17 out of 23 with levels between 1 and 2.54 mg/l, and 8 out of 13 with higher levels. Three out of 6 patients with at least twice the potentially lethal level (> 5 mg/l) also survived. Two out of 15 patients with initial cardiac arrest survived. (Note: Their data are reported in Baud and Borron, 2008).

In the 41 out of 42 cyanide poisoned patients neurological impairment was initially present, of 28 survivors 21 recovered fully, sequelae persisted in 6. Of the 66 patients with initial neurologic impairment 47 (71%) survived.

Regarding adverse effects chromaturia (n = 6), pink or red skin discoloration (n = 4), hypertension (n = 3), erythema (n = 2) and increased blood pressure (n = 2) were seen. There was no allergy (Borron et al, 2007b).

This paper was met with some critique discussing the low incidence of skin discoloration reported (6%), compared to volunteer studies (94 – 100%) with the idea that this might indicate underreporting or underrecognition of adverse events. However, this author, too, agrees to consideration of HOCO for smoke inhalation patients with coma, cardiac arrest, or cardiovascular extremis (Erdmann, 2007). The authors replied by stating that skin discoloration had not been included in the questionnaires, as being regarded as a benign reaction (Borron and Baud, 2008).

One probably related paper reports on 101 patients (53 males, 48 females, mean age 47.1 years) treated with HOCO during 8 years (1995-2003) by the Paris Fire Brigade nurses under emergency physician supervision. The HOCO doses given ranged from 1 to 10 g with a mean of 5 g and were applied about 14 minutes after initiation of emergency care, which again started 2 to 18 minutes after the emergency call.

Thirty out of 101 patients survived, 42 died, of which 17 at the fire scene and 25 in hospital (intensive care unit), and for 29 the survival status is unknown. Of 38 patients found in cardiac arrest, 21 had a return of spontaneous circulation, but 19 out of 21 died in hospital (intensive care unit). Five were found in shock. All in all 12 patients were regarded as haemodynamically unstable due to systolic blood pressure below 90 mm Hg. In 9 out of 12 improvement was observed with recovery of systolic blood pressure after 30.6 (5 to 70) minutes after start of administration. For 52 patients the GCS could be assessed. Ten out of 52 improved, 41 out of 52 had no change and 2 deteriorated. Only 2 adverse effects were observed; pink to red urine in 5 and cutaneous rash in 1 case (Fortin et al, 2006). Unfortunately, blood cyanide concentrations were never reported in the included patients.

A very recent study compared 25 patients with smoke inhalation with 12 patients with CO-inhalation from other sources. Twelve of the patients with smoke inhalation received HOCO prior to HBO-treatment. In the control group (CO-intoxications without smoke exposure) only one patient received HOCO. The mean age was 52 years, 12 male and 18 female patients were involved. The time elapsed between exposure and admission to the hospital was on average 3 hours. The results were as follows: In group I, 2 patients had an elevated cyanide level in blood higher than 39 µmol/l corresponding to 1 mg/l. One of the patients had received HOCO before blood was taken for cyanide measurement. In the group that received HOCO the cyanide level in blood prior to hyperbaric oxygen treatment was on average 15.4 µmol/l (0.42 mg/l), in the group without antidotal therapy 14.35 µmol (0.39 mg/l). Under hyperbaric oxygen therapy in 13 patients the cyanide level increased whereas in 10 patients decreased. In two cases the level did not change. The change in cyanide levels under hyperbaric oxygen treatment was very small in both groups. On average it was 1.9 µmol = 0.051 mg/l in the treated group and 2.1 µmol/l (0.057 mg/l) in the control group. The influence of the hyperbaric oxygen treatment could be seen in a decrease in lactate in blood from 5.0 µmol/l to 1.9 µmol/l in the antidote treated group and from 3.7 µmol to 1.3 µmol in the controls. In the group of CO-poisoning no patient had an

elevated blood cyanide level (Lawson-Smith et al, 2010). The severity of the different poisonings remains unclear. The involvement of cyanide in the poisonings was quite limited.

Two patients had cyanide blood levels above 1 mg/l, and are discussed in the casuistics appendix and included in the cases table.

The cases reported in the literature with HOCO are summarised in tables 28 to 30. For detailed descriptions of these casuistics please refer to Appendix C.

One case merits consideration: An indirect case of cyanide poisoning by propionitrile was reported (Baud et al, 1986; Bismuth et al, 1987). The maximum cyanide level was 5.71 mg/l, the French mixture of HOCO and STS was administered, resulting in a cyanide level of 0.93 mg/l one hour later. Urinary thiocyanate excretion increased sharply from 0 to 21.1 mg/l. The drop of cyanide in blood cannot be explained by an effect of HOCO. At maximum the HOCO dose of 4 g would have been able to reduce the cyanide level by 0.8 mg/l (Houeto et al, 1995a). The sharp increase of thiocyanate in urine suggests a decisive effect of thiosulphate in this case.

Table 24: Overview of combination of Hydroxocobalamin (HOCO) with and without other antidote(s)

	Total	Sequence	STS	AN	SN	HOCO	Co-EDTA	DMAP	Methylene blue	BAL	Coramine	STS + AN + Co-EDTA	STS + SN + Co-EDTA	STS + SN	STS + BAL
HOCO	**31**	S1	4	0	0	**19**	3	0	0	1	0	2	2	0	0
	15	S2	3	0	0	**10**	0	0	0	0	0	0	0	1	1
	5	S3	1	0	0	3	0	0	0	0	0	0	1	0	0
	1	S4	0	0	0	1	0	0	0	0	0	0	0	0	0
	0	S5	0	0	0	0	0	0	0	0	0	0	0	0	0

Data used for analysis ($n \geq 7$) are in bold.

Table 25: Cases treated with HOCO alone (Appendix C)

| Case | | | Antidote treatment | | | PSSa | | |
Age	Sex	Type of poisoning	Cumulative dose of HOCO (g)	Number of treatment steps		Before	After	Reference
39	M	NS	5	1		3	0	Baud, 1980
NS	NS	Cassava	0.5	1		3	0	Espinoza et al, 1992
NS	NS	Cassava	0.5	1		3	0	Espinoza et al, 1982
NS	NS	Cassava	0.5	1		3	0	Espinoza et al, 1982
NS	NS	Cassava	0.5	1		3	0	Espinoza et al, 1982
33	M	NS	8	1		3	0	Galliard, 1991
63	M	KCN	10	1		3	2	Baud, 2001
38	M	KCN	10	1		3	3	Baud, 2001
14	M	Hg(CN)	5	1		3	0	Baud, 2001
52	M	KCN	5	1		2	0	Baud, 2001
26	M	KCN	15	2		3	4	Baud, 2001
32	M	KCN	10	2		3	0	Baud, 2001
53	F	AuCN, KCN	5	1		1	0	Baud, 2001
38	M	BrCN	5	1		1	0	Baud, 2001
1.5	M	SNP	NS	1		3	0	Ballesteros, 2003
51	M	KCN	5	1		3	0	Weng, 2004
68	F	Amygdalin	5	1		3	0	Bromley, 2005
50	M	KCN	5	1		3	0	Coentrão, 2010
32	F	KCN	5	2		3	0	Hung, 2009
29	M	Acetone cyanohydrin	2.5	1		0	0	Roedelsperger, 2009
NS	M	Acetone cyanohydrin	2.5	1		0	0	Roedelsperger, 2009
53	M	Acetone cyanohydrin, HCN	2.5	1		0	0	Roedelsperger, 2009
22	M	NaCN	5	1		2	0	Baud, unpublished
51	F	Amygdalin	5	1		3	0	Martinelli, 2008
39	M	Acetonitrile	5	1		2	0	Baud, unpublished

NS, not stated.

Some cases treated with HOCO in combination with STS (5 cases) and/or other antidotes, see Table 26 excerpted from Table 14. HOCO has not been applied uniquely with nitrites, DMAP or Co-EDTA.

Table 26: Cases treated with HOCO in combination with other antidotes (Appendix C)

Case			Antidote treatment								Reference
			Cumulative dose of antidote (g)					Number of	PSSa		
Age	Sex	Type of poisoning	AN (pearls)	SN	STS	Co-EDTA	HOCO	treatment steps	Before	After	
With STS											
3	M	Propionitrile	-	-	8	-	4	1	2	0	Baud et al, 1986 Bismuth et al, 1987
15	F	KCN	-	-	8	-	4	1	3	1	Tassan, 1990
28	M	KCN	-	-	8	-	4	1	3	4	Jamali, 1993
27	F	Mercury	-	-	8	-	4	1	3	4	Benaissa, 1995
28	F	KCN	-	-	16	-	5	2	2	0	Baud, 2001
44	M	KCN	-	-	8	-	9	2	3	0	Baud, 2001
54	F	NaCN	-	-	8	-	4	1	3	0	Harry, 1985
With nitrites and STS											
23	M	KCN	-	0.5	75	-	0.03	1	3	4	Buchanan, 1976
With STS and Co-EDTA											
NS	M	KCN	-	-	NS	0.3	4	2	2	0	Lutier, 1971
26	M	Acetonitrile	-	-	NS	NS	NS	1	3	0	Jaeger, 1992
30	F	CN salt	-	-	16	NS	15	2	3	4	Baud, 2001
With nitrites, STS and Co-EDTA											
23	M	KCN	-	-	NS	0.3	4	1	3	0	Lutier, 1971
NS	M	KCN	-	NS	NS	0.6	4	1	2	0	Lutier, 1971
NS	NS	KCN	NS	-	NS	0.6	4	1	2	0	Lutier, 1971

NS, not stated; -, not applied.

4.6.9 Occurrence of adverse effects in clinical use

In the case series described above the only side effects were were skin and urine discoloration, which was insufficiently documented, however, some cases of elevated blood pressure and one skin rash. This spectrum fits to the known side effects of HOCO.

4.6.10 Assessment of efficacy and effectiveness in clinical use

Animal experiments suggested HOCO was inferior to STS in the treatment of SNP induced cyanide toxicity (Krapetz et al, 1981; Hewick et al, 1978) or only partially effective in cyanide poisoning (Rose et al, 1965; Offterdinger and Weger, 1969), while other authors found HOCO to be effective (Posner et al, 1976b; Wiedemann, 1976; Cottrell et al, 1992; Zerbe et al, 1993; Friedrich and Butterworth 1995, Delaney 1996) and recommended its use. This may in part have been due to doses too low being applied (molar ratio 1:25) (Krapez et al, 1981).

There is but one report of clinical application in a 1.5 months old infant with SNP infusion (Ballesteros Garcia et al, 2003).

Some papers report efficacy following the use of HOCO as cyanide antidote in heavy smokers (Forsyth et al, 1993) and in 'nervous suffering' due to chronic low-level cyanide intoxication (Vincent et al, 1981).

One paper reports the application of 500 mg HOCO each in 4 cases in boys, 8 to 11 years of age, shortly after eating rhizomes of bitter cassava. All were reported to have been critically ill with respiratory failure, bradycardia, hypotension and cardiovascular collapse. 4 further children with similar, but less severe, symptoms, received SN 0.2 ml/kg 3% solution, and an unspecified amount of STS 25%. All 8 children recovered within a few minutes and could be discharged the next day (Espinoza et al, 1992). Data related are too scarce to allow for single case evaluation.

Assessment of efficacy using sequential PSSa
Hydroxocobalamin alone

Sequence 1   n = 19   p = 0.0012 **   Median PSSa before: 3 – PSSa after: 0
Sequence 2   n = 10   p = 0.049 *   Median PSSa before: 3 – PSSa after: 0.5

Hydroxocobalamin in combination with any other antidote

Sequence 1   n = 31   p < 0.0001 ***   Median PSSa before: 3 – PSSa after: 0
Sequence 2   n = 15   p = 0.169 *   Median PSSa before: 3 – PSSa after: 1

Global assessment of efficacy and effectiveness of each antidote alone and in combination

Data from the sequential PSSa study support the following assumptions:

Hydroxocobalamin was used either alone or in combination with other antidotes to cyanide in 75 cases allowing determination of PSSa before and after its administration. In 88.7% of the total, HOCO was used in sequence 1 and 2.

Hydroxocobalamin was used alone in 29 cases. Hydroxocobalamin alone was completely efficient and resulted in full recovery of severe poisonings in 19 cases in sequences 1 and largely efficient in 10 cases in sequence 2.

Hydroxocobalamin was shown to be completely efficient when combined with other antidotes in severe cyanide poisoning which resulted in full recovery in sequence 1 and partial improvement in sequence 2 with a decrease in median PSSa from 3 to 1. The limited number of cases of each associated antidote precludes any recommendation regarding which other antidote should be preferred.

4.6.11 Gaps in knowledge

Gaps in knowledge regarding HOCO have been identified as:

- The pharmacokinetics of the high doses applied for antidotal purposes. What is known is largely based on the low HOCO doses applied for vitamin B_{12} deficiencies (2-3 mg, maximum 5 mg). Pharmacokinetics of cyanocobalamin at doses and plasma concentrations similar to those reported in the treatment of cyanide poisoning.
- Whether cyanocobalamin is the final detoxification complex or is a substrate for the Rhodanese enzyme. The liberation of cyanide from cyanocobalamin formed from HOCO was suggested, with free HOCO again available as antidote and potential for rhodanese metabolism of the liberated cyanide supported by STS.
- The penetration or not of the blood-brain-barrier, which has been reported, but is unlikely in the absence of an active transport mechanism.
- The combination effect with STS – literature reports indicate both a reduction of the HOCO effect by STS and a synergism between both.
- The possible binding of NO, which is implicated as being involved in the antidotal action of SN. This prompts the question, whether the NO binding by HOCO might reduce the antidotal potency of HOCO, and whether NO and the vasodilation caused by it is indeed involved in protection of cytochrome from cyanide (see chapter on SN).

4.6.12 Conclusion

Hydroxocobalamin is a well-defined compound with recognised antidotal action to cyanide through complexation although the kinetics of the reaction is unknown. It acts by a non-enzymatic substitution of the hydroxyl function of HOCO by a cyanide ion that results from the intrinsic affinity of the cobalt atom to cyanide producing cyanocobalamin the detoxification complex. Hydroxocobalamin acts by modifying the toxicokinetics of cyanide by increasing its elimination by non-enzymatic pathway.

Assessing the efficacy of HOCO itself is somewhat hampered by the fact, that it was used in France for many years as a mixture of HOCO and STS, which has been shown to result in the formation of thio-sulphato-cobalamin, a substance unable to bind cyanide. In addition there are reports showing a sharp increase in thiocyanate excretion after application of the mixture. So it must be left open whether the successes undoubtedly seen with the mixture, are due to HOCO, STS, or possibly to both.

The pharmacokinetics of HOCO in both healthy volunteers as well as cyanide poisoned humans is known for a large range of therapeutic doses up to 140 mg/kg, but not for antidotal doses.

Given that in some cases HOCO was administered in a polypragmatic approach, sometimes involving administration of all available antidotes within a short time, it makes it virtually impossible to ascribe positive effects to a single antidote in any situation other than when HOCO was used alone.

The effectiveness of HOCO is strongly dependent on the dose of HOCO. This dose must be adapted to the severity of the poisoning which can be assessed by clinical and biological parameters. In adults, therapeutic cumulative doses range from 5 to 15 g. A dose less than or equal to 2.5 g should not be recommended. There are paediatric dose suggestions, namely 70 mg/kg which can be repeated depending on the initial improvement.

From the casuistics (n = 38) 3 cases cannot be evaluated as the outcome PSSa is not reported. For the remaining 35 cases, it can be said that the mixture of HOCO and STS was used in 20 cases, though in part after other antidotes. In 4 cases it was fully effective when given alone and in a further 5 cases when given either together or after other antidotes. Partial success was achieved in 5 cases by the mixture alone, and in 1 case in combined application with another antidote. No positive outcome was seen in 5 cases, when the mixture was given after or together with other antidotes.

HOCO alone was applied in 15 cases. In 9 cases it was fully effective – 7 with HOCO as only antidote, and 1 each after and together with other antidotes. In two cases partial efficacy was seen with HOCO only, and in 4 cases no effect was seen (2 HOCO only, 2 after other antidotes).

Taken together both the mixture with STS (former cyanide antidote kit) and HOCO alone have been found to be fully effective (reducing the PSSa from 3 to 0) even in severe poisoning cases. That said, for the mixture it remains an open question as to whether HOCO was involved in the successful outcomes, or whether it was STS alone.

The case series reported are very difficult to evaluate both for direct and cyanogenics poisoning and for smoke inhalations so the following interpretation must be treated with caution. Looking at severe cases with direct or cyanogenics poisoning 4/5 did not survive in spite of HOCO treatment, the remaining patient suffered persistent encephalopathy. In mild and moderate poisonings 8 out of 8 patients survived, 2 after the HOCO/STS mixture and 6 after HOCO only. This would appear to indicate that neither HOCO nor the mixture were fully effective in all cases in severe poisonings, even if combined with Co-EDTA.

For smoke inhalation patients the situation is even more unclear. After introduction of HOCO into the treatment regimen in Paris in 1991, 5 out of 5 patients with cardiac arrest died in spite of high-dose HOCO treatment. This seems to have been confirmed later when a further 3 out of 3 patients with critical cyanide blood levels also did not survive in spite of HOCO treatment. All surviving patients did not having critical cyanide levels. Later fatal outcome was also reported in 13 out of 15 patients with initial cardiac arrest, which means that 2 patients might have been saved. Similarly only 2 out of 38 patients survived cardiac arrest, 19 died in hospital (intensive care unit) and 17 at the fire site.

A study showed no improvement of the GCS after HOCO application in 43 out of 52 cases. This is consistent with the findings of several casuistics, where cardio-circulatory improvement could be achieved, while the neurological status did not improve.

HOCO is completely efficient in severe cyanide poisoning as judged from the sequential PSSa.

HOCO alone was completely efficient and resulted in full recovery even of severe poisoning when administered early during the course of the poisoning. The experience of late administration of HOCO is limited thereby precluding any recommendation. In fire victims presenting cardiac arrest, the pre-hospital administration of HOCO has been associated with return of spontaneous circulation and neurological recovery more often than not without neurological sequelae.

HOCO is also effective when combined with other antidotes to cyanide. However, the experience regarding the use of combination of antidotes with HOCO is limited, again precluding any recommendation. There are no data supporting the assumption that the combination of HOCO with STS may result in any alteration of the effectiveness of either antidote alone.

HOCO is not efficacious in cases of very severe poisonings (cardiac arrest), however, that also holds true for other antidotes. Cardiovascular function may be restored, but brain damage from hypoxia from cardiac arrest may not.

Regarding practicality, in contrast to other antidotes (STS, SN, 4-DMAP and Co-EDTA) HOCO cannot be injected immediately, but must be prepared and given by infusion over 15 minutes. In desperate cases, requiring even 2 infusions, this may be a relatively long time before full effectiveness can be achieved, even though partial onset of effectiveness can be expected very quickly. Since a single 5 g infusion of HOCO can bind only 1 mg/l of cyanide thereby only lowering a potentially fatal cyanide blood level of 6 mg/l to 5 mg/l), high doses of HOCO will be needed. Furthermore, the requirement for i.v. or intra-osseous administration limits somewhat its application in conditions without medical facilities. Hydroxocobalamin is also relatively expensive.

Hydroxocobalamin has had the most extensive safety study and post-marketing follow-up of adverse events of the currently available cyanide antidotes.

The most frequent adverse effect is discoloration of urine which occurs in 100% of the treated patients with the recommended dose.

Owing to the deep red colour of HOCO, the antidote may interfere with a large number of common clinical chemistry tests (Appendix D). However, while being statistically significant, these interferences appear of limited clinical relevance. For the sensitive parameters COHb Hydroxocobalamin artificially increases the level. Therefore, in a few cases, hyperbaric oxygen may be given too generously, if the indication is based on the COHb level alone.

A transient, dose-dependent increase in blood pressure that is sometimes described in circumstances of cardiovascular failure induced by cyanide is not considered harmful by a number of authors.

Allergies, even of the anaphylactic type, can be relatively easily treated; skin, blood plasma and urine discoloration are benign effects, at worst disturbing some non-vital laboratory parameters. The problems with haemodialysis machines have only been reported once, and haemodialysis does not seem to have been or to be considered as a cyanide-poisoning treatment approach.

Overall, considering the potentially fatal consequences of cyanide poisoning, the side effects of HOCO are not critical.

Regarding efficacy, the obvious differences between caisutics as assessed for the PSSa and case series (successfuk cardiovasculat stabilisaiton, but persisting CNS effect sin a number of acses) currently cannot be resolved due to unclarities about potentitsal overlaps of the case series.

4.7   Pre-clinical drugs

4.7.1   Introduction

Historical antidotes and antidotes under development

In addition to the antidotes discussed above, there are many more alternatives having been or being evaluated. It is impossible to discuss all of them here, however, the future may well show one or more of them to take over the role as the preferred antidote for cyanide, while others have long been abandoned:

- ACTH (Lurie, 1953);
- p-amino-octanoylphenone (Bright, 1987; Marrs, 1988);
- p-aminopropiophenon (Jandorf and Bodansky, 1946; Ohkawa et al, 1972; Bright, 1987; Marrs, 1988);
- 8-aminoquinoline derivates (Steinhaus et al, 1990) ;
- ascorbate (Hatch et al, 1990);
- atropine (Vick and Vonbredow, 1996);
- BAL (Francone and Mariani, 1951);
- centrophenoxine (Marrs, 1988);,
- chlorpromazine (Levine and Klein, 1959; Way, 1983; Marrs, 1988; Salkowski and Penney, 1995);
- cobalt acetate (Evans, 1964);
- cobalt chloride (Rose et al, 1965; Frankenberg and Soerbo, 1975; Marrs, 1988; Hatch et al, 1990; Salkowski and Penney, 1994);
- cobalt-histidine (Schwarzkopf and Friedberg, 1971; Klimmek et al, 1979; Marrs, 1988);
- cobalt porphyrin (McGuinn, 1994);
- cobamide (Marrs, 1988);
- cobinamide (see below);
- cystine (Wood and Cooley, 1956; Pronczuk de Garbino and Bismuth, 1981; Marrs, 1988);
- diethyldithiocarbamate (Hatch et al, 1990);
- dihydroacetone (see below);
- diltiazem (Marrs, 1988);
- dithionite (Hatch et al, 1990);
- etomidate (Marrs, 1988);
- flunarizin (Marrs, 1988; Borowitz et al, 1995);

- glutathione (Hatch et al, 1990);
- glutathione disulphide (Hatch et al, 1990);
- DL-gylcerinaldehyde (Haustein et al, 1974; Lohs et al, 1974) ;
- haemoglobin-glutamer (Whitworth, 2007);
- hydroxylamine (Kruszyna et al, 1982; Vick and Froehlich, 1991; Bhattacharya et al, 1993; Bhattacharya et al, 1995; Salkowski and Penney, 1995; Vick and Vonbredow, 1996);
- ifenprodil (Pronczuk de Garbino and Bismuth, 1981);
- isosorbide dinitrate (Sun et al, 1995) ;
- alpha-ketobutyrate (Hatch et al, 1990);
- alpha-ketoglutarate (see below);
- mercaptopyruvates (see below);
- methaemoglobin, stromafree (Ten Eyck et al, 1984; Ten Eyck et al, 1985; Ten Eyck et al, 1985, 1986; Salkowski and Penney, 1995; Breen et al, 1996);
- methionine (Hatch et al, 1990);
- methylene blue (Geiger, 1933, Geiger and Gray, 1935);
- naloxone (Leung et al, 1986; Marrs, 1988; Borowitz et al, 1995) ;
- phenoxybenzamine (Burrows and Way, 1976; Vick and Froehlich, 1985; Marrs, 1988; Salkowski and Penney, 1995);
- procaine hydrochloride (Jiang et al, 1998);
- pyridoxale phosphate (Marrs, 1988; Hatch et al, 1990);
- rhodanese (Atkinson et al, 1933; Pronczuk de Garbino and Bismuth, 1981; Marrs, 1988);
- selenite (Hatch et al, 1990);
- sodium cobaltinitrite (Rose et al, 1965; Marrs, 1988);
- sodium ethanethiosulphonate (Marrs, 1988);
- sodium propanethiosulphonate (Marrs, 1988);
- sodium pyruvate (Schwartz et al, 1979; Pronczuk de Garbino and Bismuth, 1981; Marrs, 1988; Salkowski and Penney, 1995; Way, 1983);
- sodium tetrathionate (Marrs, 1988);
- solutions A and B (Callaghan and Halton, 1988; Nicholson et al, 1994);
- sulphate (Hatch et al, 1990);
- tetrathionate (Hatch et al, 1990);
- verapamil (Marrs, 1988).

A potential mechanism of action of some of these and partially also STS (discussed above) is by acting as substrate to mercaptopyruvate sulphurtransferase. In contrast to the mitochondrial rhodanese / cyanide sulphurtransferase this enzyme is located both in the cytosol and the mitochondria, and thus should allow for a better detoxification of cyanide. STS is not an ideal substrate, and mercaptopyruvate decomposes too rapidly in blood to be effective, but further research may turn up useful antidote alternatives (Nagahara et al, 2003).

Some of these merit specific discussion, as recent data are available.

4.7.2   3-Mercaptopyruvate and prodrugs

3-Mercaptopyruvate has been proposed as an antidote for cyanide poisoning (Way, 1983; Marrs, 1988). It is the substrate for 3-mercaptopyruvate sulphurtransferase, which is ubiquitous in the organism and converts cyanide to thiocyanate. As the enzyme is located both in mitochondria and cytosol, 3-mercaptopyruvate should be a more potent cyanide antidote than thiosulphate (Chan et al, 2011).

Indeed, 3-mercaptopyruvate has been shown to be more efficient as prophylactic treatment in hens than STS for protection against KCN – 4 minimum lethal doses were survived without symptoms, while under STS coma for 1 hour occurred before recovery (Mousa, 1991).

3-Mercaptopyruvate decomposes too rapidly in blood to be effective (Nagahara et al, 2003), and i.v. application was unsuccessful to counteract cyanide toxicity (Nagasawa et al, 2007). Therefore 3 prodrugs have been developed and tested using the righting reflex recovery time in mice poisoned by 'cyanide'. All prodrugs given orally 30 to 60 minutes before cyanide dosing were fully protective, i.p. application 5 minutes before cyanide dosing showed 2 of the prodrugs to be more efficient than nitrite/STS and HOCO. One prodrug was highly, one somewhat less efficient 5 minutes after cyanide application (Nagasawa et al, 2007). Chemical names were not given, only structure formulas, and the safety of the prodrugs seemed to be unknown.

More recently, one of these prodrugs, sulfanegen (sodium), 3-mercaptopyruvatedithiane, a 3-mercaptopyruvate dimer, given i.v. and i.m. alternatively, was shown to rapidly reverse cyanide effects on oxy-Hb and deoxy-Hb, and red blood cell cyanide in a rabbit model of sublethal NaCN poisoning (Brenner et al, 2010c).

A further paper described experiments with sulfanegen, applied either i.v. before KCN or HCN application, or together with cobinamide i.m. after KCN or HCN in mice. A beneficial effect of the combination was found, though sulfanegen alone pre-exposure also increased survival up to 100% in lethal KCN and HCN poisonings in a dose-dependent manner (Chan et al, 2011).

Besides the mechanism discussed above, pyruvate has been found to trap cyanide to form cyanohydrins – like α-ketoglutarate (Niknahad et al, 1994). Then again, *in vitro* experiments have shown the efficacy of pyruvate to antagonise cyanide toxicity to be limited (Nuskova et al, 2010).

4.7.3 α-Ketoglutarate

In recent years α-ketoglutarate (α-KG) has been postulated to be an effective cyanide antidote. However, there is no report of its use in humans until now (April 2008), and the safety of α-KG in humans has not been demonstrated either (Bhattacharya and Viajyaraghavan, 2002; Bhattacharya, 2004).

The first report on the protective effect of α-KG against cyanide toxicity is from 1986 (Moore et al, 1986). Further publications came from Japan, before a lot of research was done at the Indian Defense Research and Development Establishment.

The mechanism of action of α-KG (as of pyruvate) judging from in-vitro experiments with pyruvate has long been determined as a reaction with cyanide to form cyanohydrins (Green and Williamson, 1937). Later *in vitro* studies confirmed this binding of cyanide (Aldous et al, 1984; Norris et al, 1990).

Further attempts to elucidate the mode of action *in vitro* pointed to the importance of oxygenation and resulted in the suggestion, that sufficiently high oxygen levels displace cyanide from the cytochrome oxidase, and the cyanide is then trapped by keto acids like α-ketoglutarate (Delhumeau et al, 1994).

α-KG has been shown to be non-toxic at the most effective antidotal dose (2 mg/kg) in rats (Bhattacharya et al, 2001). α-KG does not cause formation of MetHb (Moore et al, 1986; Bhattacharya and Viajyaraghavan, 1991).

α-KG has been used as nutritional supplement at a therapeutic dose of up to 2,500 mg per day, which is about 50 – 70 times lower than the lately recommended antidotal dose (max. 2.5 g/day vs. 2 g/kg) (Bhattacharya, 2004).

Sub-acute application showed a NOAEL of 1 mg/kg. The recommended dose increased organ weights in males without histological correlate (Bhattacharya et al, 2011).

In vitro studies

In vitro pre-treatment with α-KG fully reduced the inhibition of brain cytochrome oxidase by cyanide (Norris et al, 1990).

Again *in vitro*, but this time as post-treatment, rat hepatocytes were protected by α-KG at 3 and 10 mM (among other nutrients), reducing the cytotoxicity in a dose-dependent way. The only other known antidote tested, STS, offered better protection than α-KG when given together with

the KCN, but not if given 30 minutes later. α-KG also restored hepatocyte respiration (Niknahad et al, 1994).

Cytochrome oxidase was measured in testis, heart and liver mitochondria of rats, and bovine heart enzyme in the pure state. α-KG (and pyruvate) had a protective effect, even when given after the KCN (1 mM) application. However, oxygenation played a significant role for the effect. Under conditions of high aeration the displacement of cyanide was 75%, of low aeration only 15%. Cyanide was then trapped by pyruvate or α-KG. In all experiments α-KG was less effective than pyruvate. The dose of α-KG is not reported; it was probably comparable to pyruvate (12 mM) (Delhumeau et al, 1994).

Pre-treatment and simultaneous treatment with 5 mM α-KG were effective against KCN toxicity in rat thymocytes, post-treatment application was ineffective. The positive effects seen were for eosin X exclusion and LDH leakage, while mitochondrial dysfunction, GSH depletion, and DNA damage were not improved (Bhattacharya et al, 2002).

Lately, *in vitro* experiments have shown, that α-KG, especially in combination with N-acetyl cysteine counteracts the cell death mediated by oxidative stress induced by cyanide (Satpute et al, 2010).

Studies on in vivo *prophylactic treatment and treatment simultaneous with CN exposure*

The first publication on actual antidotal use described a 5-fold increase of the lethal dose (LD_{50}) of potassium cyanide (KCN) after pre-treatment with 2 mg/kg α-KG i.p., in line with the effects of 1 g/kg STS (STS) i.p. and 100 mg/kg SN s.c. Combination of α-KG with STS and with STS plus SN applied used before KCN application increased the lethal dose by factors of 15 and 19 (Moore et al, 1986).

Studies from Japan showed a doubled LD_{50} of KCN in mice at a dose of 0.5 g/kg α-KG i.p. However, the timing of the antidote application is not clearly described, probably KCN and the antidotes ´were given at the same point in time (Yamamoto, 1989).

Further experiments in mice (s.c. application of KCN, i.p. application of antidotes) showed, that α-KG (0.5 g/kg i.p.) about doubles the lethal dose (LD_{50}) to mice of potassium cyanide (KCN). The protection factor was more than 4-fold for combinations with STS (1.0 g/kg i.p.) alone and STS (1.0 g/kg i.p.) plus SN (0.1 g/kg i.p). However, α-KG in combination with STS was not more effective than STS plus SN and did not add to the efficacy of this combination (Yamamoto, 1989).

Convulsions caused by 20 and 30 mg/kg KCN were significantly reduced by use of α-KG, especially in combination with STS. α-KG in combination with STS also reduced the decrease in brain GABA, though no better than STS alone, and the calcium increase in crude mitochondrial fractions with synaptosomes from mice brain, both caused by KCN (Yamamoto, 1990).

Pre-treatment with 2 g/kg α-KG i.p. in mice reduced the brain cytochrome inhibition by 8.5 mg/kg KCN to 20% compared to 68% without α-KG (Norris et al, 1990).

Similar experiments were done in India, again with pre-treatment of mice with α-KG (2 g/kg i.p.), STS (1 g/kg i.p.) and SN (0.1 g/kg s.c.). The LD_{50} of KCN was variable for i.p. and s.c. application between 3.5 and 8.2 mg/kg. The protection factors for α-KG ranged from 3.3 to 7.4 (i.p. and s.c.), slightly better but still comparable to STS and SN. These could roughly be doubled by addition of STS and/or SN.

The decrease in cyanide blood concentration after HCN inhalation, and s.c. or i.p. KCN application was greater for α-KG than for STS and SN (Bhattacharya and Vijayaraghava, 1991).

α-KG as oral pre-treatment (100, 250 and 540 mg/kg via gavage) before i.p. injection of KCN at 6.7 mg/kg was assessed without or with 90 mg/kg N-acetylcysteine (NAC). The mortality without antidotal treatment was 80%, it decreased after 100 mg/kg α-KG to 50%, and after 270 and 540 mg/kg to 20%. The addition of 90 mg/kg NAC further decreased mortality to 10% (270 mg/kg α-KG) and 0% (540 mg/kg α-KG). Interestingly NAC alone (90 mg/kg) reduced mortality to 40%. α-KG was most efficient if administered 10 to 20 minutes prior to KCN injection (Dulaney et al, 1991).

A comparison was done in Swiss mice with 4 g/kg α-KG s.c. and 0.4 g/kg Co-edetate s.c. 30 minutes before cyanide dosing. The authors report a better effect of Co-edetate, as it is nearly as effective as α-KG at a tenth of the dose. The table in the publication shows α-KG to be somewhat more effective, albeit at a 10-fold dose. They recommend using Co-edetate (Kravzov et al, 1994).

Pre-treatment of mice with α-KG i.p. before HCN inhalation (400 ppm) was only effective at doses of 750 mg/kg, if given alone. The mortality was not reduced at 250 and 500 mg/kg, 750 mg/kg decreased it from 90% to 60%. Again the combination with STS (1 g/kg plus 500 mg/kg α-KG i.p.) was the most effective antidote. Lower doses were effective against NaCN given i.p. – at a 4.8 LD_{50} of NaCN i.p. (26.5 mg/kg) some protection (78% mortality) was seen with 200 mg/kg α-KG and 500 mg/kg STS i.p. (Huma et al, 1995).

