Congenital Methemoglobinemia*

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ABSTRACT

This report a case of a ten-year-old female with progressive cyanosis and dyspnea on exertion. Clinical and laboratory work up ruled out a cardiac and pulmonary pathology warranting further investigation for possible hemoglobinopathies. Enzyme assay showed deficiency in cytochrome b5 reductase seen in patients with congenital methemoglobinemia. Ascorbic acid at 200mg daily afforded gradual improvement in cyanosis.

Keywords: *methemoglobinemia, cytochrome b5 reductase, cyanosis, ascorbic acid, child*

INTRODUCTION

Congenital methemoglobinemia is a rare autosomal recessive condition that is secondary to a cytochrome b5 reductase deficiency. This report aims to present and discuss the clinical presentation, pathophysiology, diagnosis, and management of congenital methemoglobinemia.

CASE REPORT

E.A., a 10-year old female, Filipino, Roman Catholic from Novaliches, Quezon City who was brought in for consult in a tertiary institution due to cyanosis.

The patient was born to a then 32-year old G3P3 (3003) mother with no exposure to chemicals, teratogens, radiation or viral exanthem. The patient was noted to be dusky at birth, which persisted throughout infancy. The patient had regular well baby check-ups with a private physician. No intervention or work-up was done. On the interim, there was progressive cyanosis *(Figures 1, 2, 3, 4 - see appendix)*, easy fatigability and dyspnea on exertion.

Two months prior to consultation, there was persistence of the symptoms. The patient had ten episodes of non-projectile, non-bilious vomiting consisting of previously ingested food approximately ½ cup per bout. Complete blood count showed polycythemia. She was referred to a hematologist and a cardiologist. ECG showed sinus tachycardia while echocardiogram result was normal. A hemoglobin electrophoresis showed an equivocal result. She was referred to a tertiary center for further work-up hence the consultation.

There was no history of hematologic or cardiac disease in the family. The patient denied exposure to any chemical, pesticide or toxin. There was no history of recent travel. The past medical, nutritional, immunization and psychosocial histories were non-contributory.

Pertinent physical examination revealed a blood pressure of 100/70 mmHg. She was tachycardic at 118 beats per minute. The respiratory rate was 18 cycles per minute and the temperature was 36.8 °C. The anthropometric measurements were normal for age. The patient had dusky skin, cyanotic mucosa and nail beds. (Figure 5 - *see appendix*) There was no murmur, organomegaly or clubbing of the fingernails. The neurological examination was unremarkable.

The patient was seen by a hematologist and a pulmonologist. Chest x-ray showed normal result. Baseline oxygen saturation was 89% at room air. Oxygen supplementation was given with no improvement in saturation. (Table 1) Pulmonary pathology was unlikely at this time. Congenital methemoglobinemia was considered hence serum analysis for cytochrome b5 reductase enzyme activity was done, which showed a low enzyme level. (Table 2)

**1st Place, 2018 Philippine Medical Association Case Report Contest*
***From Philippine Children's Medical Center, Quezon City*

Table 1: Oxygen Saturations in Percentage, Heart rate in Cycles per minute And Different Oxygen Flow Rates

Date/Time	Device	Flow rate	O2 sat	Heart rate
9/24/2017 9AM	Room air		87-88%	120
	O2 prong	4lpm	89-90%	101
	O2 mask	5lpm	89%	101
	O2 mask	6lpm	89%	101
	O2 mask	7lpm	89%	103
	O2 mask	10lpm	89%	105-108

Table 2: Result of Cytochrome b5 reductase enzyme in Uh/ Hb with accompanying Reference Value

	Value	Unit	Reference Value
Methemoglobin Reductase, B (cytochrome b5 reductase)	1.4	U/g Hb	6.6-13.3

Ascorbic acid at 200 mg per day was started with gradual resolution of cyanosis *(Figures 6 and 7 - see appendix)* and improvement in oxygen saturation.

CASE DISCUSSION

We are presented with a 10-year old female with progressive cyanosis since birth. Cyanosis is a blue or purple appearance of the skin or mucous membranes caused by inadequately oxygenated blood that perfuse peripheral tissues. Cyanosis can also result from the presence of abnormal hemoglobin forms that are unable to bind oxygen to supply end organs and tissues.[1] Outlined in Table 3 are the differential diagnoses of cyanosis.

Part of the initial assessment of cyanosis is the initiation of oxygen therapy. Clinical improvement with oxygen therapy suggests diffusion impairment whereas non-improvement with high-concentration oxygen is suggestive of ventilation-perfusion mismatch, such as shunting from a consolidated pulmonary lobule or congenital heart disease with right to left shunting. Our patient did not respond to high-concentration oxygen support.[2]

The non-improvement of cyanosis of our patient with high concentration oxygen therapy warrants reassessment of the respiratory status. The absence of fever, respiratory distress and a normal chest and lung examination make pulmonary pathology a least likely cause of cyanosis in our patient. An unremarkable chest x-ray findings further strengthen a non-pulmonary pathology.[1,3]

