FATAL AND RECURRENT METHEMOGLOBINEMIA AFTER INGESTION OF PRINTING REDUCER DYE (NITROBENZENE)

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ABSTRACT: Nitrobenzene, a pale yellow oily liquid with an odour of bitter almonds is used as intermediate in the synthesis of aniline dyes, and as a solvent for the manufacture of cellulose ethers and acetate, as a flavouring agent and in rubber industry. Nitrobenzene induces methemoglobinemia and this is responsible for the manifestations. It is important to take care of the secondary cycling of nitrobenzene from body stores in patients presenting late, after heavy exposure. Clues for diagnosis are a history of chemical ingestion. The characteristic smell of almonds, central cyanosis with no apparent respiratory distress, low pulse oximeter (observed) oxygen saturation with normal ABG (calculated) oxygen saturation and persisting cyanosis on oxygen therapy, without severe cardiopulmonary disease. Dark brown blood that fails to turn bright red on shaking suggests methemoglobinemia and this is supported by chocolate red colour of dried blood. To manage chemically induced methemoglobinemia properly, a clinician must be aware of its pathophysiology, be skilled with the use of reducing agents such as methylene blue, and understand specific physiochemical properties of the toxin. Acute poisoning with nitrobenzene is presented where clinical evaluation and timely management, with repeated intravenous methylene blue helped to save a life.
KEY WORDS: Nitrobenzene, Methemoglobinemia, Methylene blue, ABG, Ascorbic acid, G6PD.

INTRODUCTION: Acute poisoning with chemicals causing significant methemoglobinemia is uncommon but life threatening emergency requiring immediate definitive management, strongly suspected on clinical grounds that may change the patient's outcome.

CASE REPORT: A 23-year-old previously healthy married woman arrived at Sri Aurobindo hospital intensive care unit two hours after consumption of approx 200ml of pale yellow liquid used as a screen-printing reducer dye at around 3.30 pm on 10/11/2012. She was pale,

conscious, disoriented, irritable, tachypneic, cyanotic with cold clammy extremities, had 2-3 episodes of vomiting and there was a bitter almond like pungent smell from patient's body.

On examination BP was 80/60, pulse 130/min, respiratory rate 27/min, afebrile, and blood sugar 70 mg %, on auscultation lungs had few scattered rhonchi, abdomen and cardiovascular systems were normal. Treatment was started with intranasal oxygen, dextrose IV fluids, nebulisation, and intravenous hydrocortisone stat was given. Gastric lavage done with normal saline and purgation with polyethylene glycol administered. Later the relatives disclosed that the ingested reducer dye contained 20% nitrobenzene and a C9 aromatic hydrocarbon and same was confirmed by toxicological analysis of gastric lavage by spectrophotometery. [1] ABG Blood samples had a chocolate brown colour. Electrocardiogram (ECG) showed sinus tachycardia and X-ray chest was normal . Even before ABG (done by cobas b 221-blood gas system by Roche) report arrived, looking at the available clinical evidence, diagnosis of chemically induced methemoglobinemia was made and intravenously 100mg of methylene blue and 500 mg of ascorbic acid administered. ABG done with patient breathing 100% oxygen via oxygen mask showed a partial arterial oxygen pressure (PaO_2) of 120 mm of mercury, methemoglobin levels 72% and an oxygen saturation of 32% (Table 1). Ten minutes after administration of methylene blue, her colour improved and became more responsive. Two hours later repeat ABG showed a PaO_2 of 150 mm of mercury, a pH of 7.390, methemoglobin levels 49.1% with an oxygen saturation of 64% (Table 1).

Investigations done revealed haematocrit of 42.2%, haemoglobin 13 gm %, white blood count of 18.2×10^9/L (Table 2) with polymorphonuclear leukocytosis and normal platelet counts. Urine showed blood test positive with red blood cells 60-80/hpf and liver function tests were normal. Intravenous antibiotic was started looking at increased counts. Over the next 24 hours, she received an additional 200 mg of methylene blue and 1 gm of ascorbic acid in 4 divided doses. After that patient's oxygen saturation was around 70% and methemoglobin level of 30%. Though alert and oriented, she had cyanotic appearing skin with dusky blue discoloration of entire integuments, nail beds, mucosal surfaces and lips. In the next 24 hrs, looking at the ABG and clinical status of the patient she received additional 200 mg of methylene blue and 1gm of ascorbic acid in two divided doses that resulted in reduction of methemoglobin levels to 24%. On third and fourth day methemoglobin levels again increased to around 35% for which she was given 250mg of intravenous methylene blue in five divided doses with 1.5gms of Vit C and after that the levels declined spontaneously over the rest of her hospital course.(Table 1)