The same authors report in a short congress abstract, that pre-treatment with 2 g/kg α-KG i.p. in mice reduced cyanide concentrations due to 5 mg/kg KCN i.p. in brainstem and heart while

increasing the concentration in blood. Histotoxic hypoxia was reduced. α-KG afforded significantly more protection against mortality than standard doses of Co-EDTA and sodium pyruvate (Hume et al, 1996).

Further experiments were done in mice with pre-treatment or simultaneous treatment with α-KG p.o. at doses of 0.125 to 2 g/kg. The highest protection by pre-treatment was achieved by 2 g/kg with a protection factor of 7.6 compared to the LD_{50} of KCN (12.5 mg/kg). The results again were much improved by adding 1 g/kg STS i.p. with a protection factor of 21.5, and slightly improved further, if SN (100 mg/kg s.c.) was added (Bhattacharya et al, 2002).

The protective effect of α-KG was examined in female rats before and simultaneously with oral KCN application without and with addition of STS and/or SN. The most effective α-KG dose was 2 g/kg, STS was applied at 1 g/kg, SN at 25 mg/kg. The protection factors for this α-KG dose were 5.7 to 6.8, for the combination of α-KG and STS around 16. Pre-treatment with all 3 antidotes resulted in a protection factor of 28.4 (Bhattacharya and Viajyaraghavan, 2002).

In female rats given 0.75 LD_{50} of KCN pre-treatment and simultaneous treatment with α-KG (2 g/kg p.o.) was able to significantly attenuate the levels of cytochrome oxidase in brain and glutathione 60 minutes after KCN-application, though not yet after 30 minutes. Pre-treatment with α-KG improved survival time only, not survival in rats after 17 LD_{50} of KCN. The addition of STS to the treatment protocol (1 g/kg i.p.) improved the results to 33 and 50% survival. Cardiorespiratory parameters were also assessed – α-KG pre-treatment normalised mean arterial pressure after 2 hours, and heart rate and respiratory rate immediately (Bhattacharya et al, 2004).

Sub-acute oral cyanide administration at half of the LD_{50} (14 mg/kg) to rats did not cause lethality, but enzyme, glucose and glutathione alterations. These were reduced by the administration of α-KG (1 g/kg p.o.) and STS (1 mg/kg i.p.) 5 minutes after KCN, even more of both together (Tulsawani et al, 2005).

Again, female rats were administered twice the LD_{50} of KCN p.o., with pre-treatment, simultaneous treatment, and post-treatment with α-KG (2 g/kg p.o.) and/or STS (1 g/kg, i.p.). Different enzymes (cytochrome oxidase, superoxide dismutase, glutathione peroxidase) and glutathione were measured in brain and liver at the time of symptom onset and at death. Both pre-treatment and simultaneous treatment successfully could maintain enzyme levels in brain and liver at about 70 to 90% of normal (Tulsawani and Bhattacharya, 2006).

The same authors reported further experiments with α-KG pre-treatment (0.5, 1 and 2 mg/kg p.o. or i.p. and 0.1, 0.2 and 0.4 g/kg i.v.) resulting in protection factors of 5.4 to 7.6 at the highest dose given orally, i.p. or i.v. Oral administration proved to be most effective, although all gave positive results. Also, the shorter the interval between antidote and KCN administration, the

greater the apparent level of protection. Additionally the effect of α-KG on physiological parameters was assessed. Blood pressure, heart rate, respiratory rate and rectal temperature were not or less altered after α-KG, while it had no positive effect on neuromuscular transmission (Tulsawani et al, 2007).

Recently, the same working group reported on combining α-KG with STS after KCN, and with SN and STS before and after KCN. Prior s.c. injection (before KCN) of female rats with 0.025 mg SN/kgbw followed by treatment (after KCN) with α-KG at 1.0 or 0.5 g/kg bw for 2 or 4 hours, respectively, offered a 40-fold protection. Similar results were obtained using 0.5 g α-KG/kgbw plus 0.5 g STS/kgbw for 2 hours after pre-treatment with SN, STS and α-KG. Simultaneous administration (after KCN) of 1.0 g STS/kg bw i.p. and 20 g α-KG/kg bw orally, followed by 2 g α-KG/kg bw after 2 hours resulted in 38-fold protection (Bhattacharya and Tulsawani, 2009).

The toxic effect of SNP was antagonised by α-KG. The LD_{50} of 12 mg/kg in mice was increased 1.7-fold by α-KG (05 g/kg twice in 20 minutes i.p.), 5.5-fold by STS (1 g/kg i.p.) and 6.9-fold by both together. α-KG decreased the peak cyanide levels by 30%, while STS and the combination decreased them by 88 or 98%. Both antidotes were applied together with the SNP, only α-KG was repeated after 20 minutes (Yamamoto, 1992).

Studies on treatment after CN exposure

First results have been published from a study in 4 dogs exposed by gavage to 5 mg/kg NaCN. Two dogs were treated with 25 mg/kg SN and 1.25 g/kg STS i.v., half the dose at onset of symptoms, the other half within the next 5 minutes. This immediately resulted in dramatically reversed poisoning symptoms. α-KG was given at 60 mg/kg, followed by STS at the above dose. After α-KG stabilisation was seen, the addition of STS reversed the poisoning symptoms (Dalwi et al, 1990).

Further post-treatment results were reported 12 years later. 2-3 minutes after oral application of $2xLD_{50}$ of KCN to rats (p.o.) the antidotes α-KG (2 g/kg p.o.) and STS (1 g/kg i.p.) were given. While 0 out of 10 animals survived without antidote, 3 out of 10 survived after α-KG alone, and 4 out of 10 after the combination. Survival time increased from 4.07 to 5.11 and 5.54 minutes (Bhattacharya et al, 2002).

In contrast to pre-treatment, post-treatment with 2 g/kg α-KG p.o. was ineffective in attenuating the levels of cytochrome oxidase in brain and GSH in female rats given 0.75 LD_{50} of KCN. Post-treatment with the same dose of α-KG led to survival of 33% of rats given 2 LD_{50} of KCN. KCN alone was not survived, and the addition of 1 g/kg STS i.p. to α-KG led to 50% survival (Bhattacharya et al, 2004).

Otherwise unpublished work shortly referred to by the above authors showed that α-KG and STS (doses not given, but in other papers the authors used 2 g/kg α-KG p.o. and 1 g/kg STS i.p.) reduced mortality from 2 LD_{50} of KCN by 50% without increasing survival time (Bhattacharya, 2004).

Besides the positive pre-treatment and simultaneous results on brain and liver GSH and enzymes cited above, post-treatment with α-KG (2 g/kg p.o.) could restore enzyme levels only in the liver, not in the brain the time of symptom onset. At the time of death α-KG alone was not effective for the liver parameters, either, but required combination with STS (1 mg/kg i.p.) (Tulsawani and Bhattacharya, 2006).

In contrast to this it was shown later, that oral treatment with α-KG at 0.5 g/kg alone or with STS resolved the decreased brain levels of the neurotransmitters norepinephrine, dopamine and 5-hydroxytryptamine (Hariharakrishnan et al, 2010).

For cyanogens (different nitriles and SNP) poisonings α-KG was given to rats at a dose of 2 g/kg orally. The LD_{50} with and without α-KG was calculated and found to be increased only for malononitrile and SNP, while it did not significantly increase with treatment for acetonitrile, acrylonitrile, proprionitrile and succinonitrile. (Bhattacharya et al, 2009).

In a subchronic (90-day) study in KCN poisoned rats, α-KG and STS significantly reduced the cyanide toxicity, if given alone, while the combination of both gave maximum protection (Mathangi et al, 2011).

Human studies

Recently, α-KG has been given to healthy human volunteers after radiolabeling for dose fixation. Drinking of water after oral α-KG-application (2 times 10 g) increased the bioavailability to 40% as compared to a single 20 g dose with mean blood concentrations of 15 to 36 µg/ml vs. 10 to 25 µg/ml. Calculations had indicated a required dose of 140 g α-KG for humans in cyanide poisoning. After these results, 20 g in total followed by water produced α-KG-levels in blood known to be protective in animals (Mittal et al, 2010).

A new nano α-KG nebulisation formulation in ethanol / saline was tested for pharmacokinetics and safety in healthy human volunteers with satisfactory results for both aspects. It had been studied as a potential protection and post-exposure treatment mainly for inhaled HCN poisonings (Sultana et al, 2011).

Summary

While α-KG obviously offers very good protection, especially in combination with STS, if given before or simultaneously with cyanide(s) both *in vitro* and in animal studies, the data are not quite as convincing regarding post-treatment application, but are getting better, especially if applied with STS.

In dogs α-KG only stabilised the clinical status of the animals, and STS was required to reverse the symptoms, while in rodents mortality from KCN was reduced by 33 and 50% in different studies by the same group of investigators. Actually, since 1996 all publications on α-KG have come from the same research facility (Division of Pharmacology and Toxicology, Defence Research and Development Establishment, New Delhi, India).

In nitrile intoxications α-KG largely did not significantly increase the LD_{50} except for malononitrile, while it did for SNP.

From the published literature is becomes clear, that α-KG must be combined with STS at high doses to give sufficient protection.

While the safety of α-KG at therapeutical doses has been proven in rats, no such data exist for humans. There is experience with α-KG as a nutritional supplement at doses far lower than required for antidotal use at which recent dose findings studies in humans have been done.

Until now, α-KG has never been used in a human with cyanide poisoning, so no experiences exist.

Thus at the current state of knowledge α-KG cannot yet be regarded as a feasible clinical cyanide antidote. More research is required, in particular regarding safety and clinical use in humans.

From a theoretical point of view, the possibility of oral application of α-KG would make it a very valuable antidote for mass poisonings or first aiders' use.

4.7.4   Cysteine and N-Acetyl-cysteine

Results reported in acrylonitrile poisoning strongly suggest an influence of N-acetyl-cysteine (NAC) on the cyanide level. There are several cases, in which the cyanide concentration in blood dropped sharply after the infusion of NAC, and before the injection of STS (Steffens et al, 1998; Steffens, 2002). This leads to the conclusion, that NAC, the N-acetyl-derivate of cysteine, may be an effective cyanide antidote, not only by binding to the acrylonitrile.

The scarce literature on cyanide poisonings in fact supports this theory. Already in 1926 an antidotal effect of cysteine has been shown (Voegtlin et al, 1926). This was confirmed by a study result, that the antidotal effect of NAC in acrylonitrile poisoning most likely was due to cyanide detoxification (Benz et al, 1990). Known and potential cyanide antagonists were tested in mice. Thirty minutes after the antidotes graded doses of KCN were injected. Compared to the lethal dose of KCN in untreated mice, cysteine had a protection factor of 1.68 – higher than STS (1.48), leading to the suggestion to develop cysteine as a cyanide antidote (Hatch et al, 1990).

In dogs the lethal dose of cyanide was estimated after application of different antidotes during a continuous KCN infusion. Cysteine at a dose of 450 mg/kgbw increased the amount of cyanide needed to cause death, and in fact was the most effective antidote injected as a bolus (better than HOCO, SN, and STS) but did not prevent circulatory failure. It was less effective only than continuous infusions of antidotes. Probably cysteine acts as a sulphur donor to rhodanese and remains in the plasma for extended periods due to protein binding and renal reabsorption (Ivankovich et al, 1980).

Rat hepatocytes i*n vitro* could be protected from cyanide toxicity by L-cysteine, D-cysteine, cystine, thiosulphate and mercaptopyruvate, the latter probably being the main metabolite on the pathway to thiocyanate (Huang et al, 1998). N-Acetylcysteine minimised the cytotoxic changes produced by KCN *in vitro* (Bhattaharya et al, 1999).

An antidotal effect *in vitro* of NAC, particularly together with α-KG, has recently been confirmed (Satpute et al, 2008; 2010).

In regard of the published data and the mentioned results in human cases of acrylonitrile poisoning further studies are definitely warranted.

NAC would be a useful antidote, as it is available worldwide as antidote for paracetamol / acetaminophen poisoning.

4.7.5 Cobinamide

Cobinamide that had already been suggested as a cyanide antidote long ago (Evans, 1964) is the last precursor of cobalamin in the biosynthesis. It has a far greater affinity to cyanide than cobalamin (10^{10}) (Broderick et al, 2006) and binds two cyanide ions (Broderick et al, 2007). The binding affinity is much higher than for HOCO and it is water soluble (Brenner et al, 2010).

Cobinamide has been found to be a better antidote to cyanide if given pre- or up to 5 minutes post-exposure *in vitro* and to Drosophila flies (Broderick et al, 2006). It has also been shown to

be efficient in *in vitro* and animal experiments against nitroprusside-induced cyanide toxicity (Broderick et al, 2007). All these experiments were done with hydroxocobinamide and HOCO.

In rabbits cobinamide was shown to cause significantly faster and more complete recovery of oxy- and deoxy-Hb in cyanide exposed animals than HOCO at equimolar doses. Reduction of blood cyanide concentrations was much better after cobinamide than after HOCO, that was only initially more effective than saline (Brenner et al, 2010a).

Another paper reports the comparison of cobinamide given i.m. again to HOCO, but also to STS, SN and the combination of SN with STS in HCN and KCN in mice, if given before cyanide exposure. Cobinamide given i.p. was 3 times more effective than HOCO in i.p. KCN poisoning and 10 times more effective in inhalative HCN poisoning. After KCN cobinamide saved all poisoned animals, when applied i.m. maximum 2 minutes are apnoea. Cobinamide was also superior to the standard treatments with SN, STS and their combination (Chan et al, 2010).

The i.m. injection in rabbits immediately after cyanide infusion was shown to be effective, too (Brenner et al, 2010b).

Cobinamide is found in humans, probably as a contaminant of vitamin preparations. In rats, the continuous administration of 4 µg/h for 14 days was non-toxic, 59 µM were non-toxic to mammalian cells *in vitro* (Broderick et al, 2006).

4.7.6   Dihydroxyacetone

Dihydroxyacetone (DHA) has been shown to trap cyanide to form cyanohydrins – like pyruvate and α-KG (Niknahad et al, 1994).

2 g/kg DHA given 2 minutes after or 10 before KCN in mice increased the LD_{50} values by a factor of 2.1 and 3, in combination with STS by factors of 2.4 and nearly 10. Pre-treatment with DHA prevented the cytochrome oxidase inhibition and post-treatment accelerated recovery. As DHA is a physiological agent, having been used in dietary regimens and dermatological tests, it might be used as a safe antidote (Niknahad and O'Brien, 1996).

Further experiments by the same group showed a protective effect of 2 and 4 g/kg DHA given orally before KCN, increasing the LD_{50} by factors of about 2 and 3. Additional application of STS after the cyanide dosing (1 g/kg) gave a protection factor of 9.9. Intravenous application of DHA 5 minutes after cyanide (2 and 4 grams) lead to protection factors of nearly 2 and 3. Convulsions caused by cyanide were prevented by DHA (Niknahad and Ghelichkhani, 2002).

4.7.7 Hemoglobin-glutamer

A very faint future possibility of counteracting cyanide toxicity might be the use of Hb-glutamer, consisting of Hb polymerised with glutaraldehyde. It increases the MetHb level in blood, which might make it a cyanide antidote. However, this possibility has not yet really been considered by the makers.

According to an internet search there are 3 products on the market or under development, two of them made from bovine Hb and one from human Hb. One of them (Hemopure®) is registered only in South Africa for acutely anaemic adult surgical patients, and is in phase III studies in other countries, another one is registered for use in veterinary medicine for canine anaemia (Oxyglobin®). The Hb-glutamer made from human Hb (PolyHeme®) is in phase III trials.

4.7.8 Conclusion

α-Ketoglutarate is very close to first use in human cyanide poisoning, while 3-Mercaptopyruvate and its prodrugs required further animal experimental work. This is even more so for cobinamide, though this seems to be a promising antidote from a theoretical point of view. Dihydroacetone, which is a physiological substance, is in the first phases of experimental testing. Use of haemoglobin glutamers as a cyanide antidote is only a theoretical possibility so far without any supporting evidence.

5. POISONING CIRCUMSTANCES

5.1 Direct poisoning (HCN and Cyanide salts)

5.1.1 Introduction

Acute poisoning by cyanide / its salts results in the following clinical symptoms (depending on the severity of intoxication):

- General symptoms as higher perspiration, dizziness, nausea / vomiting, headache, paralysis.
- Typical smell of bitter almonds (odour threshold of 0.7 mg/m^3 in air, but which cannot be perceived by everybody).
- Absence of cyanosis though the patient is asphyxic.
- Cardiovascular / respiratory symptoms as first tachy-dyspnoea, tachycardia, hypertonia, arrhythmia, extra-systolia; in the further course bradypnoea, apnoea, bradycardia, asystolia, hypotonia and cardiovascular collapse. Lung oedema may occur as a secondary complication, not directly related to cyanide poisoning.
- Neurological symptoms start with anxiety and agitation, progressing to seizures, coma and brain oedema.

In contrast to other important intoxication requiring immediate antidotal treatment for survival, e.g. organophosphate poisoning, a typical neurological sign in patients poisoned by cyanide are dilated, unresponsive pupils, even before brain death has ensued. This allows for differential diagnosis in unclear poisoning circumstances.

Laboratory analyses indicating acute cyanide intoxication are listed below:

- Direct:
  - Blood-cyanide can be detected by two methods:
    - Qualitative detection by the von Clarmann (Mathes and von Clarmann, 1977) method.
    - Quantitative analysis (regularly used today) by the Feldstein and Klendshoy method.
  - Cyanide in exhaled air can be detected by chromatometric gas analysis. This can be done before or after intubation. However, such equipment will rarely be available.

- Indirect:
  - High thiocyanate in the urine shows the excretion of the metabolite.
  - High lactate is also a strong hint to cyanide intoxication (Baud, 2002), as is a high oxygen-saturation of the venous blood (as a result of the lack of oxygen-consumption in the cells).

However, with the possible exception of the von Clarmann qualitative method, and potentially lactate measurement, all these analyses take time and cannot be done at the location of the intoxication. At best they can be performed in the hospital/ED.

5.1.2 Identity

Hydrogen cyanide (HCN) is a clear, almost colourless liquid or gas with a faint odour of bitter almonds. It is a weak acid that is completely soluble in water. Sodium cyanide (NaCN) and potassium cyanide (KCN) are hygroscopic white crystalline solids (salts) that are odourless when dry but emit a slight odour of HCN and ammonia in damp air (Klenk et al, 1987; Gail et al, 2000). They are freely soluble in water.

The identity of these cyanides and some physico-chemical properties are listed in Table 27.

Table 27: Identity and some physico-chemical properties

IUPAC name		Hydrocyanic acid	Sodium cyanide	Potassium cyanide
CAS number		74-90-8	143-33-9	151-50-8
Formula		HCN	NaCN	KCN
Structure		H–C≡N	Na–C≡N	K–C≡N
Molecular mass		27.03	49.01	65.12
log K_{ow} [a]* at 20°C		–0.25 [a]	Not applicable	Not applicable
Vapour pressure at 20°C		830 hPa [a]	None	None
Solubility in water at 20°C		No limit (miscible in all ratios)	370 g/kg [b]	400 g/kg [b]
pKa value [c]	at 20°C	9.36 [d]	9.36 [d]	9.36 [d]
	at 25°C	9.21	9.21	9.21
	at 30°C	9.11	9.11	9.11

* Partition coefficient, log K_{ow} (octanol/water)
[a] Hansch et al, 1995 cited by US EPA, 2000
[b] Gail et al, 2000
[c] –log (acidity constant), measure of extent of acidity
[d] Izatt et al, 1962.

5.1.3 Pharmacokinetics and toxicity

In the stomach cyanide is present as HCN in presence of gastric acid. Absorption in humans is reported to be between 10 and 81% (Gettler and Baine 1938).

In small amounts cyanide is bound to the physiological met-Hb (0.5-1% of total Hb) and reversibly to plasma proteins. Therefore in low level exposures, plasma cyanide is much lower than whole blood cyanide.

No distribution volume can be reasonably defined, as it is strongly dependent on the cyanide concentration. The distribution to compartments also is concentration dependent.

Again, metabolism is concentration dependent. Only a small fraction is excreted unchanged via lung and kidneys. The most important pathway is the formation of thiocyanate by rhodanese. If there are sufficient sulphur donors e.g. thiosulphate or mercaptopyruvate, available most cyanide is excreted as thiocyanate (ATSDR, 1993).

Small amounts are excreted as CO_2 after entering the C_1-pool (ATSDR, 1993).

It is generally recognised that there is a considerable variability in regard to 'lethal doses' reported for cyanide in the literature and that this is probably due to differences in supportive care and therapy rendered during treatment. For adults, the potentially lethal oral dose of cyanide salts in the absence of medical care (antidote treatment) is between 200 and 300 mg (Bonsall, 1984).

In humans and also in other mammals, cyanide is rapidly absorbed in the form of HCN and its salts following oral, dermal or inhalation exposure. Only the more lipophilic HCN can cross the cell membranes (lipophilic media in presence of water cyanides will form also HCN (see above, equation for weak acids/bases). The onset of cyanide toxicity is normally very rapid (within seconds to minutes) in particular via the inhalation and oral routes. The following symptoms of cyanide intoxication may develop: anxiety and excitement, rapid breathing, faintness, weakness, headache (pulsating), constricting sensations in the chest, facial flushing, dyspnoea, nausea and vomiting, diarrhoea, dizziness, drowsiness, confusion, convulsions, incontinence of urine and faeces, coma and respiratory irregularities (Vogel et al, 1981; Pontal et al, 1982; Bismuth et al, 1984; all cited by Ballantyne, 1987). An early characteristic feature of acute cyanide poisoning, particularly with smaller doses, is the development of tachypnoea and hyperpnoea, resulting in increased tidal volume. This may clearly enhance the uptake of HCN in the early stages of respiratory exposure to the vapour (Ballantyne, 1987a).

The mechanism of toxicity is by inhibition of oxygen utilisation by tissues. Cyanide forms a complex with iron in cytochrome oxidase, which is located in tissues at the cellular level of the mitochondrial membrane. The complex formation inhibits oxygen from receiving electrons from the cytochrome oxidase and a so-called intracellular or cytotoxic anoxia occurs, i.e. oxygen is present but cannot be utilised by the cell. Since neurons and cardiac myocytes are highly dependent on aerobic metabolism and therefore extremely sensitive to the deprivation of oxygen, the brain and heart are the most sensitive organs to cyanide posioning.

In the case of the brain, if aerobic metabolism fails due to the inactivated cytochrome oxidase by cyanide, the neuron immediately loses its capacity to conduct nervous pulses properly and the brain fails to function with consequent loss of consciousness. If this stage continues for some minutes, the damage becomes irreversible and the neurons die. For these reasons, prolonged hypoxia, regardless of its cause, often results in injury to the brain. Toxicants that inhibit aerobic cell respiration like HCN and hydrogen sulphide have the same effect (Anthony and Graham, 1991).

Table 27: Blood levels of cyanide and symptoms after Chataigner et al (1989)

Concentration in whole blood		Symptoms
µg CN⁻/l	µmol CN⁻/l	
0 - 320	0 - 12	Absence of toxic signs, non-smokers
16 - 520	0.6 - 20	Absence of toxic signs, non ??-smokers
500 - 1,000	19 - 38	Slight intoxication, vasodilatation-tachycardia
1,000 - 2,500	38 - 96	Moderate intoxication, confusion, limited consciousness
2,500 - 3,000	96 - 115	Serious intoxication, coma
≥ 3,000	≥ 115	Potential lethal intoxication

In humans, the acute lethal dose is approximately 1.5 mg/kgbw following oral uptake. The lethal dose by the dermal route will depend upon the area of skin exposed. A dose of approximately 100 mg/kgbw is lethal even if only a small area of skin is contacted. Inhalation of 270 ppm HCN (303 mg/m^3) is immediately lethal, exposure to 110 to 135 ppm (124 – 152 mg/m^3) leads to death in 30 to 60 minutes (Bonsall, 1984). Blood cyanide levels of 2,500 to 3,000 µg CN⁻/l of total blood were related to serious intoxication and coma.

An overview can be found in the preceding cyanide report by ECETOC, JACC no. 53.

5.1.4 Casuistics

The following table gives an overview of the severe intoxications.

Table 28: Summary of severe intoxications

Number of cases	Type of poisoning, route of exposure (number of cases)	Antidote treatment (number of cases)		Outcome (number of cases)	Remark	Reference
		Main antidote	**Additional antidote**			
2	KCN oral, accidental	Supportive	Supportive	Dead	Children; one child died before treatment	Prajapati, 1992
1	KCN suicidal	Supportive	Supportive	Survived	Rhabdomyolysis	Brivet, 1983
1	KCN suicidal	Supportive	Supportive	Survived		Prieto, 2005
2	CN suicidal	Supportive	Supportive	Dead		Yen et al, 1995
1	KCN suicidal	DMAP	STS	Survived		Kampe, 2000
5	CN suicidal (4) HCN inhalation (1)	DMAP	STS	Dead (3), survived (2)	Long time between exposure and treatment (4), overdosage with adverse effects (1)	Stickel, 2008
1	Dermal-inhalation	DMAP	STS	Long-term sequelae	Consequential damage	Jakobs, 1984
2	NaCN suicidal	SN	STS (1) DMAP + STS (1)	Dead Survived	Treatment-combinations	van Heijst, 1987
1	CN suicidal	SN	STS	Survived		Lee-Jones, 1970
1	KCN suicidal	SN	STS	Long-term sequelae		Peters, 1982
1	KCN suicidal	SN	STS + HBO	Dead		Litovitz, 1983
14	CN suicidal	SN	STS	Dead (6) Survived (8)		Yen et al, 1995
1	KCN suicidal	SN	STS	Survived		De Busk, 1969
1	KCNsuicidal	SN	STS	Survived		Martín-Bermudez, 1997
1	KCN suicidal	SN	STS	Survived		Hall, 1997
1	KCN suicidal	SN (500 mg)	STS (25 g) + Co-EDTA (300 mg) + HOCO (3 mg)	Dead	Long time (2.5 h) between exposure and treatment	Buchanan, 1976
2	HCN inhalation	SN	STS	Survived (2)		Chen and Rose, 1952

Number of cases	Type of poisoning, route of exposure (number of cases)	Antidote treatment (number of cases)		Outcome (number of cases)	Remark	Reference
		Main antidote	Additional antidote			
2	HCN inhalation	AN (1) N + SN (1)	Supportive (1) STS (1)	Survived (1) Dead (1)	Patient with nitrite kit died before treatment	Würzburg, 1996
1	KCN suicidal	STS	AN	Survived		Miller and Toops, 1951
1	KCN suicidal	STS	Supportive	Dead		Lundquist, 1992
1	KCN suicidal	STS	Supportive	Dead		Mutlu, 2002
1	HCN inhalation	STS	AN	Survived		Lam and Lau, 2000
1	HCN inhalation	STS	Supportive	Survived		Bonsall, 1984
3	CN OA	Co-EDTA	STS + HOCO	Dead (1) Survived (1) Long-term sequelae (1)		Bismuth, 1984
1	NaCN oral, accidental	Co-EDTA	Supportive	Dead	Received: 15×300 mg (4.5 g)	Hillmann, 1974
1	KAuCN suicidal	Co-EDTA	AN, Glukose	Dead		Wright and Vesey, 1986
1	CN suicidal	Co-EDTA	SN, Glukose	Survived		Davis and Ewer, 1988
1	KCN suicidal	Co-EDTA	STS + HOCO	Survived		Hoang The Dan, 1981
1	HCN inhalation	Co-EDTA	Supportive	Dead	$3\times$ recommended dose	Singh, 1989
1	KCN oral, accidental	HOCO	Supportive	Survived		Hung 2009
1	KCN suicidal	HOCO	STS	Survived		Tassan, 1990
2	HgCN suicidal	HOCO	Hg-antidote	Dead (1) Survived (1)		Benaissa, 1993
5	CN suicidal	HOCO	Supportive (2) STS (2) STS + Co-EDTA (1)	Dead (4) Long-term sequelae (1)		Borron, 1996; 2007
1	NaCN suicidal	HOCO	STS	Survived		Harry, 1985
1	KCN	HOCO	STS + Co-EDTA + Methylene blue	Survived		Lutier, 1971
1	CN oral, suicidal	DMAP	Supportive	Dead		Zilker, 2000

Table 29: Summary of direct poisoning cases

Case			Antidote treatment									Reference
Age (y)	Sex	Type of poisoning	Cumulative dose of antidote (g)							Number of treatment steps	Final PSSa	
			AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO	Other			
24	M	AgCN + NaCN, oral	-	0.3	6.25	-	-	-	-	1	2	Chen and Rose, 1952
23	M	Metal cleaner	-	0.45	18.75	-	-	-	-	1.5	3	Krieg and Saxena, 1987
2.5	F	Metal cleaner	-	0.076	2.5	-	-	-	-	1	2	Krieg and Saxena, 1987
80	M	Metal cleaner	-	0.6	3	-	-	4	-	2 (SN) or 1 (STS)	3	Mannaioni, 2002
18	M	CaCN (insecticide)	NS	0.3	2	-	-	-	-	1	3	Mascarenhas, 1969
17mo		KCN	-	0.45	25	-	-	-	Methylene blue	1.5 (SN) or 2 (STS)	4	Berlin, 1970
19	M	KCN	-	0.3	NS	-	-	-	-	1	2	Wananukul and Kaojarern, 1992
22	M	NaCN 1.5 g	-	0.3	25	-	-	-	-	1 (SN), 2 (STS)	3	De Busk and Seidl, 1969
24	F	KCN 4-6 g	-	0.3	17.5	-	-	-	-	1	3	De Busk and Seidl, 1969
35	M	CN	-	0.3	12.5	-	-	-	-	1	4	Lee-Jones et al, 1970
25	M	KCN 1 g	NS	0.3	12.5	-	-	-	-	1	2	Lee-Jones et al, 1970
14	F	CN	-	0.3	12.5	-	-	-	-	1	1	Lee-Jones et al, 1970
23	M	KCN	-	0.5	25	-	0.3	3	-	1	4	Buchanan, 1976
31	M	KCN	NS	1.2	100	-	-	-	-	4	3	Peters, 1982
23	F	KCN	NS	0.3	12.5	-	-	-	-	1	2	Litovitz et al, 1983
24	F	CN	-	0.3	25	-	-	-	-	1 or 2	4	Litovitz et al, 1983
59	M	CN	-	0.3	12.5	-	-	-	-	1	2	Wesseon, 1985
34	M	KCN 1 g	-	0.3	12.5	-	-	-	-	1	2	Hall, 1987
32	M	KCN 3 g	-	0.6	25	-	-	-	Toluidine blue	2	3	van Heijst, 1987
21	M	NaCN	-	0.3	20	0.73	-	-	Toluidine blue	1 or 7 (DMAP 250 + 6 × 80)	4	van Heijst, 1987

Case			Antidote treatment									Reference
			Cumulative dose of antidote (g)							Number of treatment steps	Final PSSa	
Age (y)	Sex	Type of poisoning	AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO	Other			
29	M	KCN + NaCN	NS	0.3	12.5	-	-	-	-	1	2	DiNapoli, 1989
24	F	KCN	-	0.3	12.5	-	-	-	-	1	2	Johnson, 1989
31	M	KCN	NS	0.6	25	-	-	-	-	2	2	Selden, 1990
54	M	KCN (1.65 g)	NS	0.6	25	-	-	-	-	2	3	Goodhart, 1994
21 cases										-		Yen et al, 1995
19	F	CN	-	0.3	12.5	-	-	-	-	1	3	Martín-Bermudez, 1997
60	M	KCN 0.6 g	-	0.3	12.5	-	-	-	-	1	3	Feihl, 1982
30	M	NaCN 3 g	-	0.3	12.5	-	-	-	-	1	2	Johnson, 1988
19	F	CN	-	0.45	18.75	-	-	-	-	1.5	2	Chin, 2000
52	M	HCN inhal.	NS	0.3	25	-	-	-	-	1	2	Potter, 1950
6 individuals		CN	NS	-	-	-	-	-	-	1	1 or 2	Wolfsie, 1951
6 individuals		inhal.-dermal	-	0.3	12.5	-	-	-	-			
	M	HCN inhal.	-	0.3	1	-	-	-	-	1	3	Chen and Rose, 1952
	M	HCN inhal.	NS	-	-	-	-	-	-	1	1	Chen and Rose, 1952
	M	HCN inhal.	NS	-	-	-	-	-	-	1	1	Chen and Rose, 1952
64	M	HCN inhal.	NS	0.3	12.5	-	-	-	-	1	3	Chen and Rose, 1952
22	F	HCN inhal.	NS	0.3	-	-	-	-	-	1	2	Chen and Rose, 1952
61	M	HCN inhal.	NS	0.3	12.5	-	-	-	-	1	2	Chen and Rose, 1952
67	M	HCN inhal.	NS	-	-	-	-	-	-	1	1	Chen and Rose, 1952
44	M	HCN inhal.	NS	-	-	-	-	-	-	1	1	Chen and Rose, 1952
6 ind.	M		NS	0.3	12.5	-	-	-	-	1	1	Chen and Rose, 1952
47	M	HCN inhal.	-	NS	-	-	-	-	CaTS	1	2	Lurie, 1953
37 cases										-		Würzburg, 1996
	M	HCN inhal.	NS	0.3	12.5	-	-	-	-	1	1	Lam, 2000
35	M	HCN inhal.	NS	0.3	12.5	-	-	-	-	1	1	Brueske, 1997
41	M	HCN dermal	NS	0.3	25	-	-	-	-	1	2	Potter, 1950
19	M	KCN powder	NS	0.75	31.25	-	-	-	-	2.5	2	Thomas and Brooks, 1970

Case			Antidote treatment									Reference
Age (y)	**Sex**	**Type of poisoning**	**Cumulative dose of antidote (g)**							**Number of treatment steps**	**Final PSSa**	
			AN (pearls)	**SN**	**STS**	**DMAP**	**Co-EDTA**	**HOCO**	**Other**			
26	F	KCN	-	-	6	0.25	0.3	-	-	1	3	Daunderer, 1974
23	F	KCN	-	-	25	1	-	0.02	-	1	2	van Heijst, 1987
29	M	KCN	-	-	9	0.25	-	-	-	1	3	Kampe, 2000
35	F	CN	-	-	NS	NS	-	-	-		2	Rachinger et al, 2002
		CN	-	-	-	0.25 (4×) 0.5 (2×)	-	-	-	1 (0.25 g) or 2 (0.5 g)	1 (twice), 2 (twice), 3 (once), 4 (once)	Zilker and Eyer, 2005
34	M	KCN	-	-	10	0.5	-	-	-	2 (DMAP)	2	Stickel, 2008
23	M	NaCN	-	-	25	0.5	-	-	-	1	3	Stickel, 2008
28	F	CN	-	-	25	0.5	-	-	-	1	3	Stickel, 2008
39	M	KCN 5 g	-	-	20	0.25	-	-	-	1 (DMAP) / 2 (STS)	2	Stickel, 2008
17	M	KCN	-	-	17	0.75	-	-	-	3 (DMAP)	4	Stickel, 2008
36	F	CN	-	-	10	0.25	-	-	-	1	4	Stickel, 2008
40	F	CN	-	-	10	0.25	-	-	-	1	4	Stickel, 2008
	F	CN	-	-	-	0.25	-	-	-	1	2	Zilker, 2009 (Personal comm.)
5 individuals		CN inhal.	-	-	-	0.25	-	-	-	1	1	Zilker and Eyer, 2005
2 individuals										1	3	
56	M	NaCN inhal.	-	-	25	1	-	-	-	4	2	Stickel, 2008
	M	HCN dermal	-	-	12.5	0.25	-	-	-	1	3	Jacobs, 1984

NS, not stated; -, not applied

The following list of direct poisonings was extracted from the main PSSa database (Appendix B).

Table 30: Direct poisonings extracted from the PSSa database

Case			Antidote treatment									Reference
			Cumulative dose of antidote (g)						Number of treatment steps	PSSa		
Age (y)	Sex	Type of poisoning	AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO		Before	After	
STS alone												
27	F	KCN	-	-	NS	-	-	-	1	3	4	Favarel, 1982
NS	NS	HCN	-	-	NS	-	-	-	2	3	0	Bonsall, 1984
25	M	KCN	-	-	7.8[b]	-	-	-	1	3	0	Lundquist, 1992
83	M	KCN	-	-	NS	-	-	-	1	3	4	Mutlu, 2002
STS and nitrites												
41	M	HCN	3	0.3	25	-	-	-	3	3	0	Potter, 1950
51	M	NS	2	0.3	25	-	-	-	1	3	0	Potter, 1950
32	M	KCN	NS	-	NS	-	-	-	2	3	0	Miller and Toops, 1951
26	M	CaCN	-	NS	NS	-	-	-	2	2	0	Wolfsie, 1951
29	M	HCN	NS	0.3	7.5	-	-	-	4	3	1	Wolfsie, 1951
45	M	HCN	NS	0.3	12.5	-	-	-	2	3	0	Wolfsie, 1951
39	M	HCN	NS	0.3	12.5	-	-	-	2	3	0	Wolfsie, 1951
44	M	HCN	NS	-	5	-	-	-	2	2	0	Wolfsie, 1951
NS	M	HCN	NS	0.3	12.5	-	-	-	2	3	0	Wolfsie, 1951
NS	M	AgCN + NaCN	-	-	6.25	-	-	-	1	1	0	Chen and Rose, 1952
NS	M	HCN	-	0.6	2	-	-	-	1	3	2	Chen and Rose, 1952
22	M	HCN	NS	0.3	12.5	-	-	-	1	3	0	Chen and Rose, 1952
61	F	CNCl	20	0.6	12.5	-	-	-	2	3	0	Chen and Rose, 1952
67	M	HCN	NS	0.3	12.5	-	-	-	2	3	0	Chen and Rose, 1952
22	M	HCN	-	-	12.5	-	-	-	1	1	0	Chen and Rose, 1952
24	M	HCN	NS	0.3	12.5	-	-	-	1	1	0	Chen and Rose, 1952
25	M	HCN	NS	0.3	12.5	-	-	-	1	1	0	Chen and Rose, 1952
26	M	HCN	NS	0.3	12.5	-	-	-	1	1	0	Chen and Rose, 1952
32	M	HCN	NS	0.3	12.5	-	-	-	1	1	0	Chen and Rose, 1952

Case			Antidote treatment									Reference
			Cumulative dose of antidote (g)						Number of treatment steps	PSSa		
Age (y)	Sex	Type of poisoning	AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO		Before	After	
39	M	HCN	NS	0.3	12.5	-	-	-	1	1	0	Chen and Rose, 1952
NS	M	HCN	NS	0.3	12.5	-	-	-	1	1	0	Chen and Rose, 1952
62	M	HCN	-	-	NS	-	-	-	1	1	2	Lurie, 1953
41	F	HCN	-	0.5	8.1	-	-	-	3	3	3	Chen and Rose, 1956
24	M	NaCN	NS	0.3	25	-	-	-	2	3	0	De Busk and Seidel, 1969
31	F	KCN	-	0.3	17.5	-	-	-	1	3	0	De Busk, 1969
1.4	M	KCN	-	0.15	12.5	-	-	-	2	0	3	Berlin, 1970
25	M	CN salt	-	0.3	12.5	-	-	-	1	3	4	Lee-Jones, 1970
14	M	KCN	-	NS	> 12.5	-	-	-	1	3	0	Lee-Jones, 1970
23	F	CN salt	-	NS	> 12.5	-	-	-	1	3	0	Lee-Jones, 1970
30	M	KCN + AgCN	NS	-	NS	-	-	-	1	2	0	Trapp, 1970
3	M	KCN	NS	-	NS	-	-	-	1	3	0	Feihl, 1982
39	M	KCN	-	0.9	150	-	-	-	1	3	0	Peters, 1982
23	F	KCN	NS	0.3	12.5	-	-	-	1	2	0	Litovitz, 1983
27	M	KCN	-	NS	NS	-	-	-	1	3	2	Uitti, 1985
27	M	KCN	-	0.3	12.5	-	-	-	1	3	0	Wesson, 1985
12	M	KCN	-	0.3	12.5	-	-	-	3	2	0	Hall, 1987
2.5	M	CN salt	1	0.45	18.75	-	-	-	1	3	0	Krieg and Saxena, 1987
29	F	CN salt	1	0.076	2.5	-	-	-	1	3	0	Krieg and Saxena, 1987
30	M	NaCN	2	0.3	12.5	-	-	-	1	2	0	Johnson and Mellors, 1988
29	M	KCN + arsenic CN	-	0.3	12.5	-	-	-	1	3	0	DiNapoli, 1989
24	F	KCN	-	0.3	12.5	-	-	-	1	3	0	Johnson, 1989
46	M	KCN	-	NS	NS	-	-	-	1	3	2	Rosenberg, 1989
39	M	KCN	-	NS	NS	-	-	-	1	3	0	Feldman, 1990
31	M	KCN	NS	0.6	25	-	-	-	1	3	0	Selden, 1990
12	M	KCN	NS	-	NS	-	-	-	1	3	0	Kasamo, 1993
23	M	KCN	3	-	10	-	-	-	1	3	0	Nakatani, 1992

Case			Antidote treatment									Reference
			Cumulative dose of antidote (g)						Number of treatment steps	PSSa		
Age (y)	Sex	Type of poisoning	AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO		Before	After	
54	M	KCN	NS	0.6	25	-	-	-	1	2	3	Goodhart, 1994
NS	NS	HCN	-	NS	NS	-	-	-	1	3	4	Würzburg, 1996
NS	NS	NaCN	-	NS	NS	-	-	-	1	1	0	Würzburg, 1996
19	F	NS	-	NS	NS	-	-	-	1	3	0	Martín-Bermúdez, 1997
80	F	CN salt	-	0.45	33.25	-	-	-	2	3	0	Chin and Calderon, 2000
19	F	HCN	1	0	12.5	-	-	-	1	3	3	Lam and Lau, 2000
NS	M	HCN	1	0.3	12.5	-	-	-	1	1	0	Lam and Lau, 2000
21	M	KCN	-	0.6	3	-	-	-	1	3	0	Mannaioni, 2002
STS, nitrites and DMAP												
21	M	NaCN	-	0.3	20	> 0.25	-	-	2	3	4	Van Dijk, 1987
STS, nitrites, Co-EDTA and HOCO												
23	M	KCN	-	-	NS	-	0.3	4	1	3	0	Lutier, 1971
NS	M	KCN	-	NS	NS	-	0.6	4	1	2	0	Lutier, 1971
NS	NS	KCN	NS	-	NS	-	0.6	4	1	2	0	Lutier, 1971
STS, nitrites and HOCO												
23	M	KCN	-	0.5	75	-	-	0.03	1	3	4	Buchanan, 1976
STS and DMAP												
43	M	KCN	-	-	10	0.25	-	-	1	1	0	Werner, 1979
29	M	NaCN	-	-	16	0.25	-	-	2	3	3	Werner, 1979
20	M	KCN	-	-	12	0.25	-	-	1	1	0	Werner, 1979
19	F	HCN	-	-	8	0.25	-	-	1	1	0	Werner, 1979
25	M	KCN	-	-	5	0.25	-	-	1	2	0	Werner, 1979
25	M	KCN	-	-	3	0.375	-	-	2	3	3	Werner, 1979
30	F	KCN	-	-	25	0.25	-	-	1	0	0	Werner, 1979
38	M	KCN	-	-	10	0.5	-	-	1	2	0	Werner, 1979
26	M	KAu(CN)$_2$	-	-	NS	0.25	-	-	1	3	0	Kampe, 2000

Case			Antidote treatment									Reference
Age (y)	Sex	Type of poisoning	Cumulative dose of antidote (g)						Number of treatment steps	PSSa		
			AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO		Before	After	
23	M	KCN	-	-	10	0.25	-	-	2	2	0	Zilker and Stickel, 2008
28	M	NaCN 1g	-	-	12.5	0.5	-	-	1	3	0	Zilker and Stickel, 2008
39	F	KCN	-	-	12.5	0.5	-	-	1	2	2	Zilker and Stickel, 2008
56	M	KCN	-	-	2	0.25	-	-	2	3	2	Zilker and Stickel, 2008
17	M	NaCN	-	-	12.5	1	-	-	1	2	2	Zilker and Stickel, 2008
36	M	KCN	-	-	170 ml	3 ml	-	-	1	3	4	Zilker and Stickel, 2008
40	F	KCN	-	-	1	0.25	-	-	1	3	2	Zilker and Stickel, 2008
64	F	CN salt	-	-	1	0.25	-	-	1	3	3	Zilker and Stickel, 2008
28	F	KCN	-	-	NS	NS	-	-	1	3	2	Zaknum, 2005; Rachinger, 2002
NS	M	HCN	-	-	4	0.15	-	-	1	3	0	Steffens, 2010
NS	M	HCN	-	-	3	0.1	-	-	1	2	0	Steffens, 2010
NS	F	KCN	-	-	10	0.25	-	-	1	2	0	Zilker, 2010
26	F	KCN	-	-	6	0.25	0.3	-	2	2	0	Daunderer, 1974
STS and Co-EDTA												
22	F	KCN	-	-	20	-	0.6	-	1	3	0	Hoang The Dan, 1981
28	M	CN salt	-	-	24.5[c]	-	0.3	-	1	3	0	Lundquist, 1992
32	M	KCN	-	-	8	-	4	-	1	ND	4	Jourdan, 1993
STS, Co-EDTA and HOCO												
NS	M	KCN	-	-	NS	-	0.3	4	2	2	0	Lutier, 1971
30	F	CN salt	-	-	16	-	NS	15	2	3	4	Baud, 2001

Case			Antidote treatment									Reference
			Cumulative dose of antidote (g)						Number of treatment steps	PSSa		
Age (y)	Sex	Type of poisoning	AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO		Before	After	
STS and HOCO												
15	F	KCN	-	-	8	-	-	4	1	3	1	Tassan, 1990
28	M	KCN	-	-	8	-	-	4	1	3	4	Jamali, 1993
27	F	Mercury	-	-	8	-	-	4	1	3	4	Benaissa, 1995
28	F	KCN	-	-	16	-	-	5	2	2	0	Baud, 2001
44	M	KCN	-	-	8	-	-	9	2	3	0	Baud, 2001
54	F	NaCN	-	-	8	-	-	4	1	3	0	Harry, NS
Nitrites alone												
26	M	HCN	-	NS	-	-	-	-	1	2	0	Wolfsie, 1951
40	M	HCN + CN salts	-	NS	-	-	-	-	3	3	0	Wolfsie, 1951
33	M	HCN	-	NS	-	-	-	-	1	3	0	Wolfsie, 1951
44	M	HCN	-	NS	-	-	-	-	1	3	0	Wolfsie, 1951
29	M	HCN + CaCN	NS	0.3	-	-	-	-	2	3	0	Wolfsie, 1951
NS	M	HCN	-	NS	-	-	-	-	1	NS	0	Chen and Rose, 1952
67	M	HCN	-	NS	-	-	-	-	1	NS	0	Chen and Rose, 1952
44	M	HCN	-	NS	-	-	-	-	1	NS	0	Chen and Rose, 1952
NS	NS	HCN	-	NS	-	-	-	-	1	1	0	Würzburg, 1996
NS	NS	HCN	-	NS	-	-	-	-	1	3	0	Würzburg, 1996
NS	NS	HCN	-	NS	-	-	-	-	1	3	0	Würzburg, 1996
NS	NS	HCN	-	NS	-	-	-	-	1	2	0	Würzburg, 1996
NS	NS	HCN	-	NS	-	-	-	-	1	2	0	Würzburg, 1996
NS	NS	HCN	-	NS	-	-	-	-	1	2	0	Würzburg, 1996
NS	NS	HCN	-	NS	-	-	-	-	1	3	0	Würzburg, 1996
Nitrites and DMAP												
19	M	KCN	-	0.15	-	0.125	-	-	2	2	1	Werner, 1979

Case			Antidote treatment									Reference
			Cumulative dose of antidote (g)						Number of treatment steps	PSSa		
Age (y)	Sex	Type of poisoning	AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO		Before	After	
Nitrites and Co-EDTA												
35	M	CN salt	11	-	-	-	0.6	-	1	3	0	Naughton, 1974
DMAP alone												
26	F	KCN	-	-	-	0.25	-	-	1	1	0	Werner, 1979
50	M	Benzyl-CN	-	-	-	0.25	-	-	1	1	0	Werner, 1979
Co-EDTA alone												
32	M	KCN	-	-	-	-	NS	-	1	1	1	Paulet, 1965
68	M	NaCN	-	-	-	-	2.7	-	3	3	4	Hillman, 1974
50	M	KCN	-	-	-	-	0.3	-	5	3	0	Yacoub, 1974
42	F	KAu(CN)$_2$	-	-	-	-	0.3	-	Know	1	1	Wright and Vesey, 1986
24	M	AgCN	-	-	-	-	1.2	-	1	3	2	Brown, 1987
24	M	AgCN	-	-	-	-	0.9	-	Know	3	3	Singh, 1989
Co-EDTA and HOCO												
NS	NS	CN	-	-	-	-	NS	NS	1	3	4	Bismuth, 1984
NS	NS	CN	-	-	-	-	NS	NS	1	3	4	Bismuth, 1984
NS	NS	CN	-	-	-	-	NS	NS	1	3	3	Bismuth, 1984
HOCO alone												
63	M	KCN	-	-	-	-	-	10	1	3	2	Baud, 2001
38	M	KCN	-	-	-	-	-	10	1	2	3	Baud, 2001
14	M	Hg(CN)	-	-	-	-	-	5	1	3	0	Baud, 2001
52	M	KCN	-	-	-	-	-	5	1	2	0	Baud, 2001
26	M	KCN	-	-	-	-	-	15	2	3	4	Baud, 2001
32	M	KCN	-	-	-	-	-	10	2	3	0	Baud, 2001
53	F	AuCN + KCN	-	-	-	-	-	5	1	1	0	Baud, 2001
38	M	BrCN (halogenated CN)	-	-	-	-	-	5	1	1	0	Baud, 2001

Case			Antidote treatment										Reference
Age (y)	Sex	Type of poisoning	Cumulative dose of antidote (g)						Number of treatment steps	PSSa			
			AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO		Before	After		
51	M	KCN	-	-	-	-	-	5	1	3	0		Weng, 2004
50	M	KCN	-	-	-	-	-	5	1	3	0		Coentrão, 2010
32	F	KCN	-	-	-	-	-	5	2	3	0		Hung, 2009
22	M	NaCN	-	-	-	-	-	5	1	2	0		Baud, unpublished

Case studies are detailed Appendix C

[a] Reported as 95mmol

[b] Reported as 120 mmol

[c] Reported as 98 mmol

ND, not determined; NS, not stated; -, not applied.

Several people died when they had taken cyanide contaminated Tylenol capsules (paracetamol). All capsules of one specific lot contained some 65 mg KCN. As most of them were found dead, none of them was treated with specific antidotes (Duena, 1983).

Ten Indian children (aged between two and seven years) ate potassium-cyanide-powder which they had found by chance while playing outside. Because of their complaints about stomach pain and nausea and because of cyanosis of their lips, the parents suspected intoxication and induced vomiting. They brought their children to the emergency department. One of the young victims was already dead when reaching the ED, another one died though CPR was performed. These two children had dissolved the powder, in contrast to the other ones). The rest of the group was only given supportive treatment because of the unknown character of the poison. This consisted of gastric lavage, administration of oxygen and infusions in four patients, who had developed mild symptoms of cyanide intoxication after about 1 to 2 hours. The other four children didn't show any symptoms and because of that they were only observed. All of them could be sent home after 2 days in complete remission (Prajapati, 1992).

Cases 11-16: The remaining 6 cases (22 – 39 years old) were fire fighters overcome by HCN in a flour mill. They all wore masks. Upon removal of the masks, they all complained of nausea, shortness of breath, dizziness and coughing. Forty-five minutes later they all received AN by inhalation, 0.3 g SN and 12.5 g STS i.v., and methamphetamine. Five of 6 developed headaches. No blood cyanide concentrations were available. All were discharged within 2 to 3 hours (Chen and Rose, 1952).

5.1.5 Treatment and/or prophylaxis

186 case reports involving intoxications with hydrocyanic acid or one of its salts comprised 51 mild, 69 moderate and 66 severe poisonings.

The following graph shows prevalence of suicidal and accidental incidents in relation to the severity of intoxication:

Figure 6: Relation between severity of cyanide intoxication and background (n = 186)

	mild intoxication (n=51)	moderate intoxication (n=69)	severe intoxication (n=66)
■ oral-accidental intoxication (n=23)	11	5	7
■ accidental intoxication via skin or inhalation (n=68)	32	26	10
■ suicides (n=95)	8	38	49
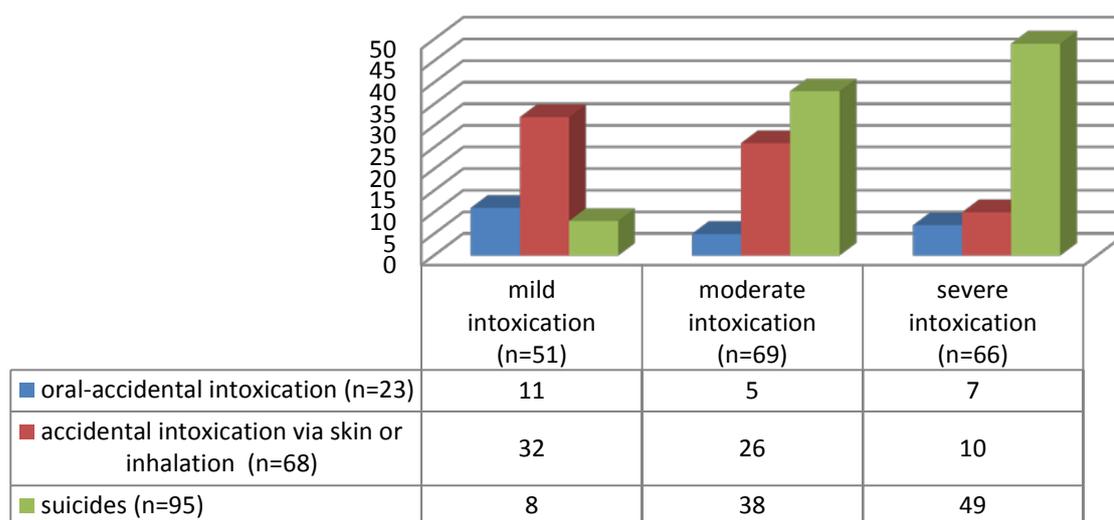
It can be seen that more or less the same numbers of patients had suffered from accidental intoxication (91 out of 186) as had attempted suicide (95 out of 186), however, severe intoxications were far more prevalent in the latter group. This perhaps can explain why accidental intoxications very infrequently result in death or long-term illnesses; whereas suicides, consistent with their supposed intention, often lead to death or irreversible damages.

Most cases (80) were treated with the nitrite antidote kit, the least number of cases with STS alone (only 9 patients). The other antidotes were administered to more or less to the same numbers of patients; 22 received HOCO, 21 received cobalt-EDTA and 24 received 4-DMAP; while the remaining 29 patients were only given supportive treatment.

Figure 7: Severity of intoxication (n = 186)
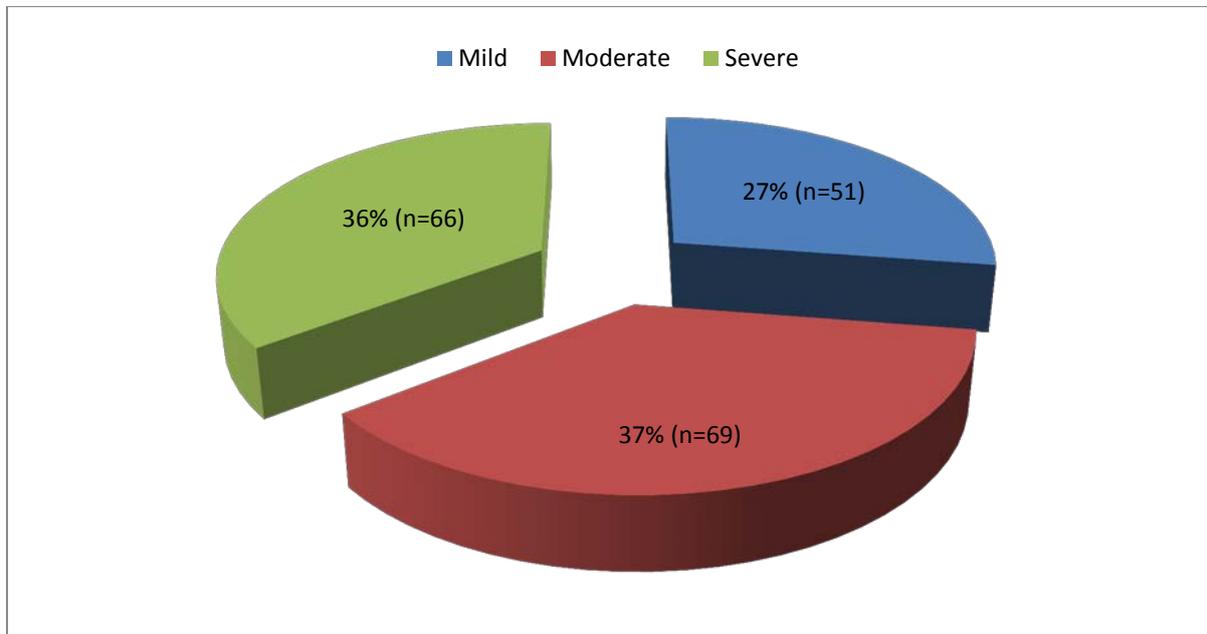
Figure 8: Frequency of administration of the different antidotes (n = 186)
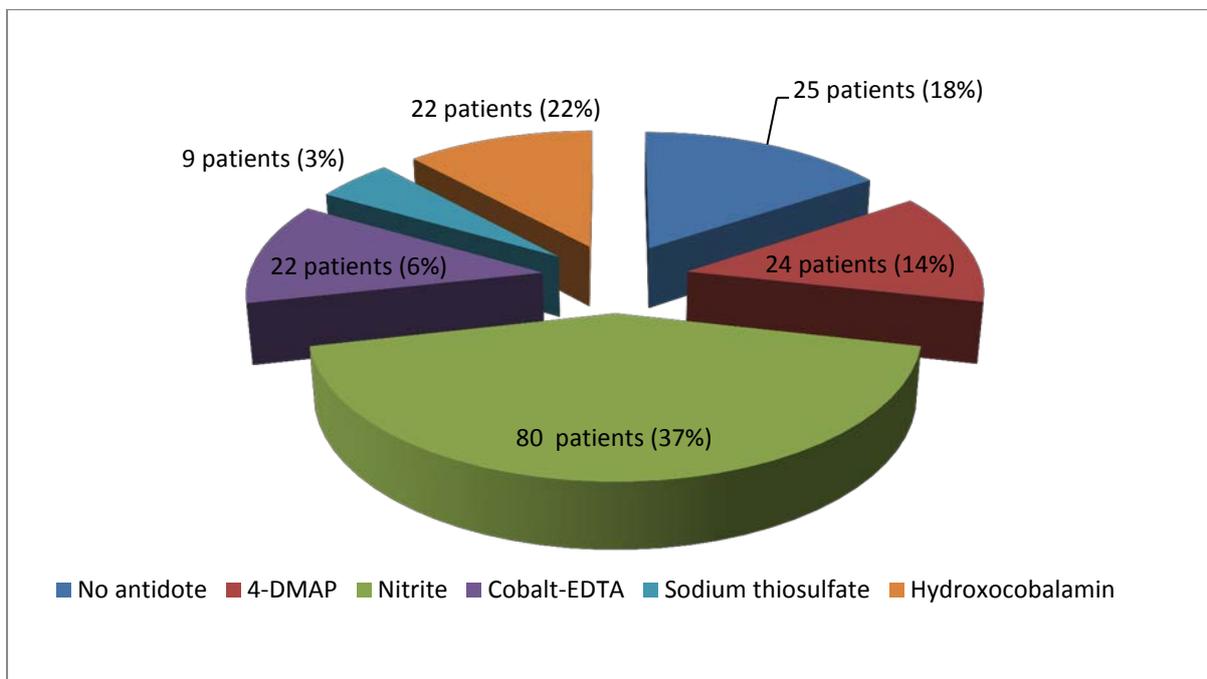
Most of the 29 patients who were only treated supportively had suffered from mild poisoning (18 out of 29). In contrast to that, HOCO was rarely used in mild intoxications (only 4 out of 22 patients) and STS was never administered to a mildly poisoned person.

The following graph shows the relation between the different forms of therapy and the severity of the intoxication.

Figure 9: Relation between the different forms of therapy and the severity of the intoxication
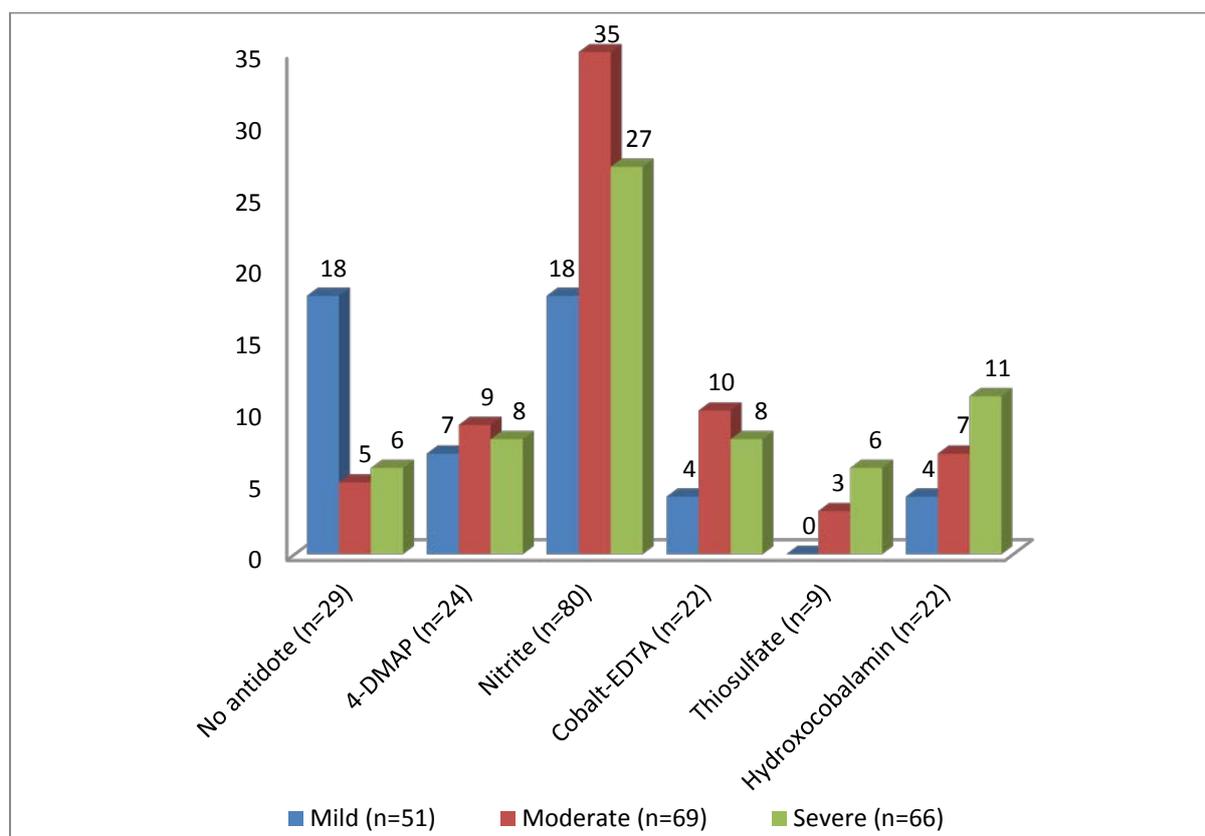
All patients who had suffered from a mild intoxication could be rescued. This did not depend on the antidotal therapy applied. The only differences were seen in the incidence of adverse side effects: none were seen under supportive treatment, STS or HOCO. 4-DMAP and nitrites only showed adverse effects if administered in high doses (higher than that recommended). In such cases the patients developed haemolysis with 4-DMAP therapy and low blood pressure / circulatory instability under SN (also under AN, but less frequently). Both antidotes could cause excessive MetHb formation with cyanosis if overdosed. The only antidote that showed adverse effects even under normal dosage was cobalt-EDTA (see cases in table above).

The patients with moderate intoxications could also be rescued, independent of the antidote or form of therapy used. Only one patient treated with 4-DMAP developed cerebral ischemia and, as a consequence, long-term cerebral damage. Adverse effects were reported under nitrite, 4-DMAP, however in this case the antidote had been extremely overdosed) and most often under cobalt therapy.

The following graph visualises these results.

Figure 10: Success of the different therapies administered in severe cyanide poisoning (n = 66)
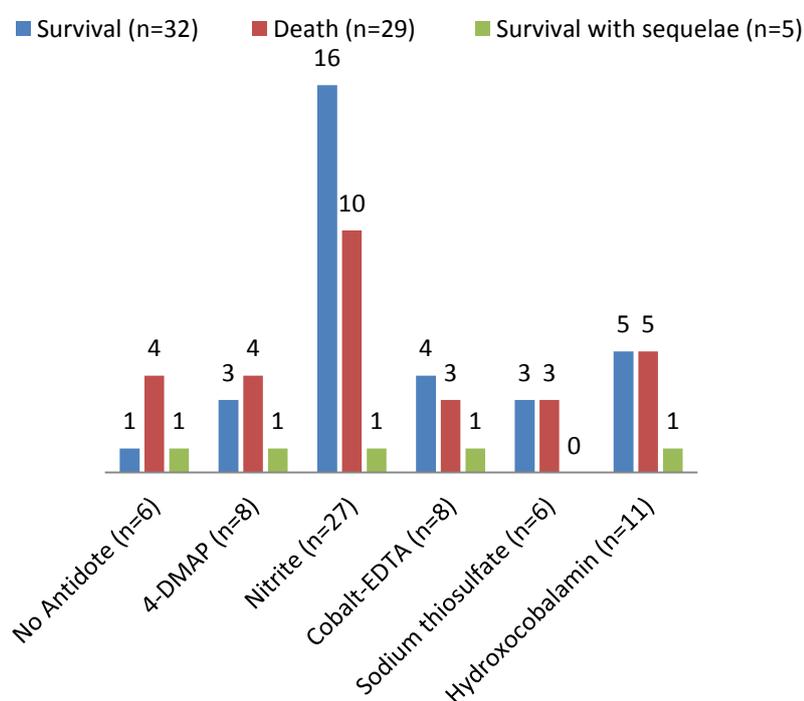
It can be seen that the specific treatment options were not administered evenly: The nitrite antidote kit, or parts of it, was most commonly used with it being administered to more than one third (41%) of the patients. The other antidotes were only administered to a few patients each (4-DMAP and cobalt-EDTA to 8 individuals each (12%), HOCO to 11 victims (16%) and thiosulphate or supportive care alone were only given to 6 individuals each (9%).

In these severely poisoned patients, one can see differences in the outcomes dependedant on the specific therapy although it is difficult to infer significant due to the considerable discrepancy in the numbers cases, the differences in time between intoxication and beginning of the treatment, and other factors of interference. However, the summary of the results shows the following:

Under merely supportive treatment, 4 out of 6 of the patients (66.6%) died.

The outcome with dicobalt-EDTA therapy was comparatively unsatisfactory: Four out of 8 patients died (50%). However, this could also have occurred because this strong, but badly tolerated antidote was often only administered after a long time had passed, after the patient had already suffered from cardiorespiratory arrest, or after the failure of other antidotes (combinations). Notwithstanding this, the significant intrinsic toxicity of the substance itself may have contributed to the outcome.

4-DMAP showed about the same efficacy as dicobalt-EDTA (4 patients died and and equal number survived) but this antidote was also often used late or in "hopeless" intoxications e.g. on victim found in cardiorespiratory arrest.

Comparable efficacy could be seen with STS and HOCO.

Under STS therapy 3 patients died and and equal number survived (50% survival).

Under HOCO therapy it was more or less the same outcome as 5 patients died and 6 survived (45% survival). It has to be mentioned that HOCO was rarely administered as single therapy but was often combined with 4-DMAP, nitrites, cobalt-EDTA or combinations of these three and with STS.

The best results were achieved with the nitrite therapy (63% of the patients survived).

Although if one compares all the cases treated with the different antidotes all forms of therapy appear to be equally effective (i.e 80% of the patients survived with only STS appearing to be less effective with an apparent survival rate of only 66.7%) this is not reliable since it does not take into account the different severity of the intoxications and the likely outcome if antidote treatment had not been given. To illustrate this point supportive treatment alone seems to be quite good, but this is only true in mild and moderate as in severe intoxications death would ensue.