Fourth day patient developed jaundice (rising bilirubin, aminotransferase levels and deranged prothrombin time) with a significant reduction in her haemoglobin and haematocrit values (Table 2) with increase in serum lactic dehydrogenase level and an uncorrected reticulocyte count of 3.0%. Peripheral blood smear showed normocytic, normochromic red blood cells that demonstrated polychromasia, anisocytosis, tear drop cells and bite cells consistent with haemolysis. There was marked leukocytosis with left shift up to myelocytes, occasional blast cells suggesting a leukemoid reaction. Coombs tests direct and indirect were negative and a glucose-6-phosphate dehydrogenase (G6PD) screening test showed no deficiency. Over next five days, the patient required three units of blood transfusion. Injection Vitamin K was given for deranged prothrombin time. Urine analysis showed microscopic hematuria. On 11th day after hospitalization patient was discharged in stable condition with normal haematological values and liver function tests.

DISCUSSION: Nitrobenzene, a pale yellow oily liquid with an odour of bitter almonds is used in the synthesis of aniline dyes and as a solvent. The first report of nitrobenzene poisoning came in 1886.[1] Nitrobenzene is slowly excreted in urine (65%) and faeces (15%) either unchanged or as the major metabolites *p*-aminophenol and *p*-nitrophenol. The lethal dose ranges from 1 to 10 g, but there are no consistent reports regarding fatalities and the dose of ingestion. [1, 2]

Acute exposure to nitrobenzene leads to the development of methemoglobinemia, a condition in which the iron within the haemoglobin is oxidized from ferrous (Fe^{2+}) to ferric (Fe^{3+}) state, resulting in the inability to transport oxygen and thus causing a brownish discoloration of the blood.[2,3]

Systemic manifestations of methemoglobinemia at different blood levels are as follows, at 10-15%, cyanosis alone is obvious, though asymptomatic. Beyond 20%, headache, dyspnoea, chest pain, tachypnea and tachycardia develop. At 40-50%, confusion, lethargy and metabolic acidosis occur and fractions around 70% are fatal.[4,5] Anaemic patients and those with G6PD enzyme deficiency suffer more severe symptoms.[6] Other effects include hepatosplenomegaly, altered liver functions, renal failure, Heinz body haemolytic anaemia[7] and contact dermatitis.[8] The onset of methemoglobinemia may be delayed for 1 to 4 hours post exposure and in some cases haemolytic anaemia may develop approximately 4 to 5 days post-exposure as was the case in our patient.

Systemic redistribution of nitrobenzene from tissue stores is likely. Initial prehepatic intestinal nitro reduction of nitrobenzene may be most important factor in generation of early methemoglobinemia as it is 150 times faster than the hepatic microsomal nitroreductase.[1,8] Recurrent methemoglobinemia suggested by persistence of significant methemoglobin levels up to 72-96 hours after exposure to nitrobenzene is rare but as our patient's ingestion quantity was large, so it is likely that the metabolism of parent compound and active metabolites was saturated because of slow hepatic microsomal nitroreductase rates leading to prolonged exposure to the active metabolite and a prolonged zero-order production of a toxic metabolite of nitrobenzene causing persistent oxidative stress.[8] So this cycling of nitrobenzene from body stores becomes a requirement for prolonged treatment.

Methemoglobinemia management can be classified into five categories:(1)reducing toxin's systemic absorption by induction of emesis with ipecac syrup / salt water and facilitating removal of toxin from gastrointestinal tract by gastric lavage, activated charcoal and a purgative (2) reduction of methemoglobin to haemoglobin via reducing agents,(3) treatment of the "functional anaemia" (hypoxic state) with hyperbaric oxygen,(4) extracorporeal removal of the chemical and (5) replacement of methemoglobin with a functional oxygen-carrying pigment.[5]

Clothing's has to be removed and changed to prevent percutaneous absorption. In the case reported, we used gastric lavage and a purgative to reduce systemic absorption.