Figure 11: Survival rates under different antidote treatments

	No specific therapy (n=29)	4-DMAP (n=24)	Nitrite therapy (n=80)	Cobalt-EDTA (n=22)	Sodium thiosulphate (n=9)	Hydroxoco balamin (n=22)	Sum
■ Survival	25	20	70	18	6	17	156
■ Death	4	4	10	4	3	5	30
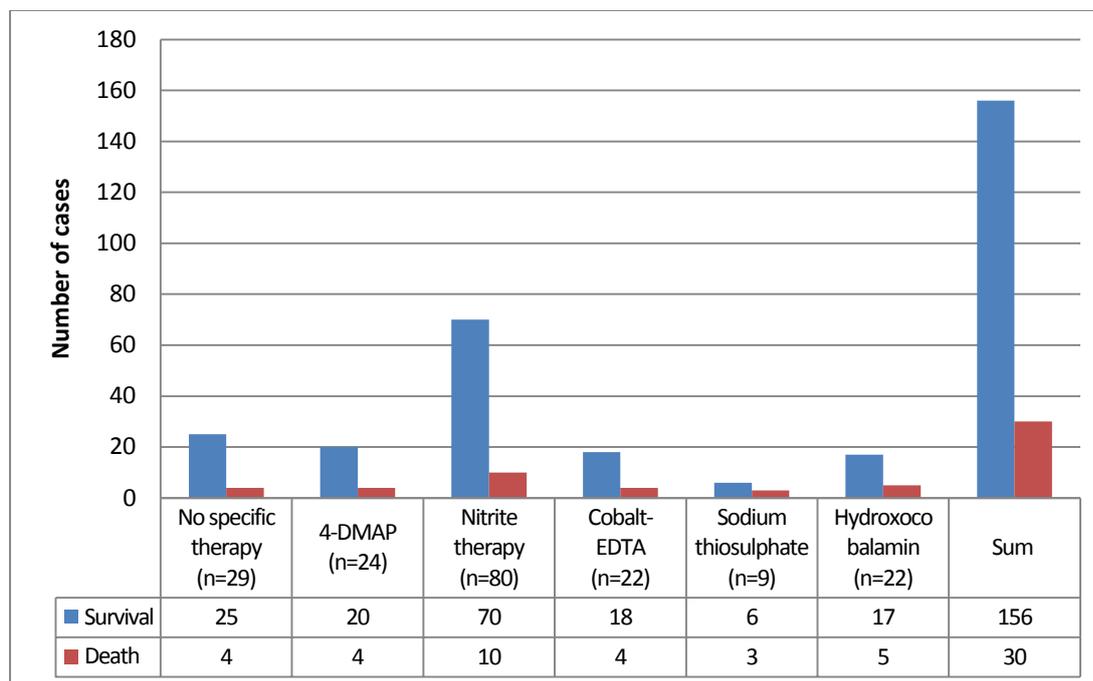
5.1.6 Occurrence of adverse effects in clinical use

Reported adverse events are events previously ascribed to the antidote and temporally related to the antidote administration.

The frequency of the adverse events and overdose was evaluated using:

- Case series.
- Case reports.
- Author's report of the adverse effect.
- Interpretation by the TF of findings reported in the cases.

The severity of the adverse events and overdose was made using the sequential PSSa, when possible.

5.1.7 Assessment of efficacy and effectiveness of antidotes in clinical use

Data from the sequential PSSa study support the following assumptions.

Cyanide salts:

Global efficacy regarding antidotes used alone.

In sequence 1:

- Hydroxocobalamin alone was efficient in poisoning with CN salts resulting in full recovery (median PSSa after 0) in severe poisoning (median PSSa before 3) (0 = 0.0335).

Global efficacy regarding antidotes used in combination.

Sequence 1: the following combinations were shown efficient.

- STS + SN was efficient resulting in full recovery (median PSSa after 0) in severe poisoning (median PSSa before 3) (p = 0.0120).
- DMAP + STS was efficient resulting in partial recovery (median PSSa after 1.5) in moderate poisoning (median PSSa before 2) (p = 0.009).

Hydrocyanic acid:

Global efficacy regarding antidotes used alone.

In sequence 1:

- The one antidote exhibiting highly efficiency was AN resulting in full recovery (median PSSa after 0) in severe poisoning (median PSSa before 3) (0 = 0.0004).
- The following antidotes were not efficient when used alone: STS, SN, 4 DMAP, HOCO.

Global efficacy regarding antidotes used in combination.

Sequence 1:

- The combination of AN + SN + STS was efficient resulting in full recovery (median PSSa after 0) in mild poisoning (median PSSa before 31) (p = 0.042).

5.1.8   Discussion and conclusion

In mild or moderate intoxications by HCN or cyanide-salts therapy with a specific antidote showed no better outcome than supportive treatment alone. The decision whether and which therapy is applied depends not only on antidote efficacy and effectiveness, but also on factors like the clinical severity, the certainty of cyanide intoxication, speed of onset of antidotal effect, possible adverse effects, and the availability of a specific antidote, as well as the costs of antidotal treatment. In other words the choice of antidote may be governed by other factors such as these rather than efficacy considerations.

Supportive care as only therapy cannot be recommended in severe intoxications because the chance for surviving decreases even in comparison to the unsatisfying efficacy of the specific treatment (20-30% less effective).

Supportive treatment and treatment with STS did not show any adverse effects. The same holds true for HOCO in the recommended dosages. 4-DMAP leads to severe haemolysis if it is overdosed. At the recommended dose there might be a mild haemolysis which does not endanger the patient. When nitrite therapy is administered one should keep in mind the suppressive effect on the BP which can also occur under AN therapy. This effect could always be compensated by vasoactive substances if used in the recommended dose.

The antidote that should not be recommended any longer, as long as others are available, is dicobalt-EDTA. Dicobalt-EDTA can show severe side effects even in normal dosing. The available data did not indicate convincing efficacy although it has to be recognised that possible reasons for this are its use only in cases of poisoning that were unlikely to survive or its administration too late in the treatment. The likelihood of severe side effects due to the antidote treatment combined with the availability of other antidotes that are better tolerated strongly supports a case for its withdrawal from use.

According to the case reports the nitrite antidote kit was tolerated quite will and was the one that showed the best efficacy in severe poisoned patients (about 60% survived). This result might be due to its use over many years and patients, which allows a greater statistical power (i.e. because more cases were observed in comparison to other antidotes). The other antidotes showed a 50% chance for survival. The bias concerning the late application or the administration in hopeless cases (cobalt-EDTA and 4-DMAP) is explained above.

Once cardiac arrest had occurred before treatment the specific antidotal treatment was effective in only very few cases. Most of these patients died, independent of the antidote administered.

Treatment recommendations

- In mild or moderate intoxications is it adequate to apply only supportive treatment. If one is sure about the cyanide intoxications and afraid of a deterioration of the patient's condition, one could use STS or HOCO. Both antidotes showed no or minimal adverse effects under clinical circumstances. The onset of the antidotal effect of Thiosulphate is delayed. If the condition of the patients allows for waiting the administration of STS can be repeated. If the patient's condition deteriorates a faster acting antidote should be used. STS should always be considered as an additional antidote to all the fast acting antidotes for it ensures cyanide removal through urinary elimination and thereby preventing a possible rebound effect. Moreover, it can be given repeatedly because of its good tolerance. Hydroxocobalamin is also an antidote with few adverse effects. But it has other disadvantages, for example, the relatively large amount of antidote required. It also it has to be stored protected against light and has a shelf-life of two years making it an expensive option. As HOCO is a comparatively new antidote there are not enough clinical data and with cyanide poisoning

becoming a very rare event it does not seem to be likely that there could be collected a reliable number within a short / straightforward reasonable period of few years.

- In case a patient is severely poisoned with coma, low blood pressure and brady-dysrhythmia thiosulphate may not act fast enough and HOCO may take too long to be administered in a sufficiently high dose. Methaemoglobin forming antidotes may be still be used successfully under these circumstances and the administration of the recommended dosages of 4-DMAP or nitrites is highly recommended Administration of 4-DMAP or nitrites will probably not create haemolysis or a falling blood pressure.

- In acute and severe cyanide-poisoning it is beyond doubt that in addition to the supportive treatment (depending on the clinical presentation) a potent antidote must be administered as fast as possible. This implicates different consequences:

  o As cyanide and organophosphate-intoxications are the only ones that require specific antidotal treatment without hesitation, a cyanide antidote should be available on every ambulance and the staff should be trained in its administration.

  o As thiosulphate acts too slowly and HOCO may not be available on the ambulance in a doses high enough for antidotal treatment, the more sensible alternatives seem to be SN or 4-DMAP. Both substances act immediately.

  o As cyanide is not removed from the system neither by nitrite, 4-DMAP nor HOCO the administration of thiosulphate is sensible following the use of them. This allows the cyanide to be removed through urinary elimination and may prevent a rebound effect.

  o Nitrites and 4-DMAP can cause adverse effects mostly if they are used without indication or if they are overdosed. For the doctor it is important to know the dosage and mode of action of each antidote so that he/she is able to react fast and properly. Nitrites regularly result in an immediate / acute and often steep drop of the blood pressure which is already low in severe poisoning. Quite often cardiopressors such as norepinephrine are needed to counteract this effect. This might be of less importance as vasopressors are needed in severe intoxications anyway. 4-DMAP, on the other hand, can lead to haemolysis and can cause alteration in the liver and renal function. Haemolysis occurs with delay so that there is time for reaction: performing blood-transfusions and/or haemodialysis. Although this side-effect might happen even under correct dosing of the antidote it does not result in negative consequences for the patient's life. Both antidotes can be administered very quickly i.v. in a small volume as the amount of antidote that is needed is low (250 mg for 4-DMAP and 300 mg for SN). They are relatively low cost and can be stored without any limitations for a long time. Repeat administration is possible for the nitrites. A second dose of 4-DMAP should be avoided and is not necessary. At the moment one of these two very potent antidotes seems to be the choice in acute and severe poisoning by cyanide.

  o AN as part of the American antidote kit is often recommended as first-aid-antidote as it can be applied easily. As far as the practicality for first responders is concerned that

might be true because the AN ampoules only have to be broken over the mouth, nose or tubus and the patient can inhale it, which means no venous cannula is needed. But one should be aware that AN is also a nitrite and as such a substance is capable of suppressing the blood pressure, too. One should be careful with its applications and not more than two or three ampoules should be broken without controlling the blood pressure. But in this dosage the possible effect may be not sufficient. So AN alone is not advisable in severe cases of poisoning.

o In addition it is an issue, that AN has been removed from the new antidote kits (SN/STS only), that it has been taken out of UK HSE recommendations and support, and that it may no longer be available as medical product. However, that data from the PSSa evaluation for AN (see above), indicate efficacy and effectiveness in moderate to severe poisonings.

o Today it cannot be ignored that cyanides might be applied by some terrorists for an attack. In such a scenario of possible mass poisoning the antidote of choice should be affordable and storable for a long time. It will have to be discarded without having been required, and replaces regularly. Most important would be its fast and easy administration in such a scenario even by staff not used to doing this every day. Few antidotes seem to fulfil this requirement. AN would be an option (see limitations described above), 4-DMAP might fulfil these criteria with the advantage of possible i.m. application. This could be given to many victims by paramedics and nurses. The i.m. injection causes significant pain, but this may be tolerable in case of mass casualties at vital risk.

Acute poisoning by cyanide is a rare event. Childhood cyanide poisonings are even more uncommon and most antidotes have never been used in children. As far as nitrites are concerned there are tables for adjusting the dose of the antidote to the patient's haemoglobin. However, determination might be difficult with acute poisoning requiring immediate action. A further fall in children's physiologically low blood pressure may need careful consideration. As Di-cobalt-EDTA and 4-DMAP may lead to dangerous side effects in children their use cannot be advised. However, there is a dosage recommendation by the maker for 4-DMAP. Besides the supportive treatment, STS and HOCO seem to be the favourable substances in these situations. They have little or no side effects and there is no reason in believing them being not tolerated in children. It is likely that in children a high enough dose of HOCO can be achieved in a short time even i.o. This makes it the cyanide antidote of choice for children (Table 70 in Section 6.4).

5.2 Cyanogenic substances

5.2.1 Cassava

Cassava, also known as manioc, tapioca or yuca is a shrub from the spurge family originating from South America but grown all over the tropics and subtropics as annual crop because of its drought resistance and high yields. Today it is a staple crop especially in many parts of Africa (Essers et al, 1992). It has an edible tuberous root rich in starch and the leaves can also be eaten.

However, leaves, roots and tubers must be processed before human consumption as they contain free and bound cyanogenic glycosides (mainly linamarin). Leaves can contain up to 5g/kg fresh weight linamarin, roots and tubers about 250 mg/kg (White et al, 1998).

Different methods of preparation will decrease the cyanogens/cyanide content and make cassava suitable for human consumption, e.g. by ventilating the flour mixed with water, by cooking or fermentation (Padmaja, 1995). The maximum cyanide content in edible cassava flour has been defined as 10 mg/kg (CAC, 1999).

5.2.1.1 Substance data

No data available or applicable as natural product.

5.2.1.2 Intrinsic properties (pharmacokinetic and toxicity data)

When the cassava plant is traumatised or uprooted glycosides are rapidly converted by the enzyme (linamarase (or linase) to cyanide (Osuntokun et al, 1969; Cheok, 1978). Linamarase converts linamarin to acetone cyanohydrin (ACH), which spontaneously at a pH above 5, at a temperature above 35ºC, or by enzymatic breakdown by hydronitrile lyase, forms cyanide. Acetone cyanohydrin is the main cyanogen in poorly processed cassava roots (White et al, 1998).

There are different cultivars of cassava. The so-called bitter cultivars give higher yields and are more drought resistant, but contain more cyanogenic glycosides (Essers et al, 1992).

Cyanide can also be formed from glycosides in the stomach by the gastric hydrochloric acid (Cheok, 1978), but this is not the primary mechanism of cyanide formation.

An increase in blood cyanide levels has been demonstrated after a cassava meal (Uwakwe et al, 1991). Gastric content was analysed on three occasions, and only one (after ingestion of fresh cassava) was positive for cyanide.

A potential role of native linamarin for the acute toxicity remains to be elucidated.

5.2.1.3 Casuistics

Acute Exposure:

Reports on acute toxicity of cassava requiring antidotal treatment for cyanide poisoning are scarce, those receiving treatment even more so. Several children's deaths in Nigeria have been attributed to cassava consumption, but no medical details are available (Aregheore and Agunbiage, 1991). Further cases have been described in more details (see appendix C).

Since cyanide blood levels have only been reported in 5 cases from 2 publications but these were taken either before treatment or long after ingestion (Akintowa and Tunwashe, 1992; Ruangkanchanasetr et al, 1999) it is not possible to ascribe these to cyanide intoxication with any certainty.

The acute cases, as far as they are sufficiently documented, are listed in Table 1:

Table 31: Acute cassava poisonings

Case				Antidote treatment						Reference
Age (y)	Sex	Latency time (h)	Cyanide poisoning?	Antidote					Final PSSa	
				AN (pearls)	SN	STS	Co-EDTA	Other		
6	M	2.5	Probable, clinical symptoms	-	Yes	Yes	-	O$_2$	2	Eiró Gonsalves et al, 1956
6	M	5	Possible, clinical symptoms	-	Yes	Yes	-	-	1	Eiró Gonsalves et al, 1956
5	F	8	Possible, clinical symptoms	-	Yes	Yes	-	-	1	Eiró Gonsalves et al, 1956
9	M	NS	Probable, clinical symptoms	-	Yes	Yes	-	-	2	Eiró Gonsalves et al, 1956
6	M	5	Possible, clinical symptoms	Yes	Yes	Yes	-	-	1	Eiró Gonsalves et al, 1956
4	F	5	Possible, clinical symptoms	-	Yes	Yes	-	-	1	Eiró Gonsalves et al, 1956
7		0.5	Yes, cyanide in gastric content	-	-	-	-	-	4	Queisser, 1966
5	M	0.5	Yes, cyanide in gastric content	-	-	-	-	-	4	Queisser, 1966
		10	Possible, clinical symptoms	-	-	-	-	-	3	Dawood, 1969
2.5	M	7.5	Possible, clinical symptoms, cyanide in food	-	-	Yes	-	-	1	Cheok, 1978
1.5	F	7	Possible, clinical symptoms, cyanide in food	-	-	Yes	-	O$_2$		Cheok, 1978
7	F	1	Possible, clinical symptoms	-	-	-	Yes	-	1	Brian, 1990
7	F	NS	Possible, clinical symptoms	-	-	-	-	-	1	Mlingi et al, 1992
4	F	NS	Possible, clinical symptoms	-	-	-	-	-	1	Mlingi et al, 1992
10	M	NS	Possible, clinical symptoms	-	-	-	-	-	2	Mlingi et al, 1992
4	F	9	Yes, cyanide in blood	-	Yes	Yes	-	O$_2$	3	Ruangkanchanasetr et al, 1999
1.5	M	9	Yes, cyanide in blood, bitter almond smell	-	-	-	-	O$_2$	2	Ruangkanchanasetr et al, 1999

Case studies are detailed Appendix C

NS, not stated; -, not applied.

Table 32: Cassava poisonings extracted from the PSSa database (Appendix B)

Case		Antidote treatment								Reference
Age (y)	Sex	Cumulative dose of antidote (g)					Number of treatment steps	PSSa		
		AN (pearls)	SN	STS	Co-EDTA	HOCO		Before	After	
STS alone										
2.5	M	-	-	12.5	-	-	1	1	0	Cheok, 1978
1.5	F	-	-	12.5	-	-	1	3	0	Cheok,1978
STS and nitrites		-			-					
6	M	-	1.5	1.8	-	-	2	2	0	Eiró Gonsalves, 1956
5	M	-	2%	10%	-	-	1	2	0	Eiró Gonsalves, 1956
9	F	-	2%	10%	-	-	1	1	0	Eiró Gonsalves, 1956
6	M	-	15 ml	15 ml	-	-	1	2	0	Eiró Gonsalves, 1956
4	M	-	0.5	6	-	-	1	3	0	Eiró Gonsalves, 1956
9	F	-	0.5	3	-	-	1	1	0	Eiró Gonsalves, 1956
NS	NS	-	0.2 ml/kg	1	-	-	1	3	0	Espinoza et al, 1982
NS	NS	-	0.2 ml/kg	1	-	-	1	3	0	Espinoza et al, 1982
NS	NS	-	0.2 ml/kg	1	-	-	1	3	0	Espinoza et al, 1982
NS	NS	-	0.2 ml/kg	1	-	-	1	3	0	Espinoza et al, 1982
1.5	F	-	0.12	6.75	-	-	1	2	0	Ruangkan, 1999
Co-EDTA										
7	F	-	-	-	NS	-	1	3	3	Brian, 1990
6	F	-	-	-	NS	-	1	2	2	Brian, 1990
7	F	-	-	-	NS	-	1	1	1	Brian, 1990
HOCO										
NS	NS	-	-	-	-	0.5	1	3	0	Espinoza et al, 1982
NS	NS	-	-	-	-	0.5	1	3	0	Espinoza et al, 1982
NS	NS	-	-	-	-	0.5	1	3	0	Espinoza et al, 1982
NS	NS	-	-	-	-	0.5	1	3	0	Espinoza et al, 1982

Case studies are detailed Appendix C

NS, not stated; -, not applied.

The latency time may vary between 0.5 and 9 hours according to these reports.

Chronic Exposure

Since the 1930s neurological syndromes have been found to be correlated to high cassava ingestion (Moore, 1934a & b) and were initially were attributed to the cyanide content (Clarke, 1936). All these reports came from Nigeria, except for a single case of 2 women from the same family in Liberia (Njoh, 1990) that had 'tropical ataxic neuropathy' (TAN) with optic atrophy, nerve deafness and sensory spinal ataxia. Angular stomatitis, glossitis, and scrotal dermatitis also had been seen. In a comparison of 2 villages one with high and and the other low cassava consumption, both the syndrome and higher plasma thiocyanate levels could be found in the village with the higher consumption (Osuntokun et al, 1969; Oluwole et al, 2000). In the meantime the disease has also been described from Tanzania (Makene and Wilson, 1972). Recently the role of cyanide in the etiology of these syndromes has come under discussion (Oluwole et al, 2002; Oluwole et al, 2004).

Konzo, a myelopathy of abrupt onset that is manifesting as permanent spastic paresis of both legs, has been linked to prolonged high dietary cyanide exposure from ingestion of cassava (Howlett et al, 1990). Konzo was first described in Zaire (Trolli, 1938; cited in Essers et al, 1992), and then further epidemics were reported from Mozambique and Tanzania (Ministry of Health Mozambique, 1984; Casadei et al, 1990; Essers et al, 1992; Cliff et al, 1997). Later affected countries included the Central African Republic (Tylleskär et al, 1994; Mbelesso et al, 2009). The occurrence of acute poisoning symptoms has been reported before the onset of Konzo (Tylleskär et al, 1991). Abortive symptoms of the disease, i.e. an ankle clonus, also have been ascribed to cyanide intake (Cliff et al, 1986). An additional cause of chronic symptoms may be a sulphur deficiency from low intake of sulphur-containing amino acids (Cliff et al, 1985). This has not been elucidated for acute symptoms.

Also, growth retardation in children has been assumed to be caused by chronic cyanide poisoning from cassava (Banea-Mayambu et al, 2000).

Higher thiocyanate and also higher CN^- levels have been found both in cassava processors and in people eating larger amounts of cassava. The blood cyanide levels were below 8 µmol/l (Osuntokun and Monekosso, 1969; Casadei et al, 1990; Okafor et al, 2002). Air concentrations in a processing plant were at a maximum of 10 ppm (Akinrele, 1986), which is significantly above current workplace limit values. Human poisoning cases have not been reported in cassava processing.

Based upon studies in animals (Akanji and Famuyiwa, 1993) it can be assumed that elevated thiocyanate levels provide a good indication of the extent of chronic cassava / cyanide poisoning.

Prolonged elevated thiocyanate levels are responsible for the development of goitre, which is described in cassava consuming populations (Cliff et al, 1986; Bourdoux et al, 1978).

Treatment and/or prophylaxis

Chronic cyanide poisoning from cassava consumption does not require antidotal treatment. Changes in preparation methods lowering the cyanide content can sufficiently abolish the chronic intoxication, while the symptoms will persist. In addition, newer publications seem to indicate that the chronic neurotoxicity may be due to unmetabolised linamarin rather than to cyanide (Mathangi and Namasivayam, 2000). For Konzo the same has been reported (Banea-Mayambu et al, 1997; Sreeja et al, 2003).

The therapeutical application of combinations of hydroxocobalamin-cystine and riboflavin-cystine in a double-blind controlled study in Nigerian ataxic neuropathy failed to show any benefit (Osuntokun et al, 1974). As both cystine (sulphur donor for the detoxification via the thiosulphate route) and HOCO are cyanide binding or detoxicating agents, this casts additional doubt on the role of cyanide in the development of the syndrome.

As acute cyanide poisoning resulting from cassava consumption obviously occurs with a latency time to ingestion, and while the treatment recommendation includes SN, practical experience shows that the use of STS alone is sufficient to terminate the poisoning symptoms and to lead to full recovery (Cheok, 1978).

It remains to be assessed whether unmetabolised linamarin might also play a role for the acute toxicity, in addition to the chronic neurotoxicity syndromes. There may be a transport of linamarin into (brain) cells via a glucose transporter (Sreeja, 2003).

Discussion and conclusion

Since no measurements of blood cyanide have been reported there is no definitive proof that the symptoms observed following poisoning from cassava are directly, or causally related to cyanide intoxication. The diagnosis and the explanatory model are primarily based on assumption, or indirect evidence of very high thiocyanate levels in the general population (Mlingi et al, 1992).

Most cases are insufficiently documented and ranged from very mild to fatal (Cliff and Coutinho, 1995; Mlingi et al, 1992; Essers et al, 1992) and administration of antidote was inneffective (Brian, 1990).

This has consequences regarding recommended therapy, as no antidote with potential side effects should be given. Since cassava poisoning appears to prevail in developing/tropical countries further consideration of cost, availability and practicability will further determine the antidote of choice.

These aspects speak in favour of using STS as antidote in acute cassava poisonings, if any.

5.2.2 Amygdalin (laetrile)

Introduction

Amygdalin is a cyanogenetic glycoside, which occurs naturally as a toxicant in the kernels of bitter almonds, apricots, choke cherries, some other stone fruits and nuts like almonds and macadamia. For example, bitter almond kernels contain many organelles with either amygdalin or ß-glucosidase. When the two come into contact, such as when the kernel is damaged, cyanide is generated. Fatalities may occur from 25-60 kernels depending on the plant variety. Cyanide contents measured have been up to 409 mg/100g wet weight (Herbert, 1979).

The commercial formulation used as an alleged cancer remedy is called Laetrile.

Substance data

IUPAC name:	[(6-O-β-D-Glucopyranosyl-β-D-Glucopyranosyl) oxy] (phenyl) Acetonitrile
Synonyms:	Phenylacetonitril, Amygdalin, Laetrile (Lätril), falsely called Vit. B17
CAS registry number:	29883-15-6
Formula:	$C_{20} H_{27} NO_{11}$
Molecular mass:	457.431
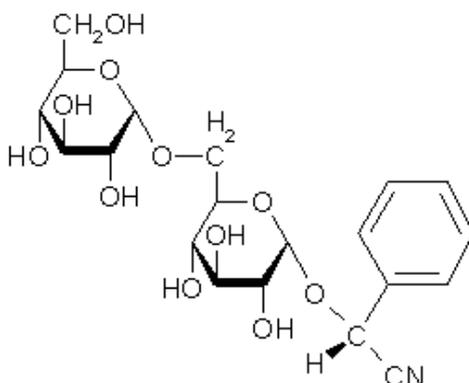
Chemical structure:

Intrinsic Properties (pharmacokinetic and toxicity data)

The kernels of bitter almonds, apricots choke cherries and some other stone fruits and nuts like almonds and macadamia contain both amygdalin and enzymes that can decompose amygdalin. For example, almonds contain a so-called emzulsin complex with 2 ß-glucosidases yielding glucose and mandelonitrile, and a hydroxynitrile lyase catalysing the dissociation of the mandelonitrile to HCN and benzaldehyde. This dissociation also occurs at room temperature without lyase (Schmidt, 1978).

Additionally, the human gastrointestinal tract contains beta-glucosidases, which cause dissociation of amygdalin to cyanide (Kalyanaraman et al, 1983).

Rat experiments showed that cyanide formation from orally administered amygdaline was much higher than from intravenous application, which caused only a slight increase after 8 hours (McAnalley, 1980). Further experiments with SPF (Specific Pathogen Free) and conventional rats showed that gastrointestinal microflora is obligatory for the production of cyanide from amygdalin. Conventional rats given a single dose of 600 mg/kg amygdalin died within 2.5 hours and had blood cyanide levels of 2.6 to 4.5 mg/l (Carter, 1980). This indicates that oral amygdalin application is 40 times more toxic than parenteral application. For humans the role of the intestinal microflora has also been established (Shragg et al, 1982).

Prior to that, the dose-dependent cyanide toxicity resulting from amygdalin intake had been shown in rats. Cyanide levels were at 3.9 mg/l after 750 mg/kg amygdalin (Khandekar and Edelman, 1979).

A literature review and animal experiments showed the time to onset of the first poisoning symptoms to be in the range of 15 to 90 minutes (Ioanid and Bors, 1961).

In dogs the simultaneous application of laetrile, at a dose equivalent to that used for cancer treatment in humans, and sweet almonds caused vomiting, incontinence, respiratory changes up

to respiratory arrest, motor ataxia, coma, convulsions and "bizarre neuromuscular manifestations" as well as 'curious behavioral patterns'.   Six dogs died of acute HCN poisoning, 3 had neurological alterations at the time of sacrifice, and one dog recovered. Cyanide levels were 0.042 to 0.122 mg/l (Schmidt et al, 1978).

Laetrile[®] (capital 'L') is commercially produced and sold as an alleged cancer treatment, also under the names nitrilosides and vitamin B17. Both laetril/amygdalin and Laetrile[®] have one benzaldehyde and one cyanide moiety, which for amygdalin are bound to two glucose molecules, and for Laetrile® to one. Both Laetrile[®] and laetrile/amygdalin are marketed as cancer remedy.

The claim of the promoters is that cyanide in minute quantities and in 'proper form' is an essential component of normal body chemistry (Herbert, 1979).

The potential of laetrile/Laetrile[®] for cancer treatment and the risks have been repeatedly questioned. In rats, amygdalin administered at very high doses of up to 750 mg/kg did not reduce growth of inoculated tumours (Khandekar and Edelman, 1979).

The US FDA in 1977 mailed a warning notice to all physicians and nearly one million health workers in the USA stating that laetrile is worthless as a cancer cure and void of nutritional value, poisonous and possibly contaminated with toxicants and molds. An analysis of commercially available tablets and liquids even found insect legs among other materials in the tablets, and only two thirds of the claimed laetrile contents (Schmidt et al, 1978). A 1982 phase II clinical study by the National Cancer Institute showed no positive effect, but symptoms of cyanide toxicity were seen (Moertel et al, 1982).

A recent review found 3 non-consecutive case series, 2 consecutive case series, 6 best case series, and 25 case reports on the effectiveness or safety of laetrile as cancer treatment. None of these publications proved the effectiveness of laetrile (Milazzo et al, 2007).

Cyanide poisonings from laetrile appear to have been known to ancient Egyptians (Herbert, 1979) and in ancient Rome laetrile was used as a poison to commit suicide.

Casuistics

The 30 cases that allow for an assessment are listed in table 33 below.

Table 33: Poisoning cases with laetrile / amygdalin

Age (y)	Sex	Exposure			Blood cyanide concentration (mg/l)	Cyanide poisoning?	Antidote treatment						Contributive for efficacy?	PSSa	Reference
		Ingested	Total dose (mg)	Latency time (h)			Cumulative dose (g)								
							AN (pearls)	SN	STS	HOCO	O₂	Other			
3	M	Choke cherries	NS	NS	-	Possible, clinical symptoms	-	-	-	-	-	-	-	4	Pardee, 1847
14	F	Choke cherry pits	NS	Days?	-	Probable, clinical symptoms	-	-	-	-	-	-	-	4	Pijoan, 1942
12	NS	Choke cherry pits	NS	NS	-	Possible, clinical symptoms	-	-	-	-	-	-	-	2	Pijoan, 1942
54	NS	Choke cherry pits	NS	NS	-	Possible, clinical symptoms	-	-	-	-	-	-	-	2	Pijoan, 1942
14	NS	Choke cherry pits	NS	NS	-	Possible, clinical symptoms	-	-	-	-	-	-	-	2	Pijoan, 1942
4	F	Peach kernels	NS	0.5	-	Probable, clinical symptoms, bitter almond smell	-	-	-	-	NS	Methylene blue	NS	3	Jeanin and Berrod, 1961
2	F	Apricot pits	NS	Minutes	-	Probable, clinical symptoms	-	-	-	-	NS	-	Yes	2	Yatziv and Simcha, 1969
3.5	F	Apricot pits	NS	NS	-	Probable, clinical symptoms, bitter almond smell	-	-	-	-	-	-	-	NS	Gunder et al, 1969
34	M	48 apricot kernels	NS	1h	-	Possible, clinical symptoms	-	-	-	-	-	-	-	1	Townsend and Boni, 1975
11mo	F	Amygdalin	500, 1 - 5 tbl.	0.5	0.29	Yes	-	NS	NS	-	NS	-	Partial effect only	3, (4)	Humbert et al, 1977; Braico et al, 1979
17.5	F	Laetrile ampoules 10.5 g	NS	10 min	-	Probable, clinical symptoms	-	-	-	-	-	-	-	3, (4)	Sadoff et al, 1978
60	M	Laetrile®	NS	1 h	6	Yes	-	-	-	-	-	-	-	1	Maxwell, 1978
NS	M	15 apricot kernels	NS	1 h	-	Probable, clinical symptoms	-	-	-	-	-	-	-	1	Cal, 1972

Case							Antidote treatment								Reference
		Exposure			Blood cyanide concentration (mg/l)	Cyanide poisoning?	Cumulative dose (g)						Contributive for efficacy?	PSSa	
Age (y)	Sex	Ingested	Total dose (mg)	Latency time (h)			AN (pearls)	SN	STS	HOCO	O$_2$	Other			
NS	F	15 apricot kernels	NS	1 h	-	Probable, clinical symptoms	-	-	-	-	-	-	-	1	Cal, 1972
NS	F	Handful of apricot kernels	NS	1 h	-	Unlikely, no cyanide in blood	-	-	-	-	-	-	-	1	Cal, 1975
3	M	Laetrile® enema,	3 d 3.5 g	'shortly'	2.14	Yes	-	-	-	-	NS	-	Yes	2	Ortega and Creek, 1978
49	F	20 – 40 apricot pits	-	0.5	3.2	Yes	NS	NS	NS	-	-	-	Yes for AN, SN, STS	2	Rubino and Davidoff, 1979
48	F	Laetrile®	-	NS	1.16	Yes	-	-	-	-	-	-	-	1	Morse et al, 1979
NS	NS	Apricot kernels	-	1.5	-	Probable, clinical symptoms, reaction to antidotes	-	NS	NS	-	NS	-	Temporary effect, possibly death due to overdose	2	Lasch and El Shawa, 1981
32	F	Laetrile®	9 g	NS	3.85	Yes	NS	NS	NS	-	-	-	Yes for AN, SN, STS	1	Moss et al, 1981
67	F	12 bitter almonds	-	0.25	-	Yes, cyanide in gastric contents	NS	NS	NS	-	-	-	Yes for AN, SN, STS	3	Shragg et al, 1982
18	M	Laetrile	3 g	1.5	-	Possible, clinical symptoms	-	-	-	-	NS	-	Yes	3	Lee et al, 1982
22	M	Laetrile	1 2 – 18 tablets	NS	-	Probable, clinical symptoms, bitter almond smell	NS	NS	NS	-	NS	-	Yes for oxygen and AN, SN, STS	3	Beamer et al, 1983
4	M	Laetrile	6 g	1.5	16.3	Yes	NS	NS	NS	-	NS	-	Yes for oxygen and SN, STS	3	Hall et al, 1986
65	F	Amygdalin	6 g i.v. 3 g p.o.	2	0.23	Yes	-	NS	NS	-	NS	-	Yes for oxygen and SN, STS	3	Leor et al, 1986
56	F	Steeped choke cherries	-	12	-	Yes, cyanide in food	-	-	-	-	-	-	-	3	Pentore et al, 1996

Case						Antidote treatment							Reference
Age (y)	Sex	Exposure			Blood cyanide concentration (mg/l)	Cyanide poisoning?	Cumulative dose (g)						Contributive for efficacy?	PSSa	
		Ingested	Total dose (mg)	Latency time (h)			AN (pearls)	SN	STS	HOCO	O$_2$	Other			
41	F	30 apricot kernels	-	20 min	1.07	Yes	NS	NS	NS	-	-	-	Yes	3	Suchard et al, 1998
32	F	6 amygdalin	-	'shortly'	-	Probable, thiocyanate in blood	-	-	-	-	NS	-	Yes	1	O'Brien et al, 2005
68	F	Amygdalin	3 g	0.5	-	Probable, clinical symptoms	-	-	-	NS	NS	-	Yes, for oxygen and HOCO	2	Bromley et al, 2005
51	F	Amygdalin	2.5 g	NS	0.4	Yes	-	-	-	NS	NS	-	No for oxygen, yes for HOCO	2	Martinelli et al, 2008

The cases are detailed in Appendix C
NS, not stated; -, not applied.

Table 34: Laetrile poisonings extracted from the PSSa database (appendix B)

Case			Antidote treatment									Reference
Age (y)	Sex	Type of poisoning	Cumulative dose of antidote (g)					Number of treatment steps	PSSa			
			AN (pearls)	SN	STS	DMAP	HOCO		Before	After		
STS alone												
51	M	Laetrile	-	-	25	-	-	1	0	0		Yeh, 1992
STS and nitrites												
35	F	Laetrile	-	NS	NS	-	-	2	3	4		Humbert, 1977; Braico, 1979
32	F	Laetrile	NS	0.3	0.05	-	-	1	2	0		Moss, 1981
4	M	Laetrile	NS	0.6	12.5	-	-	2	3	2		Beamer, 1983
59	M	Laetrile	NS	0.15	6.25	-	-	2	3	0		Hall, 1986
NS	NS	Laetrile	NS	-	NS	-	-	1	3	0		Hall, 1987
17	F	Apricot seeds	NS	0.3	12.5	-	-	1	2	0		Rubino, 1979
NS	NS	Apricot kernel	-	NS	NS	-	-	1	1	4		Lasch and El Shawa, 1981
18	F	Bitter almond	-	0.3	12.5	-	-	1	3	0		Shragg, 1982
56	F	Amygdalin	-	0.6	NS	-	-	1	3	0		Leor, 1986
41	F	Apricot kernel	-	0.45	> 25	-	-	3	3	0		Suchard, 1998
STS and DMAP												
26	F	Bitter almonds	-	-	15	0.5	-	1	1	1		Werner, 1979
HOCO												
68	F	Amygdalin	-	-	-	-	5	1	3	0		Bromley, 2005
51	F	Amygdalin	-	-	-	-	5	1	3	0		Martinelli, 2008

Case studies are detailed in Appendix C

NS, not stated; -, not applied.

Summary

Amygdalin/laetrile can lead to cyanide poisoning. In contrast to cassava poisonings the majority of cases progressed rapidly to severe poisonings within two hours or less. The severity of poisonings in most cases prompted antidote application – usually SN and STS, in single cases also HOCO. As the cyanide poisoning may occur very quickly and with sudden onset STS, though indicated in all cases, may not be sufficient for antidotal therapy. SN has been used successfully and the same can be expected for other directly acting cyanide antidotes (e.g. 4-DMAP, and HOCO).

5.2.3 Nitriles

Introduction

Secondary cyanide poisoning can occur during intoxications with aliphatic and olefinic nitriles.

The majority of exposure to nitriles is in the industrial setting. Non-occupational acute exposures may occur on rare occasions, such as the railroad accident involving loss of containment of acrylonitrile in Germany in February 2002 (unpublished) and via natural sources, such as burning of nitrogen containing biomatter.

In industry, laboratories and research acetonitrile is very commonly used, propionitrile is occasionally used as a solvent, whereas acrylonitrile is used in the production of acrylic fibre, ABS (acrylonitrile-butadiene-styrene) / SAN (Styrene-acrylonitrile) plastics, carbon precursors, NB (Nitrile-Butadiene) copolymers, adiponitrile and acrylamide (PCI, 2010). Reports in literature on nitrile poisoning are scarce, and do not deal with aspects of metabolism, cyanide levels, and treatment options. It is evident that there is a lack of data and guidelines concerning such poisonings not only in occupational medicine, but also in clinical toxicology.

A first report on animal experiments dealt with acetonitrile, propionitrile, acrylonitrile, n-butyronitrile, malononitrile, succinonitrile and acetone cyanohydrin (ACH). Only ACH inhibited the cytochrome *c* oxidase activity *in vitro* reflecting its rapid chemical decomposition to HCN while the others require metabolism. STS protected test animals against poisoning with all of these nitriles, whereas SN is reported to be effective in poisonings with acrylonitrile, n-butyronitrile, malononitrile, and ACH. HCN could be found in the tissues of the test animals and decreased after STS application. The toxic effects of the nitriles are ascribed to *in vivo* HCN liberation as a result of microsomal hydroxylation at the alpha-methylene group to form an unstable cyanohydrin (Willhite, 1980). These and other authors studied the mechanisms of toxicity. Cyanide formation is definitely implicated as the critical mechanism as although the signs of aliphatic nitrile intoxication are qualitatively similar to cyanide intoxication and both

cyanide and thiocyanate can be found in tissues after nitrile poisoning, the time course of nitrile poisoning is delayed reflecting the requirement for metabolic formation of the active substanceg. It has been confirmed that none of the aliphatic nitriles are intrinsically capable of inhibiting cytochrome *c* oxidase (Willhite, 1981; Ahmed and Farooqui, 1982).

This research also revealed that cyanide and thiocyanate concentrations in tissues were lowest and transient for acetonitrile, whereas moderate amounts could be found for propionitrile and acrylonitrile (Willhite and Smith, 1981; Ahmed and Farooqui; 1982).

In male rats acetonitrile was found to induce a much lower thiocyanate excretion in urine than the other nitriles. In addition, there was a clear difference between oral and intraperitoneal administration with the latter leading to a much lower thiocyanate excretion. This indicates that cyanide formation will be higher following oral ingestions of acetonitrile. For comparable doses of propionitrile the level of thiocyanate excretion is 6-10 times higher than for acetonitrile with no clear difference between oral and intraperitoneal administration. Acrylonitrile leads to an intermediate level of thiocyanate excretion after oral ingestion, and a low excretion after intraperitoneal injection (Silver et al, 1982).

Another group (Johannsen and Levinskas, 1986) studied the relationship between toxicity and structure of aliphatic nitriles.

The toxicity to rats of these nitriles is classified in Table 35.

Table 35: Acute toxicity of nitriles to rats

Route / Nitrile	Lethality	Classification
Oral	LD_{50} (mg/kgbw)	
Acetonitrile	2,440	Slightly toxic
Propionitrile	39	Highly toxic
Acrylonitrile	100	Moderately toxic
Dermal	LD_{50} (mg/kgbw)	
Acetonitrile	3,900	Slightly toxic
Propionitrile	40	Highly toxic
Acrylonitrile	20	Highly toxic
Inhalation	LC_{50} (mg/l), 4 h	
Acetonitrile	35.0	Practically non-toxic
Propionitrile	1.7	Moderately toxic
Acrylonitrile	1.4	Moderately toxic

Acetonitrile

Introduction

Acetonitrile is a very common solvent used by industry and especially in laboratories. It has been used in consumer products, such as nail polish remover in the past, but this is now banned in Europe and in the USA.

Substance data

Common Name:	Acetonitrile
Synonyms:	Cyanomethane, methyl cyanide or ACN
CAS registry No.:	75-05-8
Formula:	CH_3CN
Molecular mass:	41.05
Structure:	

$$H-\overset{\displaystyle H}{\underset{\displaystyle H}{C}}-C\equiv N$$
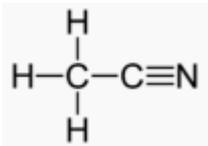
Physical State:	Clear liquid
Odour:	Ether-like
Vapor pressure:	74 mmHg at 20°C
Water solubility:	Infinitely soluble
Log K_{ow}:	-0.34
Conversion factor:	1 ppm = 1.68 mg/m³, 1 mg/m³ = 0.595 ppm (25 °, 760 mm Hg).

Nitriles in general are hydrolysed in the presence of acidic conditions to form amides. Although ACN is one of the more stable nitriles, acidic hydrolysis would be expected to yield hydrogen cyanide. Hydrolysis in water has been reported as extremely slow (WHO, 1993). Willhite (1983) found that freshly prepared solutions of ACN in distilled water did not undergo any significant hydrolysis upon incubation at 37°C for 2.5 hours (EPA, 1999).

Absorption

Like hydrogen cyanide (HCN), ACN is readily absorbed from the lungs and gastrointestinal tract, and is distributed throughout the body in both humans and laboratory animals. Absorption of ACN after inhalation of tobacco smoke is estimated to be 91% (Dalhamn et al, 1968a,b). Absorption from the gastrointestinal tract is very effective and dermal absorption is assumed to be effective (IPCS, 1993b).

Toxicity

The toxicity of ACN is believed to be mediated, in part, through delayed production of cyanide (see above) and production of cytotoxic anoxia (Albaum et al, 1946) although the conversion occurs slower than for other nitriles (Swanson and Krasselt, 1994; Mueller and Borland, 1997; Anonymous, 2001), which is due to the slow conversion of acetonitrile to cyanide with a half-life of 40 hours (Mueller and Borland, 1997) and the time elapsing until depletion of detoxification substrates (Swanson and Krasselt, 1994), and which may explain the delay in onset of acute symptoms. The conversion to cyanide may be oxygen- and NADPH-dependent, possibly mediated by P450 isozyme (2E1 or P-450j) (Freeman and Hayes, 1988) or by producing cyanohydrin through a P450 reaction, which is then decomposed by catalase to release cyanide (Ahmed et al, 1992; Feierman and Cederbaum, 1989; Willhite and Smith, 1981). Formaldehyde and formic acid are also postulated to be by-products of ACN metabolism (Ahmed et al, 1992).

It has been shown, that treatment either with ethanol or 4-methylpyrazole can inhibit the P450 2E1 pathway and may in future find a place in treatment of acetonitrile poisoning, and might reduce or abolish the need for treatment with cyanide antidotes (Kedderis and Strother, 2007).

There are difference between inhalation and ingestion. Volunteers exposed to 40, 80, and 160 ml/m³ acetonitrile vapours for 4 hours showed no measurable cyanide levels (Pozzani et al, 1959). Cyanide levels have been reported to peak after 25-59 hours (Turchen et al, 1989; Losek et al, 1991; Boggild et al, 1990). Similar though shorter delays of the occurrence of more severe symptoms have been reported several times (Kurt et al, 1991; Losek et al, 1991; Mueller and Borland, 1997). There is also a report on recurrences of severe symptoms over more than 40 hours after ingestion (Turchen et al, 1991). The latency times until onset of first symptoms vary between 20 minutes and 12 hours.

Significant interspecies differences have also been found in animal experiments with mice being more sensitive than rats (WHO, 1993).

In humans the intoxication is described as being very similar, or almost identical, to cyanide poisoning with oppression, restlessness, tachycardia, dizziness, headache, nausea, vomiting, drop in blood pressure, metabolic acidosis, pulmonary oedema, loss of conscience, seizures and respiratory arrest.

Casuistics and case series

The reports of dermal and inhalation exposures are summarised in table 36:
The assessment of the cases shows a latency time to onset of symptoms of 2-15 hours.

Table 36: Acetonitrile inhalations

Case					Antidote treatment								PSSa		Reference
			Maximum concentration (mg/l)		Antidote						Remark				
Age (y)	Sex	Latency time (h)	Acetonitrile	Cyanide	AN (pearls)	SN	STS	Co-EDTA	HOCO	Other			Before	After	
23	M	3.5		7.96	-	-	-	-	-	-	Found dead		4		Amdur, 1959
35	M	12		3.06	-	-	Yes	-	-	-			3	0	
28	M	NS		10.88	-	-	Yes	-	-	-			2	0	
28	M	12		0.72	-	-	-	-	-	-			0		
20	M	NS		0.58	-	-	-	-	-	-			0		
18	M	7		0.33	-	-	-	-	-	-			1		
42	M	12		0	-	-	-	-	-	-			1		
25	M			0	-	-	-	-	-	-			0		
19	M	12	11.8	1.12	-	-	-	Yes (0.6 g)	-	-			3	3	Dequidt et al, 1974
					-	-	-	-	Yes (4 g)	-			3	4	
2	M	9.5		6	-	-	-	-	-	-			2		Caravati and Litowitz, 1988
28	M		6.9	<LOD	-	-	Yes	-	-	NAC			0		Steffens et al, 1998; Steffens, 2002
35	M	15			-	Yes	Yes	-	-	-			3	3	Muraki et al, 2001
27	M	6			-	-	-	-	-	-			1		Tsutaoka et al, 2003
22	M				-	-	-	-	-	-			0		
23	M	2			-	-	-	-	-	-			1		
52	M	2			-	Yes	Yes	-	-	-			3	0	Zavotsky et al, 2004

Case studies are detailed Appendix C.

NS, not stated; -, not applied.

In case of oral ingestions the formation of cyanide occurs regularly. Therefore ingestion seems to differ from inhalation. Relatively many cases have been described (Jaeger et al, 1977; Caravati and Litowitz, 1988; Turchen et al, 1989; Boggild et al, 1990; Geller et al, 1991; Kurt et al, 1991; Losek et al, 1991; Michaelis et al, 1991; Turchen et al, 1991; Jones et al, 1992; Swanson and Krasselt, 1994; Mueller and Borland, 1997), though not all with biomonitoring data for acetonitrile and cyanide or with doses of antidotes given. One report describes an initial cyanide level of 3.13 mg/l in blood (8 hours after ingestion), but a maximum of 12.8 mg/l after 59 hours (Turchen et al, 1989), another one a level of 2.1 mg/l 12 hours and 3.8 mg/l 25 hours after ingestion (Losek et al, 1991). In one case of an ingestion of both acetonitrile and acetone mild symptoms developed after 8 hours, while severe symptoms and cardiac arrest occurred after 26 hours, death after 30 hours (Boggild et al, 1990). Similar though shorter delays of the occurrence of more severe symptoms have been reported several times (Kurt et al, 1991; Losek et al, 1991; Mueller and Borland, 1997). There is also a report on recurrences of severe symptoms over more than 40 hours after ingestion (Turchen et al, 1991). The latency times until onset of first symptoms vary between 20 minutes and 12 hours.

Regarding antidote applications some reports show a clear beneficial effect for the Lilly Kit and oxygen, or for STS alone and oxygen (Geller et al, 1991; Kurt et al, 1991; Losek et al, 1991), while the application of all available antidotes in other cases could not terminate the coma (Jaeger et al, 1977).

Ingestion cases are summarised in Table 37.

Table 37: Acetonitrile ingestions

Case					Antidote treatment						Remark	PSSa		Reference
Age (y)	Sex	Latency time (h)	Maximum concentration (mg/l)		Antidote									
			Acetonitrile	Cyanide	AN (pearls)	SN	STS	DMAP	Co-EDTA	HOCO		Before	After	
26	M	3	NS	NS	-	Yes	Yes	-	Yes	Yes		3	0	Jaeger et al, 1977
1.3	M	0.3	NS	3.1	-	-	-	-	-	-	Found dead	4		Caravati and Litowitz, 1988
39	F	7.3	NS	3.13	-	Yes	Yes	-	-	-		3	1	Turchen et al, 1989; 1991
22	F	8	NS	NS	-	-	-	-	-	-		3(4)		Boggild et al, 1990
3	M	13	NS	1.24	-	-	Yes	-	-	-		2	0	Geller et al, 1991
2	F	12	NS	1.82	Yes	Yes	Yes	-	-	-		3	0	Kurt et al, 1991
2	M	6	NS	2.1	Yes	-	-	-	-	-		2	2	Losek et al, 1991
					-	-	Yes	-	-	-		2	0	
30	M	5	80	7.1	-	-	Yes	Yes	-	-		1	1	Michaelis et al, 1991
					-	-	Yes	-	-	-		1	0	
48	M	NS	800	2.4	-	-	-	-	-	-	Found dead	4		Jones et al, 1992
53	F	NS	770	4.5	-	-	-	-	-	-	Found dead	4		Jones et al, 1992
39	F	NS	56	4.4	-	-	-	-	-	-	Found dead	4		Swanson and Krasselt, 1994
39	F	0.5	640	1.7	-	Yes	Yes	-	-	-		3	3	Mueller and Borland, 1997
					-	Yes	Yes	-	-	-		2	0	

Case studies are detailed Appendix C.

NS, not stated; -, not applied.

More details on the casuistics can be found in the casuistics chapter.

Table 38: Acetonitrile poisonings extracted from the PSSa database (Appendix B)

Case		Antidote treatment										Reference
Age (y)	**Sex**	**Cumulative dose of antidote (g)**						**Number of treatment steps**	**PSSa**			
		AN (pearls)	**SN**	**STS**	**DMAP**	**Co-EDTA**	**HOCO**		**Before**	**After**		
STS alone												
28	M	-	-	NS	-	-	-	1	3	0	Amdur, 1955	
28	M	-	-	NS	-	-	-	1	2	0	Amdur, 1955	
31	M	-	-	8.75	-	-	-	1	2	0	Gellier, 1991	
52	M	-	-	1	-	-	-	1	0	0	Steffens, 1998	
STS and nitrites												
39	F	NS	0.099	42.5	-	-	-	1	3	0	Kurt, 1991	
NS	M	1	-	8.25	-	-	-	2	2	0	Losek, 1991	
34	F	-	> 1.5	100	-	-	-	2	3	0	Mueller, 1996	
4	F	-	0.6	62.5	-	-	-	4	3	0	Turchen et al, 1991	
STS and DMAP												
30	M	-	> 1	-	0.25	-	-	2	1	0	Michaelis, 1991	
Co-EDTA and HOCO												
19	M	-	-	-	-	0.6	4	2	3	4	Dequidt, 1974	
HOCO												
39	M	-	-	-	-	-	5	1	2	0	Baud, unpublished	

Case studies are detailed Appendix C.

NS, not stated; -, not applied.

From Germany there are reports on a total of 10 poisoning cases in 9 individuals. 8 of these cases involved inhalation and 2 dermal exposure. Significant and potentially life-threatening cyanide poisoning was seen in cases of ACN inhalation, while the 2 dermal cases did show elevated ACN levels, cyanide was not. Symptoms included nausea / vomiting and somnolence in the more severely poisoned cases and irritation of the conjunctives and the throat. 8 cases were treated with NAC as acrylonitrile antidote, 5 of them additionally received STS (Vogel et al, 1984). *Comment: Interestingly, in 2 cases the application of NAC alone caused a significant drop in blood cyanide (Steffens et al; 1998; Steffens et al, 2001; Steffens, 2002). These cases have partly been described and cited by other authors.*

An industrial accident involving acetonitrile inhalation and possibly dermal uptake occured while resin coating a tank with acetonitrile as resin thinner. 8 workers were hospitalised, 5 more were seen as outpatients. Cyanide levels several hours after the accidents were 0.33 to 10.88 mg/l. One of the hospitalised workers died (cyanide level 7.96 mg/l) (Grabois, 1955; Amdur, 1959). *Comments: Neither the exposure times nor the thiocyanate levels in serum were in consistent relation to the cyanide levels. Acetonitrile levels were not reported.*

Treatment / respiratory prophylaxis

Antidotal treatment is targeted at the cyanide formed.

Oxygen, STS, AN and SN, as well as DMAP, have been used successfully alone or in combinations (see tables 36 and 37). Some reports show a clear beneficial effect for the Lilly Kit and oxygen, or for STS alone and oxygen (Geller et al, 1991; Kurt et al, 1991; Losek et al, 1991).

There is a description of a death of a worker who was cleaning with acetonitrile and hot water and inhaled ACN despite administration of Co-EDTA (2 x 600 mg) and HOCO (4 g). First symptoms developed after about 12 hours, cyanide in blood was 11.2 mg/l, maximum acetonitrile in blood 311 mg/l 24 hours after hospitalisation (Dequidt et al, 1974). Retrospectively the antidotes were probably given too late, more than 24 hours after onset of symptoms. In another case the efficacy of these antidotes together with STS and SN is unclear in regard of prolonged coma, which could not be terminated by antidote applications (Jaeger et al, 1977).

A further dermal exposure case has been reported. A worker was splashed on his face, neck and chest. Under the assumption of cyanide involvement he received STS (10 ml 10%) twice and was started on an N-acetylcysteine (NAC)-infusion, which caused an allergic reaction and had to be stopped. The maximum acetonitrile concentration in blood in a whole series of biomonitoring measurements was 6.9 mg/l, while cyanide could not be detected at any point in time. Either there had been no cyanide formation, or any latent formed cyanide had been efficiently captured by the STS (Steffens et al, 1998; Steffens, 2002).

Discussion and conclusion

Ingested acetonitrile will lead to cyanide formation with a relatively long latency. Particularly in the case of ingestion there may be a delay of up to 24 hours before severe symptoms start to appear. They can, in some cases, become life-threatening and require a specific therapy. However, it is not absolutely certain that inhalation or skin contact with acetonitrile will always lead to cyanide formation, as demonstrated by clinical studies, albeit with low concentrations of acetonitrile in air, did not result in cyanide formation.

The application of SN and STS, of DMAP and STS, and of STS alone; but all with oxygen, have been shown to be effective.

Propionitrile

Introduction

Propionitrile is use in chemical syntheses and to a lesser extent as solvent in laboratories so cases of poisoning are scarce (n = 4).

Substance data

Common Name:	Proprionitrile
IUPAC name:	Propanenitrile
CAS registry No.:	107-12-0
Formula:	C3H5N
Molecular mass:	55.08
Structure:	H–C–C–C≡N (with H, H on top and H, H on bottom)
Physical State:	Colourless liquid
Odour:	ether-like
Vapor pressure:	39 mm Hg at 20°C
Water solubility:	93.380 g/L at 25°C.
Log K_{ow}:	0.16
Conversion factor:	1 ppm = 2.253 mg/m³, 1 mg/m³ = 0.444 ppm (25 °C, 760 mm Hg)
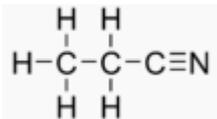
Intrinsic Properties (pharmacokinetic and toxicity data)

Little information is available on pharmacokinetics. Absorption via all routes is assumed to be good (GESTIS)[a].

The latency time to symptoms after inhalation ranged from during exposure to 7 hours later.

Casuistics and case series

The reported propionitrile and cyanide levels from the 4 cases are given in table 39.

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[a] GESTIS – Stoffdatenbank: Propionitil.
http://gestis.itrust.de/nxt/gateway.dll?f=templates&fn=default.htm&vid=gestisdeu:sdbdeu, Acc. Oct 7, 2011.

Table 39: Propionitrile poisonings

Case					Antidote treatment						Reference
			Maximum concentration (mg/l)		Antidote				PSSa		
Age (y)	Sex	Latency time (h)	Propionitrile	Cyanide	AN (pearls)	SN	STS	HOCO	Before	After	
55	M	<0.3		5.71	-	-	Yes	Yes	2	0	Baud et al, 1986; Bismuth et al, 1987
28	M	During exposure		5	-	Yes	Yes	-	3	1	Scolnick et al, 1993
34	M	During exposure		3.5	-	Yes	Yes	-	2	0	Scolnick et al, 1993
52	M	1	12.74	3.15	-	-	Yes	-	2	0	Steffens et al, 1998; Steffens, 2002

Case studies are detailed Appendix C

-, not applied

Table 40: Propionitrile poisonings extracted from the PSSa database (Appendix B)

Case		Antidote treatment					PSSa		Reference
		Cumulative dose of antidote (g)				Number of treatment steps	PSSa		
Age (y)	Sex	AN (pearls)	SN	STS	HOCO		Before	After	
STS alone									
52	M	-	-	2	-	1	2	0	Steffens, 1998
STS and nitrites									
28	M	-	0.3	12.5	-	1	3	1	Scolnick, 1993
34	M	-	0.3	12.5	-	1	2	0	Scolnick, 1993
STS and HOCO									
3	M	-	-	8	4	1	2	0	Baud, 1986; Bismuth, 1987

Case studies are detailed Appendix C.

-, not applied.

Treatment / resp. prophylaxis

STS alone and combinations of STS with direct cyanide antidotes like HOCO or SN have been shown to be effective in propionitrile poisoning.

Discussion and conclusion

In propionitrile poisoning cyanide intoxication seems to occur with a shorter latency time which means that patients may already be in a severe or critical state upon presentation. While STS has successfully been used to date a direct cyanide antidote like SN or HOCO should be considered.

Acrylonitrile

Introduction

Acrylonitrile consists of a vinyl group linked to a nitrile which means that in addition to the inherent toxicity of the vinyl group it can give rise to significant cyanide poisonings. Cyanide formation is dependant on enzyme polymorphisms and on metabolic pathway overload.

Substance data

Common Name:	Acrylonitrile
IUPAC name:	2-propenenitrile
Synonyms:	cyanoethene, vinylcyanide, ACN
CAS registry No.:	107-13-1
Formula:	C3H3N
Molecular mass:	53.06
Structure:	
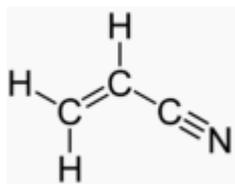
Physical State:	Colourless liquid
Odour:	Pungent, onion- or garlic-like odour
Water Solubility:	70 g//L
Log Pow:	0.11 @ 25ºC (CITI, 1996)

Intrinsic Properties (pharmacokinetic and toxicity data)

The uptake via inhalation has been determined as 52% (Jakubowski et al, 1987) and dermal uptake is up to 0.066 mg/cm²/min (Bakker et al, 1991). Only a small amount is exhaled or excreted via the urine after inhalation (Sato et al, 1975). The enzymatic metabolism occurs mainly in the liver.

For olefinic nitriles like acrylonitrile the pathophysiological mechanisms are more complex than for aliphatic nitriles (Farooqui and Mumtaz, 1991; Bolt and Lewalter, 1994; ATSDR, 1997).

Acrylonitrile is acutely toxic and intoxications with relatively small amounts, e.g. inhalation exposure to 16 to 100 ppm over 20 to 45 minutes caused central nervous effects including nausea, headache, fatigue, irritability and vertigo (Grahl, 1972). In test animals acute poisoning resulted in a biphasic course. Initially and rapidly there is cholinomimetic neurotoxicity with salivation, lacrimation, polyuria, miosis, vasodilatation, increased gastric secretion and diarrhoea, followed by tremors and seizures about 100 minutes after exposure (Benz and Nerland, 2005). This first phase seems to indicate involvement of the cholinergic system (acetyl cholinesterase) inhibition (Buchter et al, 1984, Ghanayem et al, 1991). In fact one publication describes increased acetylcholine levels (Ageeva, 1970 cited by Buchter et al, 1984). However, this publication did not assess the blood acetylcholinesterase levels and cholinesterase levels were not rasied in the 2-generation inhalation study in rats (Nemec et al. 2008). It is interesting, that atropine, but not obidoxime, has been found to show an at least partial antagonistic effect to acrylonitrile poisonings in rats (Buchter et al, 1984; Ghanayem et al, 1991). It has been proposed that the unconsciousness and Kussmaul-type breathing observed after acrylonitrile poisoning are related to formation of cyanide (Paulet and Desnos, 1961). Since CYP2E1-mediated oxidative metabolism has been shown to be a prerequisite for cyanide formation (Wang et al, 2002) and studies with inhibitors of the oxidative metabolism of AN to cyanide have been shown to abolish the early seizure activity, it has been suggested that this first phase is due to cyanide and not the parent molecule (Benz and Nerland, 2005). About 3-4 hours later a second phase of toxicity is seen with depression, clonic convulsions, and respiratory failure and arrest, paralysis and death. Since the development of convulsions and death are not abated, and CNS depression only partially attenuated, with oxidative these inhibitors, this latter phase are regarded as being due to the intrinsic toxicity of acrylonitrile (Benz and Nerland, 2005).

There are species differences in regard to the toxicity to acrylonitrile, with neurotoxic symptoms being typical for the rat, but lacking altogether in the mouse (WHO, 1983; Buchter et al, 1984; BUA, 1994), in which there are also gender differences with greater sensitivity of males (Chanas et al, 2003). There are also apparently species differeneces with regard to carcinogenicity since epidemiological studies did not show hints towards a carcinogenic effect while it is a multisite carcinogen in animal (Anonymous, 2001). IARC has classified acrylonitrile in Group 2B ("possibly carcinogenic in humans").

Acrylonitrile can lead to a significant bioavailability of cyanide in man (Dudley and Neal, 1942; Wilson and McCormick, 1949; Brieger et al, 1952).

Usually the liberation of cyanide increases with the acrylonitrile-level in the organism (Kedderis and Batra, 1993; Bolt and Lewalter, 1994). Acrylonitrile can be metabolised along different pathways, but only few pathways will lead to cyanide formation (fig. 1).

Figure 12: Metabolism of acrylonitrile[a] **(adapted from Thier et al, 2000)**
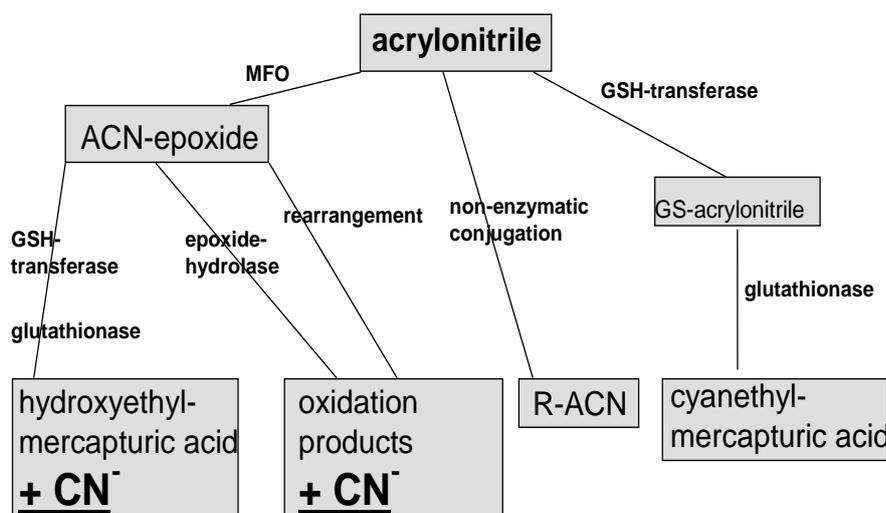
[a] GS = glutathione, GSH = reduced glutathione, R = Protein, MFO = mixed-functional oxidases

The importance of the different pathways depends first on genetic polymorphisms of enzyme status, and second on the amount of acrylonitrile to be metabolised and a possible relative insufficiency of the main pathway (Bolt and Lewalter, 1994, Thier et al, 2000). When the detoxification mechanisms via GSH-acrylonitrile-conjugates become insufficient, the metabolism via mixed-functional oxidases (MFO) will produce acrylonitrile-epoxides as intermediates and finally cyanide (Ahmed and Patel, 1981; Schaefer and Maurer, 1993; Bolt and Lewalter, 1994; ATSDR, 1997). Additionally, there are significant species differences, also resulting in different oral acute toxicity values (Ahmed and Patel, 1981; Gut et al, 1981; Woutersen, 1998), with e.g. the oxidative pathway resulting in cyanide formation being much more important in man than in the rat (Thier et al, 2000). The epoxide pathways accounts for 50% of the metabolites in the rat, but for 80% in the mouse (Fennel et al, 1991). While liver microsomes of mice show greater capacity to produce the epoxide, the mouse has lower levels of the epoxide in circulation, presumably due to an even higher detoxification capacity in the liver (Kedderis, 1993).

Its metabolites have also been shown in exposed workers (Lewalter, 1991).

In terms of antidotal treaments, as early as the 1960s it was recorded that cyanide appeared in the blood of test animals poisoned with acrylonitrile and that the cyanide vanished or was greatly diminished after pre-treatment with STS, but the toxic effects were not effectively abolished. This could only be accomplished by a pre-treatment with cysteine (Gut et al, 1981; Hashimoto and Kanai, 1985). An explanation is the inhibition by acrylonitrile of glyceraldehyde-3-phosphate dehydrogenase (Campian and Benz, 2008).

The significance of cyanide in acrylonitrile poisonings has been denied (Schwaneck, 1966) and toxicological books stress the importance of an intrinsic acrylonitrile toxicity, which is not only due to cyanide formation (Loeser, 1994). Therapeutical regimens thus stress the blocking of effects on the acrylonitrile-receptor by using substances with free SH-groups like N-acetyl-cysteine (NAC) (Buchter and Peter, 1984; Buchter et al, 1984; McLaughlin et al, 1985; van Heijst, 1986; Benz et al, 1990; US Department of Health and Human Services, 1997; Benz, 1998).

The application of cyanide antidotes also has been widely recommended with a special emphasis on sulphur donors in order to increase the cyanide metabolism capacity.

Symptoms of acrylonitrile poisoning in case of dermal contact in man include a burning of the skin, possibly followed by blisters one day later (WHO, 1983). Other symptoms in man have not been described so far (Anonymous, 2001) although AN does display significant systemic toxicity by dermal contact in animals (rat 4-hr dermal LD_{Lo}~200 mg/kg; rabbit 24-hr dermal LD_{50}<200 mg/kg compared with rat oral LD_{50}~80 mg/kg; rabbit oral LD_{50} ~90 mg/kg (SNF, 2005; Vernon et al. 1990; ECB, 2004 respectively). Thus the following casuistics are of interest also in regard of symptoms of dermal acrylonitrile poisoning in man. For inhalation exposure symptoms of headaches, upper airway and eye irritation up to lacrimation and visual problems, insomnia, nervousness, irritability, nausea, vomiting, weakness, mild jaundice, mild anaemia and leukocytosis have been reported (US-NAC, 2007).

Casuistics

Cases of acrylonitrile poisoning have been described several times.

It is noteworthy that there are some Russian publications (in Russian only) which could not be assessed including one of acute poisoning with associated damage to the nervous system, and 4 on antidotes focussing on cysteine.

From China a series of 144 cases has been reported, from which the following table of symptoms occurring in more than 50% of the cases is derived (Table 41). Most patients (102/144) had been exposed to air concentrations of more than 100 mg/m³ for 0.2 to 1.0 hours

and developed symptoms with a latency time of 0.25 to 1.0 hours. All cases survived. Blood levels of acrylonitrile or cyanide are not reported.