The definitive treatment of methemoglobinemia is the use of the reducing agent, methylene blue whose action is dependent on production of the reduced form of nicotinamide adenine dinucleotide phosphate (NADPH) by the hexose phosphate shunt and the activity of the enzyme, NADPH-methemoglobin reductase. NADPH is necessary for the reduction of methylene blue to leukomethylene blue, which is responsible for the reduction of methemoglobin into haemoglobin. G6PD deficiency leads to an impaired NADPH production within erythrocytes and precipitation of a Heinz-body haemolytic episode.[6,7] Excessive methylene blue may itself

provoke the formation of methemoglobin because of its own oxidative potential and produce a blue discoloration of the skin and bodily secretions, inducing an "apparent cyanosis".[9,10]

Role of ascorbic acid in reducing methemoglobinemia is controversial as its action is slow and offers little advantage over normal endogenous reduction of methemoglobin.

Methylene blue is available as 1% 10 ml vial and for the initial management of methemoglobinemia recommended dose is 1 to 2 mg/kg to a maximum of 5 to 7 mg/kg /day. As maximal response to methylene blue usually occurs within 30 to 60 minutes; therefore, methemoglobin levels should be monitored and repeat doses of methylene blue should be spaced at least one hour apart and after evaluating the response to the last dose. G6PD deficiency should be considered if a patient has a negligible initial response to a therapeutic dose of methylene blue. [6] Methemoglobin levels should be continuously monitored as nitrobenzene has the potential for continued methemoglobin production.[5,8]

N-acetylcysteine has a controversial role in reducing methemoglobin so its use not yet approved.[11] Exchange transfusion is indicated in severe cases[12] but we didn't require it in our patient as she improved with reducing agents and other supportive care. Hyperbaric oxygen is reserved for patients who have a methemoglobin level > 50% and or those who do not respond to standard treatment. [9]

In this case, repeated doses of methylene blue helped in tiding over the fluctuating symptoms due to the release of nitrobenzene from the body stores, without exceeding the maximum dose. Fresh blood transfusion improved the oxygen carrying capacity and haemoglobin content, improving the patient symptomatically. Taking care of nutrition, adequate urine output and hepatoprotection prevented kidney and liver failure, which have been cited as late effects.

CONCLUSION: Chemically induced methemoglobinemia is a life-threatening condition which requires immediate and definitive management. Therefore a clinician must be aware of its pathophysiology, be very skilled with the use of reducing agents such as methylene blue, ascorbic acid and other modalities of treatment like exchange transfusion and hyperbaric oxygen therapy that are usually reserved for patients who are resistant to standard treatment and understand specific physiochemical properties of the toxin especially nitrobenzene's potential for continued methemoglobin production. The authors also wish to point out the non availability of intravenous methylene blue should not be a hindrance, as methylene blue powder is available and can be made into 1% solution and sterilized in CSSD (Central Sterile Supply Department).[13]

REFERENCES:

1. Beauchamp RO, Jr, Irons RD, Rickert DE, et al. A critical review of the literature on nitrobenzene toxicity. Crit Rev Toxicol 1982; 11: 33–84.
2. Gupta G, Poddar B, Salaria M, Parmar V. Acute nitrobenzene poisoning. Indian Pediatr 2000;37:1147-8.
3. Chongtham DS, Phurailatpam, Singh MM, Singh TR. Methemoglobinemia in nitrobenzene poisoning. J Postgrad Med 1997;43:73-4.
4. Dutta R, Dube SK, Mishra LD, Singh AP, Acute methemoglobinemia. Internet J Emerg Intensive Care Med 2008;11:1092-4051.
5. Ajmani A, Prakash SK, Jain SK, et al. Acquired methemoglobinemia following nitrobenzene poisoning. J Assoc Physicians India 1986; 34: 891–2.