Table 41: Symptoms in more than 50% of cases (n = 144) **(Chen et al, 1991)**

Symptom	Occurrence (%)
Dizziness	100
Headache	100
Feebleness	100
Chest tightness	100
Knee jerk	95.1
Nausea	92.4
Dyspnoea	81.9
Pallor	75
Congestion of pharynx	72.9
Fainting	72.2
Abdominal pain	67.6
Vomiting	66
Profuse diaphoresis	66
Sore throat	60.4

Therapy consisted of AN inhalation, 10 ml SN 3% i.v. followed by 20-60 ml STS 50% i.v. According to these authors short time inhalation of high concentrations will affect the central nervous system, while prolonged inhalation of moderate concentrations of acrylonitrile will affect the liver. In severely poisoned victims damage to the heart muscle is described. The toxic effects are ascribed to cyanide formation and resulting cytotoxic hypoxia, to direct inhibition of the respiratory centre by acrylonitrile itself, to reaction with thiol-group enzymes leading to metabolic disturbances, to combination with neurotransmitters like histame, 5-HT, dopamine, norepinephrine (Chen et al, 1991).

An overview of reported cases and case series is given in Table 42. Cyanide levels have been measured only in one case series and on 3 single cases, and were significantly elevated only on 3 patients.

Table 48: Cases of acrylonitrile poisonings

Case					Antidote treatment								Reference
			Maximum concentration (mg/l)		Cumulative dose of antidote (g)						Contributive?	PSSa	
Age (y)	Sex	Latency time (h)	Acrylonitrile	Cyanide	AN (pearls)	SN	STS	HOCO	O₂	NAC			
3	F	During exposure			-	-	-	-	-	-	-	4	Grunske, 1949
10	F *	During exposure			-	-	-	-	-	-	-	4	Lorz, 1950
NS	M**	Minutes			NS	-	-	-	-	-	No	4	Van Luijt, 1963
NS	M**	Minutes			-	-	-	-	-	-	-	1	Van Luijt, 1963
NS	M**	Minutes			-	-	-	-	-	-	-	1	Van Luijt, 1963
NS	M**	Minutes			-	-	-	-	-	-	-	1	Van Luijt, 1963
22	M	0			-	-	-	-	-	-	-	2	Sartorelli, 1966
35	M	15			-	-	-	-	-	-	-	1	Orušev et al, 1972
24	M*	NS		0.184	-	NS	NS	NS	NS	-	Yes, but temporary for SN/STS, unclear for oxygen and HOCO	2	Vogel et al, 1984
48	M	0.5	0.1	3.1	-	-	-	-	-	NS	Yes	1	Steffens, 2002
51	M	0.5	0.5	3.4	-	-	NS	-	-	NS	Recovery		Steffens, 2002
32	M	4		0.5	-	-	-	-	-	NS	Unclear, no significant cyanide poisoning	1	Steffens, 2002
35	M	4	0	0.39	-	-	-	-	-	NS	Unclear, no significant cyanide poisoning	1	Steffens, 2002
37	M	0.5	0.88	0.35	-	-	-	-	-	-	-	1	Steffens, 2002
54	M	0.5	0.9	0.38	-	-	-	-	-	-	-	0	Steffens, 2002
29	M	During exposure	0.82	4.3	-	-	NS	-	-	NS	Yes	1	Steffens, 2002
56	M*	Minutes	6.45	0.1	-	-	NS	-	-	NS	Unclear, no significant cyanide poisoning	1	Steffens, 2002

Table 48: *Cases of acrylonitrile poisonings* (cont'd)

Case					Antidote treatment								Reference
Age (y)	Sex	Latency time (h)	Maximum concentration (mg/l)		Cumulative dose of antidote (g)						Contributive?	PSSa	
			Acrylonitrile	Cyanide	AN (pearls)	SN	STS	HOCO	O₂	NAC			
NS	M*	Minutes	1.54	0.1	-	-	NS	-	-	NS	Unclear, no significant cyanide poisoning	1	Steffens, 2002
41	M	During exposure	0.14	3.94	-	-	NS	-	-	NS	Yes	1	Steffens, 2002
NS	M	During exposure			-	-	-	-	-	-	-	4	Bader and Wrbitzky, 2006

Case studies are detailed Appendix C
* Dermal exposure; ** combined inhalation and dermal exposure
NS, not stated; -, not applied.

Table 43: Acrylonitrile poisonings extracted from the PSSa database (Appendix B)

Case		Antidote treatment				PSSa		Reference
Age (y)	Sex	Cumulative dose of antidote (g)			Number of treatment steps	Before	After	
		AN (pearls)	SN	STS				
STS alone								
32	M	-	-	1	1	0	0	Steffens, 1998
28	M	-	-	2	1	2	0	Steffens, 1998
56	M	-	-	6	2	1	0	Steffens, 1998
18	M	-	-	3	2	1	0	Steffens, 1998
STS and nitrites								
NS	NS	NS	-	-	1	1	3	Van Luijt, 1963
56	M	NS	39	162.5	1	2	0	Vogel, 1984

Case studies are detailed Appendix C
NS, not stated; -, not applied

Treatment / respiratory prophylaxis

The antidotes used in cases with certain cyanide toxicity (blood levels) have been effective – the Lily kit as well as NAC and a combination of NAC and STS. The issue of NAC is discussed in the chapter on Developmental and Preclinical Antidotes.

Discussion and conclusion

Acrylonitrile is itself toxic and in cases of significant intoxicaitons requires antidotal treatment with N-acetyl cystein. Experience seems to indicate that NAC, and if combined with STS, may effectively lower blood cyanide concentrations. STS in combination with sodium nitrite (and possibly amyl nitrite) has also been shown to be an effective antidote. For other cyanide antidotes the situation is less clear since there is only one single case with questionable outcome with HOCO and none for the others.

It is suggested to use the recommended antidotal treatment for acrylonitrile with NAC, preferably together with STS, and in severe intoxications with sodium nitrite. Other direct antidotes like DMAP and HOCO may also be attempted, but experience is lacking.

Other nitriles

For the few cases with other nitriles all information is given in this chapter.

One case of poisoning with **isobutyronitrile** has been described in a worker who used the wrong respiratory filter. During exposure he developed nausea, vomiting, coma, collapse, and seizures

but recovered after a single application of SN (10 ml 3%) and STS (100 ml 10%), whereas prior administration of AN had no effect.

The same worker had three prior episodes of vomiting, unconsciousness and seizures due to intoxications with **ortho-phthalodinitrile,** but no further details were reported (Thiess and Hey, 1969).

The case is contributive but not definitive for the non-efficacy of AN, and efficacy of SN and STS.

One case from Italy reported a young man who accidentally drank a few mls of **adipic nitrile**. Symptoms included weakness, headache, vertigo, nausea / vomiting, cyanosis, tachypnoea, hypotension, mydriasis, somnolence, tonic-clonic convulsion and inability to stand. All these quickly resolved after administration of STS 15 ml 25% (Ghiringhelli, 1955), which makes the case contributive for the efficacy of STS.

Two cases of methacrylonitrile inhalation in 2002 have been retrieved via a questionnaire survey:

One worker had dizziness, weakness, nausea, vomiting, tachycardia, hypotension and dyspnoea and his cyanide level in blood was 1.627 mg/l. However, a propionitrile level of 1.038 mg/l was also found which is not fully in agreement with exposure to methacrylonitrile. The second worker had the same symptoms, but no measurements were done. Both received STS (1 g) and N-acetylcysteine (12 and 14 g), which led to recovery. The latency time is unknown (personal communication Roedelsperger, 2009).

The latest case was an occupational exposure to **pentenenitrile** and unspecified solvents without the use of adequate personal protective equipment. A 38-year-old man developed progressive weakness several hours after exposure. He became syncopal and was found with atrial fibrillation and tachyarrhythmia, but awoke again with headache, nausea and vomiting. After application of AN and SN, and STS (doses unspecified), he rapidly regained sinus rhythm once more and his other symptoms regressed. Delayed cyanide poisoning was shown by elevated thiocyanate in serum before STS application (Fernández et al, 2008). This case is contributive for the efficacy of the Lilly Kit.

The cases are summarised in table 44.

Table 44: Other nitrile poisonings

Case					Antidote treatment						Reference
Age (y)	Sex	Type of poisoning	Cyanide (mg/l)[b]	Latency time (h)	Cumulative dose of antidote (g)				Contributive?	PSSa	
					AN (pearls)	SN	STS	NAC			
NS	M	Isobutyronitrile		During exposure	NS	NS	NS	-	No for AN, yes for SN/STS	3	Thiess and Hey, 1969
	Same 3×	Ortho-phthalo-dinitrile			-	-	-	-	-	3	Thiess and Hey, 1969
NS	M	Adipic nitrile			-	-	NS	-	Yes	2	Ghiringhelli, 1955
46	M	Methacrylonitrile	1.627		-	-	NS	NS	Yes	1	Roedelsperger, 2002 (2009)
40	M	Methacrylonitrile			-	-	NS	NS	Yes	1	Roedelsperger, 2002 (2009)
39	M	Pentenenitrile		Several hours	-	-	-	-	-	2	Fernández et al, 2008

The cases are detailed in Appendix C.
[b] Maximum concentration
NS, not stated; -, not applied.

Table 45: Other nitrile poisonings extracted from the PSSa database (Appendix B)

Case			Antidote treatment				PSSa		Reference
Age (y)	Sex	Type of poisoning	Cumulative dose of antidote (g)			Number of treatment steps	Before	After	
			AN (pearls)	SN	STS				
STS alone									
NS	M	Adipic nitrile	-	-	3.75	1	2	0	Ghiringhelli, 1995
NS	M	Methacrylonitrile	-	-	1	1	2	0	Roedelsperger, 2009
NS	M	Methacrylonitrile	-	-	1	1	2	0	Roedelsperger, 2009
STS and nitrites									
19	M	Isobutyronitrile	NS	3	10	1	3	3	Thiess, 1969
27	M	Pentenenitrile	NS	NS	NS	1	2	0	Fernández, 2008

Case studies are detailed Appendix C
NS, not stated; -, not applied.

In animal experiments it has been shown that the tear gas CS (o-chlorobenzylidene malononitrile) also causes cyanide formation *in vivo* via metabolism to malononitrile. The effect was less pronounced in aerosol inhalation than in i.p. injection (Frankenberg and Sörbo, 1973). In literature there are no cases in humans, in which cyanide has been measured, or in which cyanide effects have been assumed.

Critical analysis of casuistics re efficacy and safety of antidotes

The formation of cyanide from nitriles requires one or more metabolic steps and therefore contributes to the delay in onset of symptoms. If applied early enough the application of STS should theoretically be sufficient, at least for cases of aliphatic nitrile poisoning. Cyanide formed over time can thus be trapped and metabolized to thiocyanate.

Directly acting cyanide antidotes only appear to be required in poisoning cases that are already presenting high cyanide levels such as in cases of oral acetonitrile ingestion and late presenting inhalation exposure, acetone cyanohydrin intoxication and possibly acrylonitrile poisoning.

Since there is no other cause for hypoxemia in such poisonings there is no restriction regarding the selection of directly acting cyanide antidotes. HOCO and careful application of MetHb formers are possible.

In the case of acrylonitrile, for which there is a specific antidote (NAC), there are indications that NAC alone might suffice to lower cyanide in blood to non-threatening levels. In the cases reported from Germany only NAC and STS have shown to be fully sufficient (Steffens et al, 1998; Steffens, 2000). This is supported by animal experiments showing that STS is an effective antidote for acrylonitrile poisoning, abolishing lethality even if applied 10 or 30 minutes after an LD_{50} dose of acrylonitrile to mice (Mehta, 1995).

From the literature it can also be seen that in human acrylonitrile poisonings STS has been recommended as the only suitable cyanide antidote due to the slow cyanide formation (Butuc, 1969). From China a treatment using methemoglobin forming agents and STS is recommended stressing that the results are better, the sooner and more thiosulfate is given (Chen et al, 1999). A similar treatment had been advocated decades ago (Dudley and Neal, 1942). A Polish publication suggests the use of STS and Co-EDTA (Nagorzwanski, 1966).

Again, cases presenting late after exposure with high cyanide levels may benefit from the application of direct acting cyanide antidotes.

So, in general, STS should be sufficient as an antidote in nitrile poisonings giving raise to cyanide liberation. NAC may be given in support in such cases. SN, DMAP or HOCO may be indicated, but only in cases with significant cyanide poisoning and where treatment is delayed.

Acetone cyanohydrin

Introduction

For ACH there are no data available from controlled human studies. Anecdotal case reports of incidents within Task Force member companies indicate that ACH is acutely extremely toxic by the inhalation, dermal and oral routes, although there is no documentation of effects relative to dose. Due to the low vapour pressure of ACH, inhalation exposure is predominantly to HCN, the decomposition product.

Substance data

Common Name:	Acetone Cyanohydrin
IUPAC name:	2-hydroxy-2-methylpropanenitrile
CAS registry No.:	75-86-5
Formula:	C_4H_7NO
Molecular mass:	85.11
Structure:	
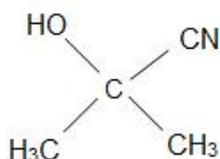
Physical State:	Clear liquid
Odour:	Faint odour of bitter almond
Water Solubility:	Completely soluble in water
Log P_{ow}:	–1 (Calculated) Rekker, 1977

Conversion factor: 1 ppm = 3.48 mg/m³, 1 mg/m³ = 0.287 ppm (25 °, 760 mm Hg)

Acetone cyanohydrin (ACH) is manufactured by the base-catalysed condensation of acetone with HCN and supplied stabilised with a combination of H_2SO_4 and SO_2 to prevent decomposition back to HCN acetone (ECETOC, 2007).

Intrinsic Properties (pharmacokinetic and toxicity data)

ACH is typically stabilised by the addition of 0.01% sulphuric or phosphoric acid. Stabilised ACH will exert a significant vapour pressure, primarily due to the presence of more volatile HCN at room temperature. Under physiological conditions, acid-stabilised ACH will be buffered by the intracellular buffering capacity resulting in its rapid and quantitative decomposition to HCN and acetone (Frank *et al*, 2002; ECETOC, 2007). Hence, although some absorption of ACH is likely the resultant systemic toxicity profile will almost certainly exhibit the combined characteristics of HCN and acetone. However, since acetone is relatively less acutely toxic, the acute toxicity of HCN will predominate.

ACH causes a very rapid poisoning and and is directly comparable to other cyanides. Antidotes (SN and STS) have been given in only 2 cases with a successful outcome in only one of them.

In none of these cases were levels of ACH or cyanide measured. Measurements or normal cyanide levels only exist for a cluster of recent cases.

Eight exposure cases with ACH have been reported via a questionnaire survey. In 7 of these cases no significant exposure had occurred as shown by the low cyanide levels in blood and the lack of symptoms in the patients. One case was a needle stick injury with a syringe containing ACH. Cyanide in blood was below the level of detection, there were no symptoms. Ten grams of STS were given. Six cases were due to skin contact. A spill across the legs of 3 to 4 litres of ACH resulted in a cyanide level in blood of 0.106 mg/l (just above the limit of detection) without decrease after the antidote (STS 2g). There had been no symptoms. Three cases of contact to 10 to 20 ml amounts of ACH did not develop symptoms and had very low cyanide blood levels. They were all treated with STS (10 g). None of the cases was really cyanide intoxication, and so antidote application had not been required. This makes efficacy assessment impossible, side effects were not reported (Roedelsperger, 2009).

Casuistics

The available casuistics are given in table 46.

Table 46: Acetone cyanohydrin poisonings

Case			Maximum concentration (mg/l)		Antidote treatment							Reference
Age (y)	Sex	Latency time (h)			Antidote				Contributive?	PSSa		
			Acetone cyanohydrin	Cyanide	AN (pearls)	SN	STS	HOCO		Before	After	
	M	3			-	-	-	-		1(4)		Sunderman and Kincaid, 1953
	M	NS			-	Yes	Yes	-	No for SN, STS	3(4)		
	M	Minutes			-	-	-	-		3		Krefft, 1955
	M	Minutes			-	-	-	-		3(4)		
	F	Minutes			-	-	-	-		4		
19	M	During exposure			-	-	-	-		3		Lang and Stintzy, 1960
23	M	Minutes			-	Yes	Yes	-	Yes for SN, STS	3		Thiess and Hey, 1969
19	M	(Needle)		<0.1 mg/l	-	-	Yes	-	N/a as no clear poisoning	0		Roedelsperger, 2004 (2009)
29	M	(Inhal.)		<0.1 mg/l	-	-	-	Yes	N/a as no clear poisoning	1		2001 (2009)
23	M	(Skin)		0.106 mg/l	-	-	Yes	-	N/a as no clear poisoning	0		2003 (2009)
	M	(Skin)			-	-	-	Yes	N/a as no clear poisoning	0		2005 (2009)
20	M	(Skin)		<0.1 mg/l	-	-	Yes	-	N/a as no clear poisoning	0		2007 (2009)
27	M	(Skin)		0.05 mg/l	-	-	Yes	-	N/a as no clear poisoning	0		2007 (2009)
52	M	(Skin)		0.035 mg/l	-	-	Yes	-	N/a as no clear poisoning	0		2007 (2009)
53	M	(Skin)		0.998 mg/l	-	-	-	Yes	Partially, decrease in cyanide level, though no poisoning symptoms	0		2008 (2009)

Case studies are detailed Appendix C.

NS, not stated; -, not applied.

Table 47: Acetone cyanohydrin poisonings extracted from the PSSa database (Appendix B)

Case		Antidote treatment					PSSa		Reference
Age (y)	Sex	Cumulative dose of antidote (g)				Number of treatment steps	Before	After	
		AN (pearls)	SN	STS	HOCO				
STS alone									
29	M	-	-	10	-	1	0	0	Roedelsperger, 2009
NS	M	-	-	2	-	1	0	0	Roedelsperger, 2009
27	M	-	-	10	-	1	0	0	Roedelsperger, 2009
52	M	-	-	10	-	1	0	0	Roedelsperger, 2009
53	M	-	-	10	-	1	0	0	Roedelsperger, 2009
STS and nitrites									
44	M	NS	3.3	10	-	2	3	0	Thiess and Hey, 1969
HOCO									
29	M	-	-	-	2.5	1	0	0	Roedelsperger, 2009
NS	M	-	-	-	2.5	1	0	0	Roedelsperger, 2009
53	M	-	-	-	2.5	1	0	0	Roedelsperger, 2009

Case studies are detailed Appendix C.
NS, not stated; -, not applied.

Treatment / resp. prophylaxis

Due to the relative instability of ACH and its propensity to form HCN within the body, antidotal treatment is based upon the rationale of targeting the cyanide formed, rather than the parent molecule.

There is extremely limited experience with the use of antidotes for treatment of ACH poisonings.

Notwithstanding this a combination of STS and sodium nitrite was without success in one case, but effective in another. For HOCO a partial efficacy was seen by lowering the blood cyanide level in a case without poisoning symptoms (Roedelsperger, 2009).

Discussion and conclusion

ACH causes a very rapid poisoning and and is directly comparable to other cyanides. Antidotes (SN and STS) have been given in only 2 cases with a successful outcome in only one of them. The efficacy of HOCO is unclear.

In regard of the rapid cyanide formation, STS alone is not assumed to be sufficient for antidotal treatment. Quick acting antidotes like sodium nitrite, possible also DMAP or HOCO are expected to be required.

5.2.4 Sodium nitroprusside

Introduction

The hypotensive effect of SNP was discovered in 1887 and used in a clinical trial in 1928 (Leeuwenkamp et al, 1984). The use of SNP as an i.v. adminsitered drug started in the early 1950s (Page et al, 1955).

The toxicity of SNP has been recognised since 1886 (Herrmann, 1886) and has been confirmed by experiences from its extensive clinical use as detailed below.

SNP is used for treating hypertensive emergencies and acute heart failure syndromes (Elliot, 2004).

Identity

Common name:	Sodium nitroprusside
IUPAC name:	Sodium pentacyanonitrosylferrate (II)
CAS Registry number:	13755-38-9
Formula:	$Na_2[Fe(CN)_5NO] \cdot 2H_2O$
Molecular weight:	298
Chemical structure:	
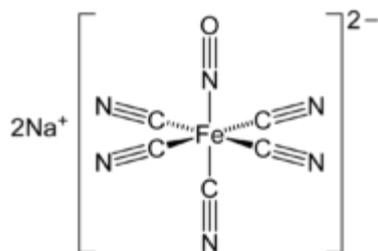
(Wikimedia Commons, 2012)

$\log K_{ow}$ –0.53 at 20°C (TerraBase, 2012)

SNP is a hydroscopic red crystalline salt that can be dissolved in water, and to a lesser degree in alcohol. Based on the acute toxicity data SNP is classified and labelled as toxic if swallowed, harmful if inhaled or in contact with skin. Additionally it is irritating to skin and eye.

Pharmacokinetics and toxicity

If SNP is swallowed, cyanide is formed immediately by hydrolysis with gastric acid, and will be distributed very quickly via blood.

Therapeutical admiistration as a hypotensive drug is by continuous i.v. infusion only as it has an elimination half-life of 2 minutes (Delanty, 2004; Cole and Vessey, 2000).

The hypotensive effect of SNP is based on the liberation of nitrogen monoxide (NO). The cyanide moieties do not contribute to the hypotensive effect.

The ligands (five cyanide and one nitric oxide) of nitroprusside are liberated very quickly in the blood by biotransformation (Froldi et al, 2001), interaction with oxy-Hb, dissociation, MetHb formation and subsequent release of the cyanide and nitric oxide moieties (Smith and Kruszyna, 1974; Tinker and Michenfelder, 1976; Friederich and Butterworth, 1995). More precisely, SNP is rapidly broken down by free and intracellular Hb by electron

transfer via a non-enzymatic reaction with Hb-iron, forming MetHb and an unstable nitroprusside radical. The radical splits and releases the 5 cyanide ions, of which one binds to MetHb to give cyano-methaemoglobin, while the 4 others remain as cyanide in the organism, until they are detoxified in the liver to thiocyanate. The detoxification rate is limited by the amount of available sulphur-donor in the body (Drew, 1983).

Though the reaction described is very rapid, in literature toxic cyanide blood levels have been reported after 50 minutes to 14 days during SNP treatment (Rindone and Sloane, 1992). In volunteers highest levels were reached 3.5 hours after the initiation of SNP infusion and were sustained for at least 210 minutes (Cottrell et al, 1978).

A recent paper has shown cyanide formation by free Hb in blood specimens from 25 cardiac surgery patients undergoing cardiopulmonary bypass (Cheung et al, 2007).

In a group of 26 patients receiving up to 95.4 mg SNP the maximum plasma cyanide level was 0.0533 mg/l and the maximum red blood cell cyanide level 5.512 mg/l (Vesey et al, 1976).

Another series of patients without poisoning symptoms has been reported, in which doses of SNP between 12 mg/15 min and 783 mg/85 min were given. A dose dependant range of cyanide levels was found between 0.18 mg/l and 2.05 mg/l (Aitken et al, 1977).

In a series of 50 patients cyanide was found at a maximum of 1.8 mg/l, with a mean of 0.333 mg/l, after infusion of 50-1230 µg/kg and an infusion time of between 50 and 120 minutes (Bogusz et al, 1979).

In a study series levels of up to 100 nmol/ml erythrocytes were reported (Schulz et al, 1982).

The same author reported a level of 13 mg/l in the red blood cells in a child treated with SNP (Schulz et al, 1982).

In 52 paediatric patients 62 cyanide measurements were taken. The highest level found was 2.141 g/l, 4 measurements were in the assumed toxic range (above 0.5 mg/l) but no clinical signs of cyanide toxicity were observed. The measurements were prompted by a child with cyanide levels up to 2 mg/l becoming obtunded and hypoxic but this might have been due to pacemaker dysfunction (Linakis et al, 1991).

An oral suicide case mentioned above had a total cyanide level of 5 mg/l with 3.3 mg/l of free cyanide (Froldi et al, 2001).

In 26 patients, consisting of 17 females and 9 males aged between 15 and 77, a linear relationship of plasma (0-194 nmol/100 ml) and red blood cell cyanide (0.042 - 17.35 µmol/100 ml) to total SNP dose were seen after doses of between 4 and 95.4 mg SNP (Vesey et al, 1976).

In a recent case a level of 3.1 mg/l cyanide was seen after administration of a total of 200 mg SNP over 56 hours (Quinlan et al, 2008).

In a series of 13 hospital homicides, allegedly all with SNP, in 3 cases elevated cyanide levels of between 0.2 and 0.4 µg/ml could be found even after exhumation. Since these levels cannot be related to acute levels immediately after poisoning they are not considered in the table 48 below (Ehrlich et al, 2009).

Table 48: Cyanide concentrations in blood in cases of cyanide toxicity from sodium nitroprusside therapy from single reports and from the overview publications

CN⁻ concentration (mg/l)	Outcome	Source
5	Death	Davies et al, 1975a, Davies et al, 1975b
2.99	Death	Posner et al, 1977
3.6	Survived	Schulz et al, 1979
3.64	Death	Kim et al, 1982
13.6	Death	Schulz et al, 1982
13 (red blood cells)	Survived, delayed death	Schulz and Roth, 1982
0.17	Death	Robin, and McCauley, 1992
0.8	Survived	Patel et al, 1986
0.575	Survived	Patel et al, 1986
3.66	Death	Patel et al, 1986
0.517	Death	Patel et al, 1986
0.555	Survived	Patel et al, 1986
0.546	Survived	Patel et al, 1986
1.6	Death	Patel et al, 1986
2.141	Survived	Linakis et al, 1991
1.73	Death	Robin and McCauley, 1992
4	Death	Robin and McCauley, 1992
6	Death	Robin and McCauley, 1992
1.69 [b]	Death	Robin and McCauley, 1992
0.43 [b]	Not known	Robin and McCauley, 1992
7.13 [b]	Survived	Robin and McCauley, 1992
4.68 [b]	Survived	Robin and McCauley, 1992
2	Death	Sipe et al, 2001

Case studies are detailed Appendix C.

[a] Some figures given in µmol/l were converted using a factor of 0.026.

[b] Stated as 169, 43, 713 and 468 without unit. Presumably this refers to µg/100 ml or µmol/l (nmol/ml).

It should be acknolwedged that there are also fatalities with symptoms reminiscent of cyanide toxicity (unstable myocardial function, hypo-perfusion state) in which no cyanide could be detected in serum, urine and brain (Merrifield and Blundell, 1974). As cyanide in tissues is labile and its analysis is not without technical difficulty these may, however, be false negatives.

In animal experiments the acute oral toxicity was 99, 61 or 34 mg/kgbw in the rats, mice and rabbits, respectively (http://fscimage.fishersci.com/msds/30681.htm). Whereas the i.v. LD_{50} values (mg/kgbw) are reported as 11.2, 8.4, 2.8 and 5.0 in the rats, mice, rabbits and dogs, respectively [htp://www.rxlist.com/nitropress-drug.htm]. Another source quotes 9.3, 6 and 1.8 mg/kgbw in trats, mice and rabbits, respectively [www.drugfuture.com/toxic/q56-q457.html].

Casuistics

The toxicity of SNP has been known since 1886 (Herrmann, 1886). The first two fatalities from suicides by oral ingestion of SNP were described in 1941 and 1942 and cyanide was detected in the stomach in both (Lazarus-Barlow and Norman, 1941; Hill, 1942).

In 1982, 10 cases of cyanide poisoning from SNP were summarised (Schulz et al, 1982). In 1992 a publication reported on 17 cases found on Medline from 1970 to 1992, of which 8 were new compared to the older publication (Rindone and Sloane, 1992). In the same year an overview detailed 52 further poisoning cases reported to the FDA, of which 29 have been fatal (Robin and McCauley, 1992). Four cases seem to be redundant to cases published before. Unfortunately the data given in this publication do not allow for an assessment – the cases with reported cyanide blood levels are listed below above (table 48).

Not included in these lists were 4 cases from Canada (Aitken et al, 1977) and one case from the USA (Michenfelder and Tinker, 1977).

Publications could be retrieved on a total of 77 cases, 41 of which were fatal.

Table 49: Potential and certain acute cyanide poisoning cases from sodium nitroprusside therapy

Case						Antidote treatment								Reference
		SNP		Cyanide concentration (mg/l)	Cyanide poisoning?	Cumulative dose (g)						Contributive for efficacy?	PSSa	
Age (y)	Sex	Total dose (mg)	Time			AN (pearls)	SN	STS	DMAP	HOCO	O₂			
23	F	NS	NS	Low amount in stomach	Possible	-	-	-	-	-	-	-	-	2, (4)	Lazarus-Barlow and Norman, 1941a
NS	M	NS	NS	Cyanide in stomach	Probable	-	-	-	-	-	-	-	4	Lazarus-Barlow and Norman, 1941a
NS	F	NS	NS	Low amount in stomach	Possible	-	-	-	-	-	-	-	4	Hill, 1942a
20	M	750	5 h	Low thiocyanate found	Unlikely	-	-	-	-	-	-	-	3, (4)	Merrifield and Blundell, 1974a,b
NS	M	750	NS	Metabolic acidosis	Possible	-	-	-	-	-	-	-	3, (4)	Jack, 1974a,b
42	F	250	90 min	Symptoms, metabolic acidosis	Possible	-	-	-	-	-	-	-	2	MacRae and Owen, 1974a,b
40	M	400	80 min	5	Yes	-	-	-	-	-	NS	Negative	4	Davies et al, 1975a,b
		0.783/kg	85 min	1.8, acidosis	Yes	-	-	-	-	-	-	-	2	Aitken et al, 1977
		0.735/kg	75 min	2.05, acidosis	Yes	-	-	-	-	-	-	-	2	Aitken et al, 1977
		0.496/kg	55 min	1.1, acidosis	Yes	-	-	-	-	-	-	-	2	Aitken et al, 1977
		0.340/kg	35 min	0.9	Yes	-	-	-	-	-	-	-	0	Aitken et al, 1977
14	M	NS	NS	0.18	Possible	-	-	-	-	-	-	-	4	Michenfelder and Tinker, 1977
66	M	238	38 h	2.99	Yes	-	-	-	-	-	-	-	3, (4)	Posner et al, 1977a,b
14	M	130	5 h	Symptoms, reaction to antidote	Probable	-	-	NS	-	-	NS	Yes for oxygen failure and STS efficacy	3	Perschau et al, 1977b
66	F	490	28 h	Metabolic acidosis	Possible	-	-	-	-	-	-	-	1	Humphrey and Nash, 1978a,b
27	F	1,120	48 h	Symptoms, acidosis	Possible	-	-	-	-	-	-	-	4 (resusc.)	Rauscher et al, 1978b
42	M	13	90 min	Symptoms, metabolic acidosis	Possible	-	-	-	-	-	-	-	4	Montoliu et al, 1979a,b

Case						Antidote treatment								Reference
Age (y)	Sex	SNP		Cyanide concentration (mg/l)	Cyanide poisoning?	Cumulative dose (g)						Contributive for efficacy?	PSSa	
		Total dose (mg)	Time			AN (pearls)	SN	STS	DMAP	HOCO	O₂			
52	F	1,093	34 h	Symptoms, metabolic acidosis, thiocyanate in blood	Possible	-	-	-	-	-	- -	-	3	Mellino and Phillips, 1980a,b
28	F	480	NS	-	-	-	-	-	-	-	- -	-	3, (4)	Case Nuremberg, 1979; in Schulz et al, 1982
10 d	M	28	NS	13.6 in erythrocytes	Yes	-	-	-	-	-	-	Delayed death	2, (4)	Case Cologne, 1980; in Schulz et al, 1982; Schulz and Roth, 1982b
43	M	2,878	14 d	Symptoms	Possible	-	-	-	-	-	- -	-	3, (4)	Kim et al, 1982b
58	F	1,000	6 d	5	Yes	-	NS	NS	-	-	-	- (not reported)	2	Marbury et al, 1982
62	F	721	80 h	0.8	Yes	-	-	NS	-	-	-	Yes	3	Patel et al, 1986b
65	M	157	30 h	0.517	Yes	-	-	NS	-	-	-	Temporarily effective	2, (4)	Patel et al, 1986b
72	F	447	160 h	0.575	Yes	-	-	NS	-	-	-	Yes	2	Patel et al, 1986b
71	M	682	55h	3.66	Yes	-	-	NS	-	-	-	No, delayed death	2, (4)	Patel et al, 1986b
61	M	431	74	0.555	Yes	-	-	NS	-	-	-	Yes	2	Patel et al, 1986b
71	M	126	26 h	0.546	Yes	-	-	NS	-	-	-	Yes	2	Patel et al, 1986b
58	M	661.6	81 h	1.6	Yes	-	-	NS	-	-	-	No, death	3, (4)	Patel et al, 1986b
NS	NS	NS	NS	Symptoms	Possible	NS	-	-	-	-	-	Yes ('dramatic improvement')	1	Cetnarowski and Conti, 1986
53	M	NS	NS	Symptoms	Possible	-	-	NS	NS	-	NS	Yes for the combination	2, (3)	Ram et al, 1989a
78	F	1,300	5 d	2	Yes	-	NS	NS	-	-	-	No, no improvement	2	Sipe et al, 2001
41	F	6,000 p.o	NS	5	Yes	-	-	-	-	-	-	Found dead	4	Froldi et al, 2001
1.5mo	NS	NS	NS	No cyanide in blood	Unlikely	-	-	-	-	NS	-	Improvement but poisoning uncertain	3	Ballesteros Garcia et al, 2003
58	M	NS	NS	Symptoms	Possible	-	-	-	-	-	- -	-	1	Nicoletta et al, 2007

The cases are detailed in Appendix C.
[a] Cited by Schulz et al, 1982b: cited by Rindone and Sloane, 1992.
NS, not stated; -, not applied.

Table 50: SNP poisonings

Case		Antidote treatment								Reference
Age (y)	Sex	Cumulative dose of antidote (g)					Number of treatment steps	PSSa		
		AN (pearls)	SN	STS	DMAP	HOCO		Before	After	
STS alone										
14	M	-	-	0.75 about	-	-	1	3	0	Perschau, 1977
58	M	-	-	> 0.1	-	-	2	2	0	Schulz, 1982
72	F	-	-	18.75	-	-	1	3	0	Patel, 1986
71	F	-	-	0.1 or 0.2	-	-	1	2	0	Patel, 1986
65	M	-	-	0.1 or 0.2	-	-	1	2	3	Patel, 1986
61	M	-	-	0.1 or 0.2	-	-	1	2	2	Patel, 1986
71	M	-	-	0.1 or 0.2	-	-	1	2	0	Patel, 1986
58	M	-	-	0.1 or 0.2	-	-	1	2	0	Patel, 1986
NS	M	-	-	0.1 or 0.2	-	-	1	3	4	Patel, 1986
STS and nitrites										
62	F	-	0.3	12.5	-	-	1	3	0	Marbury, 1982
78	F	-	0.6	25	-	-	1	2	4	Sipe, 2001
16	F	-	0.3	18.5	-	-	1	0	0	Quinlan, 2008
STS and DMAP										
23	M	-	-	12.5	0.25	-	1	3	0	Ram, 1989
Nitrites										
NS	NS	-	NS	-	-	-	1	2	0	Cetnarowski, 1986
HOCO										
1.5	M	-	-	-	-	NS	1	3	0	Ballesteros, 2003

The cases are detailed in Appendix C.
NS, not stated; -, not applied.

Treatment and/or prophylaxis

It has been clearly shown that the co-administration of STS with SNP will prevent cyanide poisoning from SNP in humans (Wiedemann, 1976; Schulz et al, 1982; Schulz, 1984; Hall and Guest, 1992). This has also been confirmed in animal experiments (Michenfelder and Tinker, 1977; Höbel et al, 1978; Drew, 1983; Vesey et al, 1985). In 1992 the use of STS together with SNP has been recommended by the FDA, the recommended dose being 1 g STS per 100 mg of SNP as a continuous infusion (Schulz, 1984; Dwyer and Morris, 1993). Also prophylactic treatment with α-ketoglutarate (500 mg/kg twice in 20 minutes) showed some effect in animals, albeit much smaller than that of STS (1 g/kg) (Yamamoto, 1992).

In a clinical study red blood cell cyanide levels during treatment with SNP alone (about 0.61 mg/kg) averaged approximately 0.8344 mg/l, with plasma levels about 0.035 mg/l, whereas with additional administration of HOCO (12.5 mg/30 minutes) the levels in red cells were about 0.332 mg/l and in plasma about 0.022 mg/l. Acidosis was prevented by the HOCO (Cotrell et al, 1978a,b).

This approach has also been corroborated in experiments in baboons with administration of 22.5 mg HOCO per 1 mg of SNP (Posner et al, 1976). However, a later study concluded that HOCO was effective against SNP toxicity only when given prophylactically, while support for its use once signs and symptoms had developed was lacking (Drew, 1983).

A comparison was made in animals between STS (75 mg/kg) and HOCO (1.5 mg/kg). Plasma cyanide levels were highest under HOCO, though oxygenation was not impaired. However, in contrast to STS, HOCO significantly increased the vascular response to SNP (1.5 mg/kg for 1 hour). At these doses STS was more effective than HOCO (Krapez et al, 1981).

A comparison in rats showed that HOCO slows the depressor response to SNP, but did not affect the degree of blood pressure lowering, while STS did not influence the depressor response (Hewick et al, 1978).

STS has been used as an antidote for acute poisoning and of particular interest is respect is the case series of Patel, cited by Rindone (Rindone and Sloane, 1992). Seven cases with measured cyanide concentrations are reported, all received STS as an antidote, not as a prophylactic. Four cases with severe neurological impairment died, four survived and improved after treatment with STS. However, except for two cases reported in more detail, the information provided is scarce (Patel et al, 1986). Another paper showed the efficacy of a STS bolus (10.6-38.5 mg/kgbw) given immediately on cessation of SNP infusion (Cole and Vesey, 1987).

As long ago as 37 years a high dose of HOCO was being recommended as an antidote to SNP toxicity (22.4 mg/mg SNP) (Wiedemann, 1976). Since then doses of 12.5 mg/30 minutes

(Cotrell et al, 1978) and 25 mg/hour (Zerbe and Wagner, 1993; Friederich and Butterworth, 1995), and an unspecified i.m. dose have also been shown to be effective clinically (Garcia et al, 2003).

Other immediately effective antidotes (MetHb formers), SN and DMAP have been used and/or recommended for the treatment of SNP poisoning, but not for prophylaxis (Marbury et al, 1982; Ram et al, 1989; Hall and Guest, 1992; Friederich and Butterworth, 1995; Sipe et al, 2001).

Recently, a review of literature indicated that STS therapy may only be required in patients with risk factors like impaired kidney function, prolonged and/or high dose SNP infusion and drug interactions (Thomas et al, 2009).

Discussion and Conclusion

Cyanide is formed from SNP and can accumulate to dangerous levels during high-dose infusion if not controlled by STS. Fatal outcomes have been seen at cyanide levels as low as 0.17 mg/l, whereas STS treatment resulted in survival at blood cyanide levels of 3.6 mg/l and delayed death at levels of 13 mg/l.

There are clear recommendations to use STS for prophylaxis in parallel with SNP at a dose of 1 g STS per 100 mg SNP. The efficacy of STS has clearly been demonstrated. HOCO has also been tested, used and recommended for prophylaxis. If no prophylaxis has been applied and dangerous cyanide levels are found, STS has been successfully used. Primary and rapidly acting cyanide antidotes like MetHb formers or HOCO may also be of use.

An antidote for cyanide poisoning resulting from treatment with SNP should be efficient and have few side effects. As such poisonings will occur in clinical settings, issues of shelf-life and cost might be secondary. However, following the clear official advice on prophylactic STS administration in parallel should make further antidote considerations unnecessary.

5.3   Fire smoke (mixed intoxications)

Introduction

An important source of cyanide poisonings is smoke inhalation. Many natural substances like wool, silk or paper, and synthetic polymers like acrylics, styrenes, nylons, plastics, melamine resins and polyurethanes can yield cyanide (HCN), when burning or especially when smouldering. Building materials, many substances and articles of daily use can release cyanide when incompletely combusted e.g. in confined fires.

Together with the large amount of other toxins released in fires, from chlorine, phosgene, hydrochloric acid gases, nitrous oxides, carbon monoxide and dioxide, to organics like acrolein and benzene, HCN may well play an important part in the toxicity of smoke. Yet the contribution of oxygen depletion and heat in fires should not be underestimated.

Most prominently smoke inhalation may lead to a combined severe systemic intoxication with the 'toxic twins' CO and HCN (Zilker, personal communication).

However, the allegedly typical signs of cyanide poisoning – elevated lactic acid levels and metabolic acidosis – may also be caused by oxygen depletion as well as by CO poisoning (Barillo, 2009).

Furthermore, since the auto-ignition temperature of HCN (538°C) will easily be exceeded in a typical room fire (537-1160°C; Barillo, 2009) a significant proportion of HCN produced will likely combust and CO inhalation toxicity will predominate over that of cyanide.

5.3.1 Toxicity - Cyanide concentrations

Cyanide concentrations in fires

Biomass (vegetation) burning is the principle source of cyanide in the environment (ECETOC, 2007). The amount of cyanide evolved in a fire will vary significantly depending upon fire conditions such as N content, temperature and oxygen availability with almost double the amount of HCN released from smouldering vegetation compared to that under flaming conditions (Lobert and Warnatz, 1993).

Most fire casualties are the result of house fires and will involve combustion conditions that will vary considerably since, for example, there will be a wide variety of both natural and synthetic materials available to combust and oxygen availability may be impeded. As a result it is extremely difficult to predict the level of cyanide evolved in these situations.

There are two ways to quantitate cyanide concentrations in house fires, these being experimental fires and direct measurements in fire-exposed fire-fighters. The results obtained using these two approaches have varied greatly.

Experimental fires revealed concentrations of either below 5 or 2500 ppm depending upon the amount of synthetic polymers, negligible in the first and 27% in the second experiment (Morikawa et al, 1987). Test animals had blood levels of 3.2 mg/l when exposed to the high CN concentrations.

An attempt to reconstruct an actual nightclub fire revealed a maximum HCN concentration of about 250 ppm, falling very rapidly to 20 ppm after five minutes (Davies, 1986).

Actual measurements by personal sampling in fire-fighters have not been positive in some of the fires studied (Brandt-Rauf et al, 1988; Burgess et al, 1979; Gold et al, 1978; Jankowicz et al, 1981; Lowry et al, 1985; Treitman et al, 1980) see table 51.

In addition, the studies with positive findings revealed lower HCN-concentrations than the experimental fires with a mean of 0.14 to 5.0 ppm with a maximum of 75 ppm (Burgess et al, 1979), the latter being cited from another publication (Brandt-Rauf et al, 1988),which found HCN in concentrations of 0 to 75 ppm, but only once higher than 10 ppm. It is interesting, that another sample from the same fire did not contain any HCN at all.

In 2 out of 173 victims of an Australian bushfire cyanide was detectable at 0.5 and 2.7 mg/l (Gerostamoulos et al, 2009).

An overview cites HCN concentrations in fire atmospheres between non-detectable and 'over 20' ppm (Shusterman, 1993).

Table 51: Cyanide exposures in fire-fighters

Author	Number of fires	% HCN⁻ positive	Range positive ppm	Calc. mean ppm	>1 ppm	>10 ppm (STEL)	>50 ppm (IDLH)
Gold et al, 1978	90	48	0.02-0.48	0.04	6	0	0
Burgess, 1979	253	10.7	0.2-3.6		5	0	0
Treitman et al, 1980	>200	10	0.2-4		5	0	0
Lowry et al, 1985	NS	12	NS-40	3.7	NS	NS	0
Brandt-Rauf et al, 1988	14	57	0.8-75	12.92	7	1	1
Jankovic et al, 1991	22	55	0.6-20		7	3	0

NS Not stated.

Cyanide can only be detected in about 10 to 50% of actual fires, rarely in excess of workplace limits for peak exposures (set for 15 minutes exposure) and very rarely in excess of IDLH values.

Assuming a concentration of 90 ppm as dangerous (Loeser 1994), cyanide does hardly seem to reach such concentrations. Judging from these personal sampling studies in fire fighters, cyanide at first view does not seem to pose a significant risk in fires. The above authors agree that CO is the main toxic factor in fires, and in addition sometimes acrolein is reported to occur in dangerous concentrations (Treitman et al, 1980).

Cyanide blood levels in fire / smoke victims

There is a multitude of studies reporting cyanide levels in victims of fires and smoke inhalations, from samples taken at fire scenes or in hospital to samples taken at autopsy. Even values in fire fighters are available.

Determination at autopsy

Most available studies report cyanide levels at autopsy, which usually is at best on the day following the accident. In many cases several more days passed until the actual measurement of cyanide in blood, which will have impaired the result reliability.

First results of cyanide analyses in fire victims were reported in 1966 (Wetherell, 1966). In 39 of 53 analysed cases cyanide could be detected in a range of 0.17 to 2.2 mg/l.

A study found cyanide levels in 89 fire deaths of 0 to 4.2 mg/l with a mean of 1.1 mg/l (Halpin and Berl, 1976; cited by Anderson and Harland, 1982).

A study in Maryland revealed cyanide levels above 2 mg/l in 9% of 256 cases (Caplan, 1977). The same figure is given in an enlarged study in the same population describing 463 fire deaths. Only 4 victims had cyanide levels above 3 mg/l with a maximum of 6 mg/l, however, also here only 256 blood samples have been analysed for cyanide (Berl and Halpin, 1979).

In the UK 15/43 victims from 13 fires were tested for cyanide in blood, giving positive results in 13 individuals in a range of 0 to 1.8 mg/l. A calculated mean would be 0.8 mg/l (Silcock et al, 1978).

A very large study with a cohort of 530 fire fatalities (in the first 6 hours after the fire) is reported by Birky et al (1979; 1981). Cyanide levels were determined in the last 272 cases. The mean concentration was 0.99 mg/l. 10% of the patients had levels above 2 mg/l, but unfortunately the exact levels are not reported. The same authors also present data of 10 out of 42 victims of a jail fire (Birky et al, 1979; 1980). The cyanide concentrations were 0.05-1.83 mg/l, with a mean of 0.76 mg/l.

A Scottish study reported first on 52 (Symington et al, 1978) and later on 100 fatalities and 41 non-fatalities (and on exposed firemen, healthy volunteers and fatalities from other causes than smoke) (Anderson et al, 1979). The fire victims had cyanide levels of 0 to 6.2 mg/l with a mean of 0.88 mg/l, much higher than in survivors. In the fire victims the cyanide levels correlated with the carbon monoxide levels. This study was continued and reported again 3 years

later (Anderson and Harland, 1981). In 139 fire victims the same concentrations of cyanide in blood were found (0-6.2 mg/l), but the mean was now higher at 1.2 mg/l.

In Germany cyanide could be found in 12/48 cases in a range of 0.52 to 6.24 mg/l, a theoretical mean being at 2.15 mg/l (von Meyer et al, 1979). 75% of the fire victims did not have measurable cyanide concentrations.

Victims of a hotel fire in Las Vegas are reported again by (Levin, 1990) referring to the original publication (CCFD, 1981). In 84 fatalities concentrations were between 0 and above 4 mg/l.

In 18/23 fire victims from house fires in the USA, cyanide was found in a range of 0 to 2.1 mg/l with a mean of 0.5 mg/l (Barillo et al, 1986a). A continuation of this study revealed cyanide in 24/29 victims in a range of 0 – below 2.5 mg/l with a mean of 0.74 mg/l (Barillo et al, 1986b). Eight years later, the authors reported on 433 fatalities, again mostly from house fires. In 279/365 cases cyanide could be detected. In 248 cases the cyanide level was below 3 mg/l, which is regarded as the potentially lethal threshold. 31 cases had cyanide concentrations in blood of 3.09 to 7.4 mg/l with an average of 1 mg/l. CO was elevated in 375/433 cases, of which in 195 cases the levels were in the lethal range above 50%. There was a positive correlation between the CO and the CN⁻ levels. The CO levels averaged 62.5% in cases in which the cyanide level was above 3.0 mg/l. The authors think that few fatalities are due to the combined intoxication and argue that the exposure to carbon monoxide can explain the fatal outcomes (Barillo et al, 1994).

A Canadian study reports on 64 fire victims, 50 had measurable cyanide concentrations, 9 had levels above 2 mg/l and 1 above 3 mg/l (4.15). The mean concentration was 0.87 +/- 0.88 mg/l (Péclet et al, 1982).

In 20/26 victims of fires in Florida (Copeland, 1985) cyanide could not be detected or was present just at trace level. 1 victim had a concentration between 1 and 2 mg/l, 3 victims between 2 and 3 mg/l and 2 victims above 3 mg/l.

Data on 18 victims of residential fires from Sweden (Lundquist et al, 1989) showed a range of cyanide concentrations of 0.016-2.63 mg/l with a calculated mean of 0.91 mg/l.

Fatalities from a hotel fire in Puerto Rico (n = 77) (Levin et al, 1990) showed a range of cyanide levels from 0.3-3.9 mg/l with one victim having a level of 11.4 mg/l. The calculated mean is 1.38 mg/l, without the extreme value 1.25 mg/l.

A Japanese study on 18 fatalities from house fires found strikingly different cyanide concentrations in the left and right ventricle, the ones in the left ventricle being significantly higher. The range of left ventricular concentrations was 0.11-18.12 mg/l, with a calculated mean

of 3.06 mg/l. The latter is purely theoretical and due to the one case with an extreme concentration (Shiono et al, 1991).

Airplane crashes are a special issue. Surprisingly many victims of airplane crashes had potentially lethal levels, showing that airplane fires can be exceptional and should not be compared to residential fires. Victims of 4 plane crashes in the United States between 1970 and 1972 were examined for cyanide levels in blood. Though the number of total victims and the percentage of positive cyanide identifications are not given, a range of 0.007-3.9 mg/l is reported (Mohler, 1975).

A further plane crash was described from the USA. The cause of death was assessed in 47 individuals as smoke/fire. All but one of these victims had cyanide concentrations of 0.03-1.34 mg/l with a mean of 0.56 mg/l (Salomone et al, 1987).

The cyanide levels in 55 victims of a later air plane crash and fire in the UK were 0.55-8.4 mg/l, the calculated mean was 2.75 mg/l (Hill, 1989; Mayes, 1991).

A publication on analytics found cyanide levels above 2 mg/l in 9 victims in a range of 2.1 to 60.4 mg/l (Zamecnik and Tam, 1987).

In a crash at Los Angeles the cyanide levels in 20 victims were 0-4.8 mg/l with a mean of 1.4 mg/l (Veronneau et al, 1992).

Aircraft fire victims were evaluated from 1990 to 2002. Of 201 fire-related cases 103 had a cyanide blood level of or above 0.25 mg/l (Canfield et al, 2005).

Cyanide levels in serum, not in whole blood, have been reported, too (Jones et al, 1987). In 4 of 6 fatalities concentrations of 1.4-2.4 mg/l could be detected with a mean of 1.78 mg/l.

These data are summarised in the following table (table 52)

Table 52: Cyanide concentrations in fire victims

Author, Year	No. of cases	Cyanide in blood mg/l - range	Cyanide in blood mg/l - mean	% above 3 mg/l
Wetherell, 1966	53	0 – 2.2	0.65 39/53 detected 0.48 all	0
Halpin, 1976	89	0 – 4.2	1.1	NS
Caplan, 1977,Berl	256	0 – 6	NS	1.6
Silcock, 1978	15	0 – 1.8	0.8	0
Birky, 1979	272	0 - > 2.0	0.99	NS
Anderson, 1979	100	0 – 6.2	0.88	6
Von Meyer, 1979	12/48	0.52 – 6.24	2.16	16.7/4.2
Birky, 1979/80	10	0.05 – 1.83	0.76	0
CCFD/Levin,1990	84	0 - >4	NS	2.4
Anderson, 1982	139	0 - 6.2	1.2	8.6
Péclet, 1982	64	0 - 4.15	0.87	1.6
Copeland, 1985	26	0 - >3	NS	7.7
Barillo, 1986	18/23	0 - 2.1	0.5	0
Barillo, 1986	24/29	0 - <2.5	0.74	0
Jones, 1987, Serum	4	1.4 – 2.4	1.78	0
Lundquist, 1989	18	0.016 – 2.63	0.91	0
Levin, 1990	77	0.3 – 11.4	1.38	3
Shiono, 1991	18	0.11 – 18.12	3.06	27.8
Barillo, 1994	364	0 – 7.4	1.0	8.5
Airplane crashes				
Mohler, 1975	51	0.007 – 3.9	NS	NS
Salomone,1986	47	0 – 1.34	0.56	0
Zamecnik, 1987	67	NS – 60.4		10.4
Mayes, 1991	54	0.55 – 8.4	2.75	37
Veronneau, 1992	20	0 – 4.8	1.4	NS
Canfield et al, 2005	201	=/> 0.25	In 103/201	NS

NS Not determined.

The results indicate that lethal cyanide concentrations (> 3 mg/l) are uncommon in victims of fires or smoke inhalation. Since the German and especially the Japanese study report very high percentage of cases with potentially lethal cyanide concentrations there may exist country or regional differences, perhaps corresponding to differences in building construction/house content materials. Many other studies, however, do not report any such cases at all although it has to be recognised that cyanide determinations at autopsy may give a wrong impression due to post-mortem changes in blood cyanide.

Determination made in hospitals

The above-mentioned Scottish study also reported cyanide levels in survivors of the fires. Blood samples were taken on arrival at the hospital. In 1978 and 1979 (Symington et al, 1978; Anderson et al, 1979) surviving smokers had levels of 0.1-0.59 mg/l, mean 0.34 mg/l, and surviving non-smokers had levels of 0-0.75 mg/l, mean 0.27 mg/l. 3 years later 41 non-fatalities (smokers plus non-smokers combined) had levels of 0.008-3.3 mg/l with a mean of 0.54 mg/l (Anderson and Harland, 1982).

Another study from the UK reports on 53 fire survivors of which 36 had clinical evidence of smoke inhalation (Clark et al, 1981). In this group the cyanide levels were 0.05-3.3 mg/l with a mean of 0.67 mg/l. Among the patients without smoke inhalation, a cyanide range of 0.057-0.34 with a mean of 0.17 mg/l was seen in smokers, whereas non-smokers had a range of 0.013-0.1 with a mean of 0.055 mg/l.

In 1985 a small cohort of cases was published. All the patients arrived unconscious from house fires. The range of cyanide levels was 0.35-3.9 mg/l, a mean of 1.6 mg/l can be calculated (Hart et al, 1985).

From France a group of 4 fire exposed patients that survived is reported in a publication on HOCO. The cyanide levels ranged from 0.6 to 5.4 mg/l (Baud et al, 1990).

Blood cyanide levels in a large cohort of 144 fire burn patients ranged from 0 to 11.5 mg/l with a mean of 0.65 mg/l (Silverman et al, 1988). There was no correlation between cyanide level and the burn size. A subset of 12 patients was described as having 'lethal' cyanide levels, which were reported at 1.2 - 11.5 mg/l with a mean of 3.56 mg/l. Eight of these 12 patients died. However, results from a further categorisation of the data were not conclusive: Four surviving patients had levels of 1.4 - 6.0 (mean 3.7) mg/l, 3 early deaths had 1.2 - 2.72 (mean 1.7) mg/l, and 5 late deaths had 1.4 - 11.5 (mean 4.56) mg/l. The concentrations in these 12 patients are shown in Table 53):

Table 53: Cyanide concentrations in fire burn patients **(Silverman et al, 1988)**

Cyanide in blood, mg/l	Sequelae
1.2	Early death
1.2	Early death
1.4	Late death
1.4	Survived
1.6	Late death
2.2	Survived
2.6	Late death
2.72	Early death
5.2	Survived
5.7	Late death
6.0	Survived
11.5	Late death

43 of the 144 patients were dead on arrival at hospital, their cyanide levels were 0.3-6.3 mg/l with a mean of 1.87 mg/l. It was not reported how many fatalities had levels above 2 mg/l.

Another small group of patients was reported from the US. Six Patients had cyanide levels on admission to the hospital of 0.96-3.38 mg/l, a theoretical mean being 2.08 mg/l (Kirk et al, 1989).

In 1996 a study was performed on 40 smoke inhalation patients. Cyanide was detectable in 23/39 cases and ranged from 0(below detection limit)-2.79 mg/l with a mean of 0.25 mg/l (Shusterman et al, 1996).

Table 54: Overview of the results in hospitalised patients

Author, year	No. of cases	Cyanide in blood mg/l - range	Cyanide in blood mg/l - mean	% above 3 mg/l
Anderson, 1979	21, s*	0.1 – 0.59	0.34	0
Anderson, 1979	19, ns*	0 – 0.75	0.27	0
Clark, 1981	36	0.05 – 3.3	0.67	5
Anderson, 1982	41	0.008 – 3.3	0.54	?
Hart, 1985	5	0.35 – 3.9	1.60	20
Silverman, 1988	144	0 – 11.5	0.65	2.8
Kirk, 1989	6	0.96 – 3.38	2.08	33
Shusterman, 1996	40	0 – 2.79	0.25	0

Case studies are detailed Appendix C.

* 's' = smokers, 'ns' = non-smokers.

Again, there are relatively few cases of cyanide levels in a potentially lethal range. However, two studies stand out, but both have very small numbers.

Under these sampling circumstances there is just one report of an airplane fire, from which 15 passengers have been rescued. The cyanide levels measured from 45 minutes to 3 hours after the crash ranged from 0.53 to 8.4 mg/l. Unfortunately it is not clear, whether only the survivors were tested, as data on COHb measurements in 55 fatalities were also reported in this publication (Hill, 1989).

Determination on site of accident

Some publications report blood sampling on the site of the fire. As they all come from the same working group in Paris it is not completely clear whether all publications really deal with different patient groups, or whether reporting is redundant.

Two papers described 50 cases although unfortunately dead and surviving patients are combined and there is no data on the number of positive cyanide detections. The levels were 0-8.9 mg/l with a mean of 2.0 mg/l. The number of patients with levels above 3 mg/l and the distribution between victims and survivors was not reported (Baud et al, 1988; Barriot et al, 1988).

In a further series of 109 cases the range of cyanide levels was 1.5 +/- 2.0 mg/l. The subgroup of surviving patients had a range of 0.56 +/- 0.95 mg/l, the subgroup of deceased patients of 3.0 +/- 2.3 mg/l. This publication is of high importance as it is the only one which shows a significantly higher cyanide levels in patients dying from smoke inhalation than in survivors. It also shows that it is mandatory to take blood samples for cyanide measurement on site and not later in hospital (Baud et al, 1991).

In 50 fire victims the range of cyanide concentrations was 2.16 +/- 1.9 mg/l. No distinction was made between victims and survivors. Again, it can be seen that cyanide levels were significantly higher when samples are taken on site as opposed to in hospital (Favier et al, 1993).

12 further survivors of house fires had cyanide levels of 0-3.51 mg/l. All patients with a cyanide level higher than 1 mg/l died. However, there was no clear cut correlation as patients with cyanide levels of 0.7 mg/l and a relatively low COHb of about 13% also died (Houeto et al, 1995; Houeto et al, 1996).

Finally, results of measurements in a group of 49 fire victims were in the range 0-0.42 mg/l with a mean of 0.2 mg/l. The levels in victims were higher than in survivors (Houeto et al, 1999).

The studies are summarised in table 55.

Table 55: Cyanide concentrations in samples taken on site

Author, Year	No. of cases	Cyanide in blood mg/l - range	Cyanide in blood mg/l - mean	% above 3 mg/l
Baud et al, 1988; Barriot et al, 1988	50	0 – 8.9	2.0	NS
Baud et al, 1991	109	1.5 +/- 2.0	1.5	NS
Baud et al, 1991; dead	43	3.0 +/- 2.3	3.0	
Baud et al, 1991; survived	66	0.56 +/- 0.95	0.56	
Favier, 1993	50	2.16 +/- 1.9	2.16	NS
Houeto, 1995	12	0 – 3.51	0.98	8
Houeto, 1999	51	0 – 0.42	0.2	0
Houeto, 1999; dead	7	0.93 – 2.54	1.7	
Houeto, 1999; survived	44	0 – 0.33	0.14	
Steffens 2002	17	< 0.1 (LOD)		0

Case studies are detailed Appendix C.
NS, Not stated.

If a lethal level of 3 mg/l is assumed (which may be too high a value), the following overview results (table 56) are found assessing studies that allow for calculation of a percentage ratio.

Table 56: Cyanide concentration above lethal levels

	n =	CN > 3 mg/l	%
Residential fire victims	1328	65	4.9
Airplane fire victims	168	9	5.4
Hospital sampling	261	9	3.4
On site sampling	80	1	1.3

Most studies found a positive correlation between CO and CN^- concentration in blood after smoke inhalation. Only in one retrospective study in 35 Argentinean prisoners, who had died in a smoke atmosphere, was there an indication that cyanide may inhibit the uptake of carbon monoxide (Ferrari et al, 2001). In contrast to these findings another study indicated that in smoke exposed persons CN^- levels were never elevated if carbon monoxide levels were in the normal range (Clark et al, 1981). It must be concluded, therefore, that the main focus of attention in patients with smoke inhalation should be on the clinical status and not on laboratory results.

Some authors assume that cyanide is more toxic on the CNS and cardio-respiratory system than CO. This may lead to the loss of orientation and to the impaired ability of individuals to leave the

danger zone unassisted thereby increasing the likelihood of a fatal outcome (Breen et al, 1995a,b; Eckstein and Maniscalco, 2006; Alarie, 2002).

Studies on the efficacy of cyanide antidotes on combined carbon monoxide / cyanide intoxication in animals

In vivo experiments in dogs with a combined CO plus CN exposure of the animals did not show a significant improvement after the use of STS alone. STS did not normalise lactate and cyanide levels. Only thiocyanate levels increased during the administration of STS. The elimination of CO was mainly dependent on the administration of pure oxygen during artificial respiration. The efficacy of the therapy with thiosulphate could be augmented by reducing the COHb (Breen, 1995a,b).

Moore et al (1987) published a study in animals using male ICR mice which were poisoned with CO by inhalation and cyanide by intraperitoneal injection. AN, SN and 4-DMAP were used as cyanide antidotes. AN and SN did not reduce mortality. Mortality was even enhanced by the use of these antidotes. Inhalation of AN elevated the death rate in the animals by 43% after one minute of inhalation and by 59% after two minutes. The intraperitoneal administration of SN enhanced mortality by 25%.

4-DMAP had no negative effect on the animals. The conclusion was that the reason was not the formation of MetHb but the hypotension that was induced by the nitrites. This is in accordance with the clinical experience published by Hall et al (1989). There are no reports on the use of 4-DMAP in smoke inhalation, which could affirm the animal data.

5.3.2   Case series and casuistics

The majority of case series have been reported by the Paris working group which predominantly uses HOCO as antidote. To avoid duplication the reader is referred to chapter 4.8.

An additional 13 cases of confirmed cyanide poisoning were identified after the cut off for data collection and were not entered into the PSSa database as they would not have significantly altered the outcome of the analysis. In the following table these cases are summarised.

Table 57: Antidotal treatment of smoke inhalation cases – casuistics

Case			Antidote treatment						PSSa		Reference
Age (y)	Sex	Blood cyanide concentration (mg/l)	Cumulative dose of antidote (g)						PSSa		
			AN (pearls)	SN	STS	DMAP	HOCO	O_2	Before	After	
78	M	0.34	-	0.6	-	-	-	-	3	4	Hall et al, 1989
20	M	3.38	-	0.3	12.5 (2×)	-	-	-	2	0	Kirk et al, 1993
39	M	3.16	-	0.3	12.5	-	-	-	3	3(4)	Kirk et al, 1993
64	M	0.96	-	0.3	12.5	-	-	NS	2	0	Kirk et al, 1993
26	F	0.7	-	0.3	12.5	-	-	NS	2	0	Kirk et al, 1993
29	M	1.5	-	0.3	12.5	-	-	-	2	0	Kirk et al, 1993
2 children		1.15, 1.1	-	0.4 g/kgbw	-	-	-	-	2	0, 1	Persson, 1993
42	M	NS	-	-	-	-	5	-	3	NS	Baud and Borron, 2007
37	M	NS	-	-	-	-	5	-	3	NS	Baud and Borron, 2007
47	M	3.4 *(after 5 g HOCO)*	-	-	-	-	10	-	3	3(4)	Fortin et al, 2007
54	F	NS	-	-	-	-	5	-	1	0	Cescon and Juurlink, 2009
NS	NS	1.5	-	-	-	-	NS	-	2	NS	Lawson-Smith et al, 2010
NS	NS	1	-	-	-	-	NS	-	3	3(4)	Lawson-Smith et al, 2010

Case studies are detailed Appendix C.
NS, not stated; -, not applied.

In 7 cases nitrites were administered after smoke inhalation. The Lilly antidote kit without AN (0.3 g SN in 100 ml and 12.5 g STS) was given to several paramedics' headquarters. It was advised that they should use it in cases of smoke intoxications which had to be ventilated, or had impaired consciousness or circulatory instability or persistent metabolic acidosis despite supportive treatment. Furthermore cyanide, carbon monoxide and MetHb were measured in blood (Kirk et al, 1993).

Five firemen (Case 3-7) were exposed to hydrogen cyanide gas for about 5 to 30 minutes. They were wearing protective clothing and face masks while working in the contaminated room. Having finished they complained about headache, dizziness, nausea, irritations of the eyes and throat and a narrowness of the chest. All were decontaminated completely and oxygen was applied. The symptoms regressed within a few hours (Lam and Lau, 2000).

Treatment

In the above cases SN/STS (Lilly Kit) has been used successfully in 4 cases and without success in one case. In a further case it was overdosed by applying a double dose of antidote.

For HOCO the situation is less clear, in the most part because data on 3 patients are missing. In the 3 cases that can be assessed, it was not successful in 2 severe poisoning cases, and it was successful in a case of mild poisoning. Regarding the case series treated with HOCO see chapter 4.8.

Two children (both 2.5 years of age) with unconsciousness and severe acidosis after smoke inhalation were treated with STS alone (400 mg/kg) and hyperbaric oxygen therapy. Their cyanide blood concentrations were 1.15 and 1.1 mg/l and both survived without sequelae (Unreferenced source cited by ICPS, 1993).

So far the suggestion for treatment of smoke inhalation is inconsistent. All authors suggest the use of oxygen inhalation and to treat supportively. As CN^- intoxication is uncertain during smoke inhalation, and the results of laboratory examination come too late to decide on treatment, antidotes with severe side effects like Cobalt-EDTA are not recommended. Despite having been used successfully in several cases together with STS, unless overdosed, nitrites are also not the treatment of first choice, as they produce methaemoglobinaemia (less than 4-DMAP) which further decreases the already impaired (by CO) oxygen transport capacity in blood (Hall et al, 1989a,b; Breen et al, 1995a,b). The current recommendation of the Task Force is that HOCO and/or STS are the therapeutic options of first choice since they have very little side effects and no influence on the oxygen transport system in blood.

5.3.3 Gaps in knowledge

It is unclear why the results of blood cyanide measurements vary so greatly and often do not show high levels in smoke inhalation victims, while French reports show high and potentially fatal concentrations in many cases. Possible explanations include differences in the combustion conditions, exposure duration, or degradation of cyanide between sampling and analysis.

Regarding antidote use there are discrepancies between animal experiments with SN and human cases. In humans the treatment seemed to be rather successful, while it was detrimental in studies in animals. For STS there is just one human case report. Again, in animals it did not seem to be effective.

5.3.4 Conclusion

Overall there are several case studies on the combined carbon monoxide / cyanide intoxication in smoke from fires. Variations in treatment, however, compromise their interpretation making a clear recommendation for the treatment of combined carbon monoxide / cyanide intoxication impossible.

The current situation for the time being is that cyanide poisoning cannot be proven on location, or within a reasonable time in the Emergency Room, and can only be assumed by signs and symptoms, e.g. soot in airways or by proxy laboratory results (lactate, acidosis) although the later may be elevated for other reasons. As there is a significant risk of treating patients with cyanide antidotes when they do not really require them this effectively rules out the use of antidotes with significant side effects. Co-EDTA clearly is not an option in such cases, SN/STS in combination may be usable but it has a potential for significant side effects due to its ability to lower blood pressure in the already compromised victim. DMAP may be more favourable in this respect.

The risk of producing a life-threatening reduction in Hb available for oxygen transport by adding e.g. 15% met-Hb to a pre-existing COHb level of say 50% is high, which speaks against the use of these MetHb forming antidotes in severe smoke inhalation poisoning cases.

STS has rarely been used – from a theoretical point of view it might be suitable, however, an animal experiment suggests otherwise.

The remaining antidote HOCO has the advantage of few and insignificant side effect apart from rare severe allergic reactions, which are treatable in emergency medicine. However, the results of HOCO use in severe poisonings are not fully convincing, as shown by the above cases and by the case series (see chapter 4.8).

It is of great importance however to remind emergency doctors or ambulance staff of the possibility of a combined CO/CN intoxication when they are confronted with household-fires. Cyanide antidotes are so far not used on a regular basis. This specific antidotal treatment should be carried out if there is sufficient justification from the signs and symptoms to suspect cyanide poisoning. Blood samples should be taken as early as possible for subsequent analysis. While this will come too late for treatment decision, it will further the understanding of cyanide involvement in smoke toxicity.

5.4   Poisoning with an unknown substance

Suspected Cyanide Poisoning / Clinical Symptoms of Cyanide Poisoning.