6. Rosen PJ, Johnson C, McGehee WG, et al. Failure of methylene blue treatment in toxic methemoglobinemia. Association with glucose-6-phosphate dehydrogenase deficiency. Ann Intern Med 1971; 75: 83–6.
7. Yoo D, Lessin LS. Drug-associated "bite cell" haemolytic anaemia. Am J Med 1992; 92: 243–8.
8. Holder JW. Nitrobenzene carcinogenicity in animals and human hazard evaluation. Toxicol Ind Health 1999; 15: 445–57.
9. Wright RO, Lewander WJ, Woolf AD.: Methemoglobinemia: etiology, pharmacology, and clinical management. Ann Emerg Med 1999; 34: 646–56.
10. Wright RO, Magnani B, Shannon MW, Woolf AD. N-acetylcysteine reduces methemoglobin in vitro. Ann Emerg Med. 1996 Nov;28(5):499-503.
11. Mier RJ. Treatment of aniline poisoning with exchange transfusion. J Toxicol Clin Toxicol. 1988;26(5-6):357-64.
12. Harvey JW, Keitt AS. Studies of the efficacy and potential hazards of methylene blue therapy in aniline induced methemoglobinemia. Br J Hematol 1983; 54: 29-41.
13. Dutta R, Dube SK, Mishra LD, Singh AP. Acute methemoglobinemia. Internet J Emerg Intensive Care Med. 2008; 11:1092-4051.

TABLE 1:- Oxygen Saturation Values, ph, methemoglobin and blood lactate levels and Doses of Methylene Blue Administered During the First 6 days after ingestion of Nitrobenzene.

Time after Ingestion (hours)	ph	PaO_2(mm Hg)	Oxygen Saturation (percent)	HCO_3^- mmol/L	Methemoglobin Level (percent of Total pigment)	Blood lactate (mmol/L)	Dose of Methylene Blue (mg)
2	7.32	120	32	18.1	72	8.7	100
4	7.38	150	64	18.6	49.1	3.7	-
6	7.39	105	50	19.1	54	4.1	100
12	7.39	112	66	19.5	28.9	3.1	-
18	7.38	115	52	19.9	42	4.5	100
24	7.39	151	70	20.1	30	2.9	-
30	7.38	121	72	20.3	38.3	3.1	100
36	7.39	120	74	21	32.1	2.1	-
42	7.39	118	63	20.3	39.7	3.0	100
48 (2nd day)	7.39	120	79	22	24	2.5	-
54	7.41	121	72	21.6	35.9	2.6	50
60	7.42	123	74	21.8	31.9	2.8	50
66	7.41	131	78	21.5	29.8	2.2	50
72 (3rd day)	7.43	115	81	21.1	24.5	2.1	-
78	7.45	134	75	22	33.2	2.2	50
84	7.41	121	79	23.2	29.5	1.9	50
90	7.4	130	85	23	24	1.6	-
96 (4th day)	7.41	125	90	24.6	19.1	1.6	-
108	7.42	135	96	24	15.4	1.5	-
120 (5th day)	7.42	130	98	23.9	08	1.4	-
144 (6th day)	7.41	110	98.6	24	03.1	1.4	-

TABLE 2:-Hematologic and Liver function test Values

Days After Ingestion	Haemoglobin (grams Per dl)	Haematocrit (percent)	Serum Bilirubin Direct/Total (mg per dl)	Serum AST* (IU per litre)	Units of Blood transfusion	TLC
Initial	13	42.2	1/1.5	42	-	18,200
1	11.6	36	-	55	-	19,800
2	9.5	29	-	-	-	18,500
3	7.5	24	-	-	1	17,200
4	6.9	22.5	1.5/3.5	125	1	15,900
5	7.7	24.3	2.1/5.2	210	1	12,200
6	8.5	27.1	2.8/6.1	247	-	11,000
7	9	28	2.1/4.2	217	-	10,500
8	9.3	28.5	1.9/3.2	128	-	9,200
9	-	-	-	-	-	-
10	-	-	-	-	-	-
11	9.5	29.2	1.3/2.1	50		8,300

AST = aspartate amino transferase.