5.4.1   Introduction

In 2004, 2.4 million exposures to toxins were reported to the American Association of Poison Control Centres. Cyanide poisoning, however, is rare (Erickson, 2007).

Most poisoned patients do not present a typical symptomatology, but more or less severe clinical symptoms and signs that may suggest any kind of medical condition, including a poisoning. The following is a proposal for a standardised approach to treating patients suspected of having been poisoned by some unknown substance taking cyanide into account.

5.4.2   Approach to treating a patient suspected of having had an overdose or having been poisoned by an unknown substance

For the "Approach to the Patient with an Unknown Overdose" classical clinical skills, such as carefully history taking, including especially histories provided by third parties, and physical examination, are an essential part of case management and initial emergency treatment (Erickson et al , 2007).

Table 58: *Approach to the patient with an unknown overdose* (adapted from Erickson, 2007; Figure 1)

Poisoned Patient	
↓	↓
Diagnosis:	**Treatment:**
History	Airway
Physical examination	Breathing
Toxidrome recognition	Circulation
Diagnostic tests	Decontamination
	Enhanced elimination
	Focused therapy
	Get tox help

Recognising a toxidrome is considered crucial to initiating a clear diagnosis and therapy in the case of poisoning with an unknown substance. Well-documented toxidromes have been described for cholinergic, nicotinic, anticholinergic, sympathomimetic, and opioid reactions (Erickson, 2007).

Reports of certain odours may be useful for diagnosing a poisoning or metabolic disorder. Hydrocyanic acid has smell of bitter almonds that is often very subtle (Erickson, 2007). In addition, 20-40% of the population are unable to smell HCN, which also can cause olfactory fatigue [http://www.atsdr.cdc.gov/mmg/mmg.asp?id=1141&tid=249].

Although not a specific finding, cyanide is listed among poisoning agents that cause a lactic acidosis and anion gap (Erickson, 2007).

Table 59: Agents causing an elevated anion gap (METAL ACID GAP)

Methanol, metformin, massive overdose
Ethylene glycol
Toluene
Alcoholic ketoacidosis
Lactic acidosis
Acetaminophen (large overdose)
Cyanide, carbon monoxide, colchicine
Isoniazid, iron, ibuprofen
Diabetic ketoacidosis
Generalised seizure-producing toxins
Acetylsalicylic acid or salicylates
Paraldehyde, phenformin

Adapted from Erickson, 2007, Box 6.

5.4.3 Symptomatology of Cyanide Poisoning

Several authors described a general symptomatology of cyanide poisoning (Holland, 1983; Holland and Kozlowski, 1986; Hall, 1986; Baskin, 1997; Way, 1984; ATSDR, 1993; WHO, 2004) and provided information in varying detail on sources of cyanide, mechanisms of intoxication, mechanisms of antagonism, and treatment but not specific case reports.

According to Holland (1983), cyanide poisoning affects the circulatory, respiratory, and central nervous systems:

Clinical symptoms of the central nervous symptoms range from headache, vertigo, nausea, agitation, anxiety, and combative behaviour to generalised convulsions. Symptoms of the circulatory system may initially be elevated blood pressure with bradycardia, followed by lowered blood pressure with tachycardia, ST segment elevation or depression, and arrhythmias secondary to oxygen deficit. The respiratory system may be characterised tachypnoea, laboured breathing, respiratory paralysis, and pane.

Holland and Kozlowski (1986) also report signs and symptoms of acute cyanide toxicity. Early and late stages are distinguished for pulmonary and cardiovascular effects.

Table 60: Signs and symptoms of acute cyanide toxicity

Central nervous system effects	Headache
	Drowsiness
	Dizziness
	Seizures
	Coma
	Paralysis
Pulmonary effects - Early stage	Dyspnoea
	Tachypnoea
Late stage	Rapid decrease in respiratory rate
	Apnoea
	Pulmonary oedema
Cardiovascular effects - Early stage	Hypertension
	Reflex bradycardia
	Sinus or AV nodal arrhythmias
Late stage	Hypotension
	Tachycardia
	Complex arrhythmias
	Cardiovascular collapse
Local effects (after oral ingestion)	Burning of the tongue and mucous membranes
	Nausea
	Gastrointestinal irritation

(Holland and Kozlowski, 1986, Table 2).

Way (1984) provides a highly comprehensive overview of the biological mechanisms of cyanide poisoning. In his work, he proposes that cyanide affects the central nervous system more than it does the heart. Initial hyperpnoea, followed by dyspnoea and then by convulsive seizures, is described. The occurrence of cardiovascular signs is related to higher doses of cyanide. The brain is more sensitive to cyanide than the heart; as in cases where a lethal dose had been administered, frequently noted that electrical activity of the brain had stopped and the heart was still beating (Way, 1984).

Isom (1993) focuses on the effects on the central nervous system, the part of the body that cyanide selectively poisons. Neurological dysfunction including unconsciousness and respiratory depression occurred within seconds exposure, death from cardiorespiratory arrest occurred within seconds to minutes (Isom, 1993).

Isom also reports acute and subacute and chronic effects as well as post-intoxication sequelae.

Table 61: Classes of cyanide toxicity

Type of toxicity	Manifestations
Acute	Respiratory depression
	Cardiovascular collapse
	Unconsciousness
Post-intoxication sequelae	Parkinson-like rigidity
	Dystonia
	Memory deficits
Subacute/chronic	Spastic paraparesis
	Optic atrophy
	Cortico-spinal damage
	Nerve deafness
	Stomatitis
	Glossitis

(Adapted from Isom, 1993, Table 1).

Table 62: An overview of diseases associated with chronic cyanide toxicity is also provided

Disease	Symptoms
Konzo	Spastic paraparesis
	Upper motor neuron dysfunction
	Central visual field defects
Toxic ataxic neuropathy	Ataxic polyneuropathy
	Peripheral neuritis
	Areflexia
	Stomatoglossitis
Tobacco amblyopia	Optic atrophy
	Centro-ceacal scotoma

(Isom, 1993, Table 2)

Hall and Rumack (1986) describe the relationship between cyanide levels and associated symptoms, as well as cyanide concentrations in the air and the expected responses.

Table 63: Cyanide levels and associated symptoms

Whole Blood Cyanide		Symptoms
µg/ml	µmol/l	
0.2–0.5	8–20	None
0.5–1.0	20–38	Tachycardia. Flashing
1.0–2.5	48–95	Depressed level of consciousness
2.5–3.0	95–114	Coma
3.0	114	Death

(Hall and Rumack, 1986, Table 4), (also compare Baskin, 1997, Table 10-3)
Adapted from Rumack, 1983.

Table 64: Air cyanide concentrations and expected responses

	Concentration	
	mg/m3	ppm
Fatal immediately	300	270
Fatal 10 min	200	181
Fatal 30 min	150	135
Fatal within ½ to 1 hour or more, dangerous to life	120–150	110–135
No immediate or late effects with exposure for ⅓ or 1 hour	50–60	45–54
Mild symptoms with several hours exposure	20–40	18–36

(Hall and Rumack, 1986, Table 3).

Concerning laboratory findings metabolic (lactic) acidosis generally reported: "A high anion-gap (lactic-acid) metabolic acidosis is frequently found" (Holland, 1983).

Baud (1996a,b) relates lactate level to cyanide levels and the severity of acute cyanide poisoning.

5.4.4   Review of Case Reports

We reviewed 31 case reports from 14 different sources of reference (De Busk, 1969; Thomas, 1970; Moeschlin, 1980; Hausmann, 1982; Hall, 1983; Singh, 1989; Binder, 1991; Wesson, 1995; Chin, 2000; Lam and Lau, 2000; Gökhan, 2002; Mannaioni, 2002; Prieto, 2004; Stacie, 2006).

All cases were initially of unknown cause, while cyanide poisoning was considered during the course of intervention. The case reports vary in quality, so we tried to obtain the information that seemed essential to determining the cause and providing support for the choice of a particular therapy.

The ages of the patients ranged from 12 to 83 years. There were 10 female and 19 male patients.

Causes of poisoning and symptomatology

Twelve cases were occupational and thus fall under work-related accidents. Six suicide cases and ten criminal cases were reported. Three cases remained unexplained. Ten patients had chemistry or laboratory jobs, 5 were firemen and the others a painter, a disinfector, a tank cleaner, and an office clerk. During the course of observation, 19 patients were unconscious or became unconscious. The odour of bitter almonds was reported in six cases by medical personal or bystanders.

Eye and skin irritation was present in seven cases, one of which showed macular erythematous lesions. Five patients complained of throat discomfort. Dilated pupils or low GCS was observed in 11 cases; 8 patients reported vertigo and dizziness. Other neurological symptoms observed were cloni and seizures, tingling, and numbness of the hands.

Chest discomfort and angina were reported in two cases. Collapse and hypotension was observed in 10 cases. Nausea and vomiting did occur in two cases, and there was one case of hyperventilation reported. Red skin or flushing was reported in seven cases, cyanosis or greyish skin in two cases.

Reported heart rates were at a relatively mild tachycardia at 96 to 136 a minute. ECG findings were atrial fibrillation in three cases, and ST segment changes in four cases. One case of QT shortening was reported, and one patient showed signs of anterolateral ischemia.

Laboratory findings revealed acidosis (lactic acidosis, metabolic acidosis) in 13 cases.

The following tables give an overview about these 31 cases (De Busk, 1969; Thomas, 1970; Moeschlin, 1980; Hausmann, 1982; Hall, 1983; Singh, 1989; Binder, 1991; Wesson, 1995; Chin, 2000; Lam and Lau, 2000; Gökhan, 2002; Mannaioni, 2002; Prieto, 2004; Stacie, 2006). Causes, symptoms, PSSa classification and antidote use are presented (Tables 64, 65).

Table 64: Causes of poisoning in 31 cases

Job title	Occupational	Suicide	Uncertain	Criminal
Chemist (laboratory)	3	5	1	1
Fireman	5			
Painter	1			
Disinfector	1			
Tank cleaner	1			
Office clerk	1			
Other / Unknown		1	2	9
Total	12	6	3	10

Table 65: Clinical symptoms and findings in 31 cases

Unconsciousness during course of observation	19
Metabolic acidosis / lactic acidosis	13
Collapse and hypotension	10
Neurological findings	21
Dilated pupils, low Glasgow coma score	11
Cloni and seizures	2
Vertigo and dizziness	8
Red skin / flushing	7
Cyanosis	2
Odour of bitter almonds	6
Eye and skin irritation	7
Throat discomfort	5
Chest discomfort	2
Hyperventilation	1
Nausea, vomiting	2

Table 67: Poison Severity Score (PSSa) and symptomatology

Poison Severity Score (PSSa) (n)		Smell of bitter almonds	Eye or skin irritation	Throat discomfort	Nausea / vomiting / abdominal discomfort cramps	Neurological findings	Unconsciousness / unresponsiveness	Hyper-ventilation / difficulty breathing	Angina / chest discomfort	Hypo-tension / collapse	Red skin / flush	Cyanosis / grey color of skin	Metabolic acidosis / lactic acidosis
1	(6)	1	4	4	0	7	1	1	2	1	3	0	0
2	(2)	0	1	1	0	3	0	0	0	0	0	0	1
3	(10)	2	1	0	2	7	10	0	0	5	2	2	7
4	(13)	3	1	0	0	4	8	0	0	4	2	0	5
	(31)	6	7	5	2	21	19	1	2	10	7	2	13

Table 68: Poison severity score (PSSa) and use of antidotes

Poison Severity Score (PSSa) (n)		Amyl-nitrite	Sodium-nitrite	Sodium-thiosulphate	4-DMAP	Co-EDTA	HOCO	Oxygen	Other antidote (e.g. Narcan)	Other (e.g. gastric lavage, haemodialysis)	No cyanide antidote
1	(6)	0	0	1	0	0	0	5	0	0	1
2	(2)	1	1	1	0	0	0	2	0	0	0
3	(10)	2	6	7	0	0	1	10	2	5	0
4	(13)	0	1	1	0	1	0	7	1	1	6
	(31)	3	8	10	0	1	1	24	3	6	7

Use of antidotes

The most often used treatment was oxygen (24 out of 31 cases). STS was used in 10 and SN in eight cases. Three patients received AN, Co-EDTA, and HOCO each. No 4-DMAP was reportedly used. Three patients were initially treated with AN. Naloxon was used in three cases prior to cyanide antidotes in anticipation of an opioid poisoning. In six cases, other detoxification procedures (gastric lavage or haemodialysis) were carried out. Seven patients did not receive a cyanide antidote.

SN and STS were used primarily in severe cases: PSSa 3 or 4. Only two cases, ranked PSSa 1 and 2, involved antidote treatment with either SN or STS, or both. One case, PSSa 4, was treated with Co-EDTA. Hydroxocobalamin was used in one case, PSSa 3, but in combination with another antidote. All but one PSSa 1 to 3 cases received treatment with oxygen. Six cases, all PSSa 4, did not receive any antidotal treatment. These cases involved victims who may have been considered dead upon arrival or soon thereafter, so no further measures were taken.

5.4.5 Conclusion

The most common clinical findings of cyanide poisoning in initially uncertain cases are neurological symptoms and unconsciousness during the observation.

The laboratory finding most often reported is lactic acidosis. A relationship between amount of cyanide intake and lactic acidosis has been proposed.

However, neither the clinical symptoms nor the lactic acidosis are unique to cyanide poisoning.

In contrast, so-called classical findings, such as red or flushed skin, or the odour of bitter almonds, are apparently observed in a small number of cases only. If indicated, however, an odour may prove to be the ultimate clue for determining the cause of poisoning with an unknown substance (Hall and Rumack, 1986; Holland and Kozlowski, 1986).

Twelve out of these 31 cases of unknown poisoning were attributable to accidental exposure at work. In five out of six suicide cases involving cyanide, the patient had a chemistry or laboratory job. In 18 out of 31 cases ($\sim 58\%$), the patients' or victims' occupations were related to chemistry, laboratory, or fire fighting. Therefore, besides taking a general history, clinical diagnosis, and laboratory tests, the third party history and occupational history may be an extremely helpful, if not essential clue, for diagnosing cyanide poisoning.

Thus, the diagnosis of cyanide poisoning on purely clinical grounds will always remain a challenge. Baskin (1997) concluded that "These signs are not specific for cyanide poisoning,

which makes the distinction from other types of poisoning very difficult without the history of exposure".

It is therefore recommended that in the situation of an unknown poisoning antidote with significant side effects should not be used. Antidotes that may synergistically add to the effects of a possible other poison involved in the intoxication, like CO or any met-hemoglobin former, require caution. This speaks against the use of DCE all, and for caution in using nitrites and 4-DMAP which would not be the antidotes of first choice.

HOCO, possibly followed by STS, would be the safest option, AN followed by STS is also acceptable, as the met-hemoglobin formation by AN is limited. If these approaches are not (fully) successful and the patient remains in a critical state, careful application of SN or 4-DMAP, each followed by STS, can be considered.

6. CONCLUSIONS AND RECOMMENDATIONS

6.1   Classification of antidotes to cyanide and mode of action

Antidotes to cyanide have been developed as drugs for use in what are in reality relatively rare poisoning events. Furthermore, these events cannot be directly compared in many respects since they result from very different circumstances of exposure. The circumstances of poisoning vary from immediate onset (HCN and salts) to delayed, sometimes up to many hours later (nitriles and other cyanogens). They may also arise out of mixed exposure, such as in the case of smoke inhalation where there is co-exposure to CO, irritant gases and many other toxins plus oxygen depletion and heat from the fire. Consequently, it is impossible to give a recommendation for one single antidote for all cyanide poisoning situations since too many caveats would have to be included to account for, e.g.

- Is there an urgent need to counteract cyanide toxicity thereby necessitating both a fast acting antidote and rapid administration, i.e. without the need for preparation and infusion; or is cyanide liberation delayed sufficient for there to be enough time to use an antidote with slower onset of action?
- Is the intoxication life-threatening, thereby justifying significant side effects; or will the severity of poisoning be so mild that the side effects outweigh the risk from cyanide?
- Do storage requirements for transport and storage allow for worldwide dispersal and use of the antidote, or do specific requirements preclude use in some regions of the world, e.g. requirement for refrigerated storage in tropical countries?
- And finally, consideration of the availability and financial cost of antidote, including aspects of shelf-life and frequency of replacement etc., although from an ethical point of view this should not be paramount.

Consequently, the development of antidotes to cyanide never could, nor can, meet criteria usually required in the preclinical and clinical phases of new medications. No randomised double-blind studies, no phase 1 to phase 3 studies have been performed, or indeed are possible. Furthermore, since there are so very few cases of cyanide poisoning, observational studies of cohorts of patients receiving antidotes could not nor can be organised.

It is even more complicated, when the objective is to compare antidotes on a like-for-like basis. In clinical practice, there is no clinical study comparing the efficacy and effectiveness as well as the safety of the various antidotes under specified condition. Again, the low number of cases and the immediate need for action in the case of severe poisonings prevent and prohibit conventional comparative studies.

To date, therefore, preference for the use one antidote over another has drawn on expert-based opinion rather than on evidence-based medicine. This is further complicated by the existance of country-specific tendencies to use specific antidotes. Only the combination AN/SN/STS has been used worldwide, and still is in use in many regions. Germany (and a few neighbouring countries) have been using DMAP/STS, the UK has tended to use Co-EDTA. The latter has also been used in France, but has been replaced by HOCO; which again, is finding increasing favour in other countries / regions, especially in Europe and the USA.

These commonly used antidotes aim at modifying the toxicokinetics of cyanide poisoning in one of two different ways.

- Displacement of cyanide from the cells to the blood in the case of MetHb forming agents (AN, SN, DMAP) and HOCO, or
- Enhancing elimination of cyanide from the body by enzymatic as well as non-enzymatic detoxification by STS, Co-EDTA and HOCO.

There are apparent gaps in knowledge relating to how nitrites are effective without inducing significant MetHb levels. Additonal mechanisms involving alteration of nitric oxide (NO) have been implicated, yet the mechanism is unclear.The nitrites are either supposed to form a complex with NO, or to increase the NO levels. However, clinical use does not indicate problems, so the NO involvement may be irrelevant.

The common toxicokinetic mechanism of action of antidotes to cyanide does, however, allow comparison of the basis of efficacy, effectiveness, toxicity and safety.

Finally, oxygen cannot be regarded as a true cyanide antidote, though it is obvious that its use will increase the effect of other antidotes, thus making oxygen an indispensable mainstay of any treatment of symptomatic cyanide poisoning.

6.2 Practicality

Regarding practicality of the antidotes pharmaceutical preparation, storage conditions, shelf life, route of application and costs have to be considered (Table 69).

Table 69: Practical considerations

Antidote	Pharmaceutical preparation	Storage conditions	Shelf life	Route of application	Costs per single dose
STS	a. Ampoules 10 ml 10% b. Infusion vials 100 ml 25%	15 - 25°C	a. 3 y b. 2 y	Intravenous a. injection or b. infusion	a. 3.86 €(D) b. 22.69 €(D)
Cyanide Antidote Kit (AN, SN, STS) – 2 doses		< 40 °C, preferably 15-30 °C	1.5 y	See below for substances	213.31 US$ (USA)
AN	12 breakable ampoules (0.3 ml each) for inhalation	See above for kit	See above for kit	Inhalation, 1 ampoule for 15 - 30 seconds, every minute until SN available	See above for kit
SN	2 ampoules 10 ml (300 mg each)	See above for kit	See above for kit	Intravenous injection of 300 mg over 2 - 5 minutes	See above for kit
STS	2x50 ml vials (12.5 g each)	See above for kit	See above for kit	Intravenous injection of 12.5 g over 10 minutes after SN	See above for kit
DMAP	Ampoules 5 ml (250 mg)	Max 20°C, dark	3 y	Intravenous injection (3 - 4 mg/kg,), followed by STS 100 - 500 mg/kg	41,41 €(D)
Co-EDTA	Ampoules 20 ml (300 mg)	25°C, dark	3 y	Intravenous injection	31.41 US$ (CDN)
HOCO	2x250 ml (2.5 g each) glass vials, to be reconstituted with NaCl 0.9% 100 ml each = 1 dose		3 y	Intravenous infusion over 15 minutes	844,48 €(D)

Note: Cost estimated on July 4, 2013 (for the Cyanide Antidote Kit April 30, 2012), internet.
At least for DMAP availability is limited (to certain countries, mainly in Europe). For other antidotes this needs verification.

Safety (intrinsic effects)

Oxygen is safe when administered over limited periods of time. However, as stated previously, it is not a cyanide antidote, but rather a basic supportive treatment.

6.2.2   STS

STS has few intrinsic toxic effects and these are relatively non-serious. They include nausea / vomiting, headache, and a tendency towards hypotension due to the formation of thiocyanate. One study has reported further side effects, but these have not been reported elsewhere in spite of comparatively frequent use.

6.2.3   Methaemoglobin-forming agents

For the MetHb forming agents AN, SN and DMAP the formation of MetHb could be regarded as a 'toxic' effect, but this, rather than being a side effect ,is the intended therapeutical principle or mode of action. In overdose MetHb will impair oxygen transport and cause haemolysis,

particularly in individuals suffering from G-6-PDH deficiency. G-6-PDH deficiency is the most common human enzyme defect, being present in more than 400 million people worldwide with a greater prevalence in malaria regions such as Africa and SE Asia (Cappelini and Fiorelli, 2008). In a life-threatening poisoning situation the benefit of administration of a MetHb former will outweigh the risk, but this will not be the case of mild intoxication for example following ingestion of cyanogenic plants like cassava in Africa.

Nitrites

The nitrites, particularly SN, decrease blood pressure as a consequence of vasodilatation, which is in itself not desirable in cyanide poisoning since this would predispose to a tendency toward circulatory failure. However, it can be corrected by the standard emergency treatment of hypotension / shock with catecholamines. Also headache and flushing can occur, which are irrelevant compared to cyanide poisoning.

DMAP

Besides the MetHb formation, DMAP will induce haemolysis to a mild, non-significant, degree at recommended doses and to a higher degree in overdose.

In toddlers and small children MetHb reductase is not fully effective, so administration of MetHb formers require special caution, if they are to be used at all. The maker of 4-DMAP recommends a dose of 3.25 mg/kg bw for children (3-4 mg/kgKG for adults; Product Info Koehler Chemie). Antidotes for excessive MetHb formation are available (toluidine blue and methylene blue), but their administration will prompt cyanide liberation from met.Hb.

6.2.4   Co-EDTA

Co-EDTA frequently induces severe side effects, including extensive urticaria; angiotic oedema, which may result in life-threatening dyspnoea; ventricular rhythm disturbances; severe hypertension; and seizures requiring medical intervention. These side effects are more frequently observed in non-cyanide poisoned patients, which is consistent with cyanide binding apparently 'detoxifying' Co-EDTA. Consequently, cyanide poisoning should be certain before Co-EDTA is administered, if at all. Even then adverse effects may occur in cyanide poisoned humans.

6.2.5   HOCO

HOCO can increase blood pressure to a mild to moderate degree. This side effect, however, may be rather desirable in cyanide poisoning since cyanide causes hypotension and circulatory failure. Anaphylactic allergies have been described, but such can be treated with usual anti-allergic emergency measures. Other adverse effects are related to the discoloration of skin, plasma and urine from HOCO and cyanocobalamin. This transient discoloration may interfere with laboratory analyses and haemodialysis machines, and give rise in uninformed medical personnel to the suspicion of urinary tract bleeding or rhabdomyolysis. All of these, however, are no serious effects. In summary, regarding the frequency of occurrence of moderate to severe toxicity, the antidotes to cyanide can be ordered in their increasing risk of harm as as: STS < HOCO < DMAP < SN and AN < Co-EDTA.

6.3   *Efficacy and effectiveness*

As stated, any conclusions about the efficacy and effectiveness of antidotes to cyanide will have to be drawn from experience-based rather than evidence-based knowledge. Therefore, it is important to consider the frequency of use of the antidotes alone, and in combination, whatever the circumstances. The available data allowed us to identify a total of 468 "sequential poisoning severity scores" from 289 cases.

This was comprised of 116 sequencies in which the antidote was used alone):

- STS (n = 34),
- Co-EDTA (n = 29),
- HOCO (n = 25),
- AN (n = 16),
- DMAP (n = 2).

Noteworthy, SN was never reported to have been used alone.

There were 352 sequences, when antidotes were used in combination[a]:

- STS (n = 136),
- SN (n = 90),
- AN (n = 58),

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[a] Combination: where more than one antidote had been given with a clear intervals between successive treatments enabling discrete treatments to be identified.

- DMAP (n = 28),
- HOCO (n = 22), and
- Co-EDTA (n = 18).

Oxygen has been regarded as basic life support treatment of patients presenting, or at risk of presenting, cyanide-induced organ dysfunction so it is not possible to assess its frequency of use. In the past, when oxygen was not yet included in basic life support, in 95 sequences assessed for PSSa cyanide poisoning was treated with an antidote to cyanide but without oxygen.

6.3.1 STS

STS has been found to be completely efficient and resulted in complete recovery in moderate poisonings. In contrast there are insufficient data on the use of STS alone in severe poisoning cases to allow an assessment of its efficiency in this category.

Although the reaction of thiocyanate formation with rhodanese is relatively slow, STS appears to have an effect earlier than previously assumed. In animal experiments STS had an effect within 4 to 10 minutes even after lethal cyanide doses (Schubert and Brill, 1968; Friedberg and Grützmacher, 1968; Schwarzkopf and Friedberg, 1971). For life-threatening poisonings STS is not effective on its own, but is effective if given in combination after a direct (enzymatic or non-enzymatic) antidote such as the MetHb forming nitrites and DMAP, and potentially also HOCO. For Co-EDTA the situation is less clear.

For cyanogens, early administration of STS may have a prophylactic role against cyanide poisoning in SNP treatment as well as a therapeutic role in the case of delayed cyanide formation, e.g. from nitriles, laetrile / amygdalin or cassava, MetHb and maybe also cyanocobalamin. In these situations STS will scavenge cyanide ions as they are liberated converting them to thiocyanate which is subsequently eliminated via the kidneys. Delayed treatment after occurrence of acute and severe symptoms of cyanide poisoning will require the initial administration of a direct acting antidote.

6.3.2 Amyl nitrite

AN somewhat surprisingly has been found to be both efficacious and effective in cyanide poisoning, both when administered alone and in combination with other antidotes. Even severe, at least more than moderate, poisoning cases could be treated successfully with AN (mean PSSa 2.5 ≥ 0 for AN only, mean PSSa ≥ 0 in combination). AN appears to work better, if given early in the therapy, since administration after other antidotes seems to be less effective.

The ease of administration of AN; by simply breakable ampoules into the breathing air or directly into the mask or intratracheal tube as in the case with mechanical ventilation, points to making it an ideal first-line antidote and also one of the two antidotes that can be administered by first aiders and medical personnel not allowed to administer i.v. drugs. Unfortunately, AN appears to be no longer available as a medication or antidote in some countries (USA, UK, Germany), so it may be difficult to obtain.

6.3.3   Sodium nitrite

SN has never been reported as having been used as an antidote alone. In combination with other antidotes, primarily STS, it is clearly efficacious and effective in severe poisonings, even when given late in the course of poisoning and after unsuccessful application of other antidotes. In nearly all cases it has been adminsitered before STS, which is rational considering the action of STS (Section 6.4.1).

6.3.4   AN/SN/STS

The use of the 'cyanide antidote kit' has also been found to be efficacious and effective in severe poisonings, which is obvious since the individual substances have individually been found to be effective in severe (AN, SN) and moderate (STS) cyanide poisoning.

6.3.5   DMAP

DMAP also has been found to be effective in combination with other antidotes. The number of cases with DMAP alone, however, is too small for meaningful analysis. Given the recommendations of the manufacturer and the open literature to give STS after DMAP, it can be suspected that STS also has been applied in some, if not all of these few cases without having been mentioned.

From the Munich casuistics it can be concluded that DMAP can restore circulation even in (some) cases of cardiac arrest. However, as with other antidotes, neurological damage due to cerebral hypoxia often cannot be reversed.

DMAP can be administered by i.m. injection although this will be painful and there is a risk of necrosis at the site of injection (Baskin and Brewer, 1997). This may allow for its use in mass poisonings in desperate situations, if administered by medical personnel allowed to give i.m. injections.

6.3.6 Co-EDTA

While the case reports dealing with the treatment of cyanide poisoning with Co-EDTA clearly showed the efficacy and effectiveness of Co-EDTA, the severity of side effects occurring early after administration of Co-EDTA detract from the therapeutical effectiveness.

Due to the low number of cases in which Co-EDTA has been used alone or in combination with other antidotes its overall efficacy cannot be confirmed. Furthermore, there is insufficient clinical experience with Co-EDTA use alone in severe cyanide poisoning to judge its effectiveness. Indeed, the median PSSa before treatment with Co-EDTA alone, or in combination, was only two (see the corresponding PSSa analysis).

6.3.7 HOCO

From the PSSa evaluation it can be seen that HOCO also is efficacious and effective in severe poisoning cases both when administered on its own and in combination with any other antidotes, reversing PSSa of 3 to 0, if given in the first sequence i.e. early in treatment. For HOCO alone, this also holds true for sequence 2 (given as second treatment), while combinations (HOCO and other antidote) were not significantly successful.

Regarding combinations of HOCO with STS it is unclear whether HOCO contributes any effectiveness over and above that of STS alone, as both antidotes applied together may result in formation of an inefficient combination.

Further observational studies will be required to determine whether there is a different efficacy in reversing cardio-circulatory and neurological effects of cyanide poisoning, as seen for HOCO, but also for other antidotes.

6.4 Poisoning circumstances and antidotes

6.4.1 Direct poisonings – HCN and salts

For mild or moderate poisonings, antidote therapy does not seem to be more effective than symptomatic and supportive treatment. In such cases the administration of an antidote does not seem to be indicated. If an antidote had to be chosen, it should perhaps be STS based upon its good effect-risk ratio. The same holds true for HOCO although in such situations it is apparently not superior to STS.

In severe poisonings administration of STS alone is probably insufficient due to concern over its delayed onset of action and absence of clinical experience (clinical experience with STS used alone is limited to moderate poisoning). While HOCO is efficient in severe poisonings it does require additional time for preparation and infusion and immediate counteraction of cyanide toxicity is paramount in such situations.

As there is no problem with parallel CO poisoning in such cases, both nitrites and DMAP can be administered. AN, since it is administered by inhalation, has the advantage of being effective and bridging the time until an i.v. injection can be given.

In the case of the nitrites, side effects like blood pressure drop must be accounted for and the possibility of excessive MetHb formation must be kept in mind for both nitrites and DMAP.

Continued use of Co-EDTA is not recommended due to its significant inherent toxicity and the availability of less toxic alternatives.

6.4.2 Cyanogens

In the case of cassava poisoning there remains some uncertainty whether the reported poisonings and/or deaths are due to cyanide or linamarin. This prohibits the use of antidotes with significant side effects. Given that resources may be limited in countries where cassava poisoning can occur, STS is the most suitable option, if an antidote is to be administered. Only in very severe, life-threatening cases would administration of SN be an option. It is unlikely that either DMAP or HOCO would be available and even if they were the high cost of HOCO is likely to be prohibitary.

For laetrile / amygdalin poisoning, which often manifests with severe symptoms in a short timeframe, a direct acting antidote may be required. AN/SN/STS or HOCO/STS are the preferred options, but DMAP/STS would also be suitable if available. In mild to moderate poisoning cases no antidote seems to be required although administration of STS on its own can be considered.

Nitriles are associated with a delayed liberation of cyanide but this can be counteracted by administration of STS alone. In life-threatening cases any of the direct antidotes SN, HOCO or DMAP are indicated but these must be followed by administration of STS.

SNP should never be given without STS, as per long-standing recommendations. STS has been shown to effectively protect from cyanide poisoning. So no other antidote options need to be considered. In the unlikely event of STS not being co-administered and acute and severe cyanide poisoning occuring, any of the direct acting antidotes (SN, HOCO, DMAP) could be used.

6.4.3   Smoke

The main issue regarding smoke inhalations poisonings is the potential for co-exposure to CO and HCN, even though currently available data would suggest that CO is more important and critical in most fire situations. Since CO impairs oxygen transport by formation of CO-Hb, further impairment of oxygen transport capacity by administration of MetHb-forming agents would not seem advisable. Indeed, this is the general advice of subject matter experts, even though few, if any, problems have been seen with the infrequent administration of such antidotes (at correct dosage) in smoke inhalation. Notwithstanding this, the advice for treatment of severe smoke inhalation cases with a reasonable suspicion of cyanide poisoning must be administration of HOCO, followed by STS. In mild to moderate cases STS alone can be regarded as sufficient to account for the cyanide poisoning component.

6.4.4   Unknown poisonings

In mild to moderate cases STS can be given as a precautionary trial treatment, though generally no antidote seems to be required. In severe cases direct antidotes followed by STS would be recommended. As impairment of oxygen transport is not to be expected, both HOCO and the MetHb formers AN/SN and DMAP can be considered. From the point of view that treatment will be on a trial basis in this scenario, AN or HOCO should perhaps be the antidotes of first choice, since any improvement would be expected to be seen quickly in the case of true cyanide poisoning and this would allow for the option of following up with SN or DMAP if necessary.

Two specific scenarios seem to merit special consideration:

1. Children: As MetHb reductase is not fully active in toddlers and infants, nitrites and DMAP should not be given, despite the manufacturer of DMAP indicating a recommended dose for children. This leaves HOCO and STS as the preferred antidotes for the treatment of children.
2. Mass poisoning (e.g. terrorist attacks): AN (inhalation) and DMAP (i.m. injection) are the only antidotes, that seem to be feasible in mass poisoning since there would be an urgent need for antidotal treatment of larger numbers of individuals, especially by emergency medical staff not allowed to give i.v. injections.

6.5   Summary

To summarise, these recommendations are presented in Table 70:

Table 70: Antidotes recommended for acute poisoning by cyanides

Circumstance	Poisoning	
	Mild or moderate	**Severe**
Direct		
HCN or its salts	None or STS (HOCO)	AN/SN or DMAP, followed by STS
Cyanogenic compound		
Cassava	None or STS	STS, in very rare most severe cases SN followed by STS
Laetrile, Amygdalin	None or STS	AN/SN/STS or HOCO(/STS) (or DMAP/STS)
Nitriles	None or STS	STS, in very severe cases SN/STS or HOCO(/STS) or DMAP/STS
SNP	STS	STS
Smoke		
CO and **cyanide**	None or STS	HOCO (followed by STS). Neither AN/SN nor DMAP
Unknown	None or STS	HOCO or AN, followed by STS. If then required SN/STS (or DMAP/STS)
Child	None or STS	HOCO and/or STS
Mass poisoning	None	AN (or DMAP i.m.)
First aider	None	AN

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