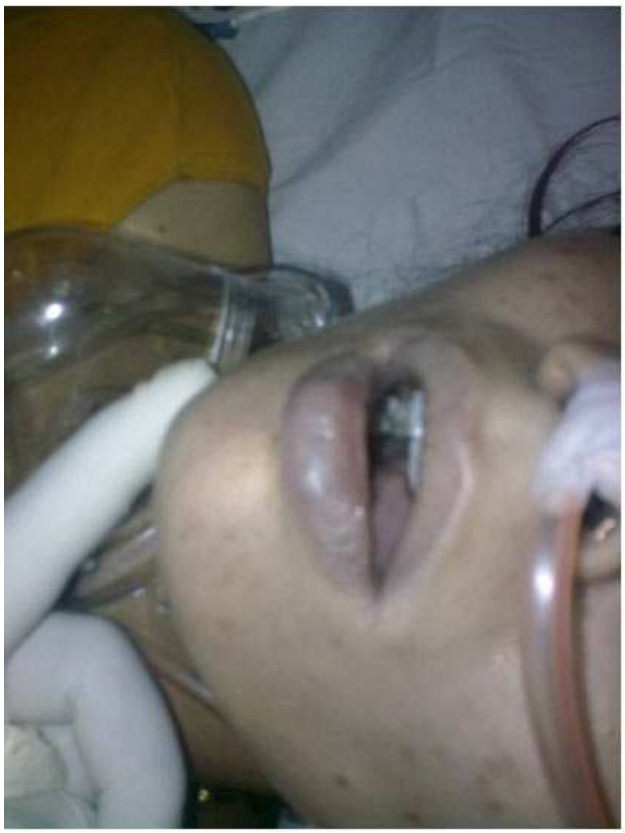

Figure 1 : Cyanotic lips in a patient with Methemoglobinemia and hypoxia

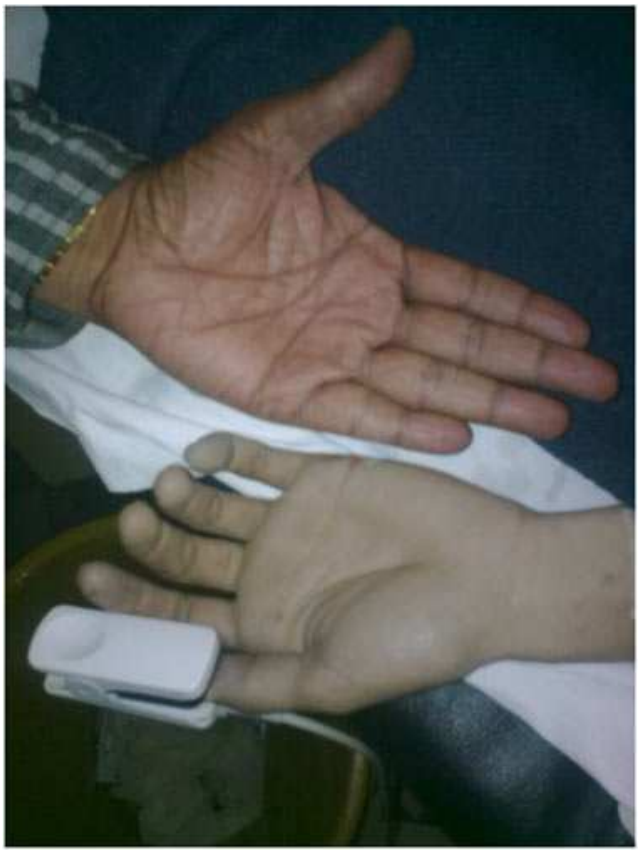

Figure 2: Severe peripheral cyanosis (the hand with SpO_2 probe in situ is of patient's) following nitrobenzene poisoning.

Figure 3: Choclate brown colour of blood drawn for investigation purpose.

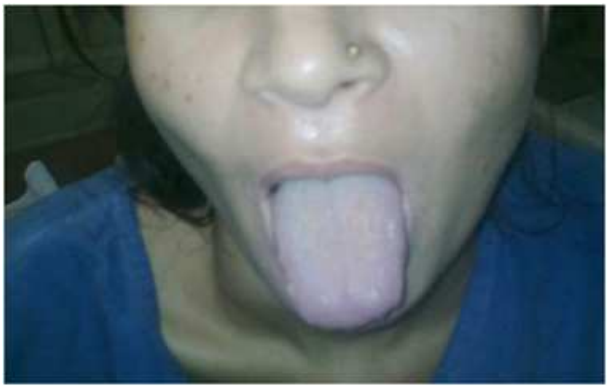

Figure 4 : Cyanosis improved after treatment of Methemoglobinemia and hypoxia