Cardiac pathology may also be considered in patients with central cyanosis. Our patient did not present with easy fatigability, growth retardation and other findings suggestive of a cardiac disease. Cardiac examination was essentially normal. Radiologic and echocardiographic findings are unremarkable. Hence, in a patient presenting with persistent cyanosis in the absence of significant pulmonary or cardiac disease, the rare possibility of an abnormality in hemoglobin structure or disorders of oxygen affinity was entertained.[3,4,5]

It is imperative to discuss hemoglobin structure and synthesis. Hemoglobin is a tetrameric protein composed of 2 pairs of globin chains. Each globin chain, or subunit, is associated with a heme group in its center. The dominant form of adult hemoglobin is hemoglobin A (HbA), which is made up of 2 α chains and 2 β chains. The synthesis and structure of the different globin chains is under tight genetic control. Defects in these genes can cause the abnormal production of hemoglobin and anemias, a disorder called hemoglobinopathies. These genetic defects can result in structural defects in the hemoglobin molecule, diminished production of the hemoglobin subunits, or abnormal association of subunits.[6]

Hemoglobinopathies are classified according to the (I) qualitative nature of the resultant hemoglobin wherein there is production of structurally abnormal globin chains (ie, sickle cell disease) and the (II) quantitative amount of hemoglobin produced wherein there is structurally normal but decreased amount of globin chains (ie, thalassemia).[6,7] Most hemoglobinopathies are not clinically apparent, while others produce abnormal laboratory findings and a few cause serious disease.[6]

As enumerated in table 4, quantitative disorders of globin chain synthesis such as thalassemia in infants and children usually presents with pallor and does not present with cyanosis. Patients with thalassemia show a variable quantity of HbA_2 in haemoglobin electrophoresis. Our index patient presented with a normal hemoglobin electrophoresis which rule out quantitative disorders of globin chain synthesis.[8]

Qualitative disorders of globin structure produce mutations or alterations of the globin protein producing pronounced changes in the functional property of the hemoglobin specifically oxygen affinity and solubility. Structural hemoglobinopathies presenting with cyanosis include those with abnormalities with oxygen binding such as those with low oxygen affinity mutants (Hemoglobin M) and methemoglobinemia.[7] These are abnormalities that result from a change of its iron from the Ferrous form to its Ferric state which is unable to carry oxygen.[9]

Hemoglobin M is an autosomal dominant hemoglobin variant that causes cyanosis as a result of structural changes in the alpha or beta chains that stabilize the hemoglobin in the ferric state. In such cases, hemoglobin M may be differentiated from methemoglobin by its corresponding absorption spectrum in hemoglobin electrophoresis.[10] A normal hemoglobin electrophoresis in our index patient excludes hemoglobin M as the possible cause of cyanosis.

Congenital methemoglobinemia was considered in our patient due to the presence of central cyanosis and low oxygen saturations, which were unresponsive to oxygen therapy. Hence, serum analysis for cytochrome b5 reductase enzyme activity was done, which showed a low enzyme level.

To elucidate the importance of cytochrome b5 reductase enzyme in central cyanosis, let us discuss the role of this enzyme. Cytochrome b5 reductase, an enzyme system present within the RBCs, converts methemoglobin to hemoglobin. It maintains intracellular methemoglobin to less than 1%. Decreased activity of this enzyme promotes the accumulation of methemoglobin leading to central cyanosis and symptoms related to hypoxia. An alternative pathway that is not physiologically active utilizes nicotinamide adenine dinucleotide phosphate (NADPH) generated by glucose-6-phosphate dehydrogenase (G6PD) in the hexose monophosphate shunt. However, there is normally no electron carrier present in the red blood cells to interact with NADPH-methemoglobin reductase. Exogenously administered electron acceptors such as methylene blue are required for this pathway to be activated. This non-physiologic pathway becomes clinically important for the treatment of methemoglobinemia.[4]

Methemoglobinemia results from an oxidation-reduction imbalance, either due to (I) excessive oxidation of hemoglobin (increased production of methemoglobin) following exposure to various oxidant drugs/toxins or (II) decreased in the activity of reducing enzymes secondary to a genetic defect in red blood cell metabolism or hemoglobin structure. The newly formed methemoglobin causes an increase in its oxygen affinity, but a functional decrease in its oxygen binding capacity shifting the oxygen dissociation curve of the oxidized hemoglobin to the left which hinders the release of oxygen in the tissues. This now leads to tissue hypoxia and a relative or functional anemia (i.e., the amount of functional hemoglobin is less than the measured level of total hemoglobin) due to the reduction of free hemoglobin to transport oxygen to the tissues. Clinically, these two mechanisms will produce central cyanosis which is unresponsive to oxygen therapy.[10,11,12]

Methemoglobinemia can be congenital or acquired. Acute or acquired methemoglobinemia results from exposure to several oxidizing agents. (Table 5) There is no exact prevalence of methemoglobinemia but studies have shown that acquired methemoglobinemia are more frequent than congenital types. Symptoms in patients with acquired methemoglobinemia tend to be more severe than in patients with congenital methemoglobinemia.

Acquired methemoglobinemia is life-threatening when methemoglobin comprises more than 40% of total hemoglobin.[4] The diagnosis of acute methemoglobinemia is least likely in this case due to the chronic presentation of the symptoms and the absence of exposure to oxidizing agents, hence, congenital methemoglobinemia was considered.

Congenital methemoglobinemia results from either cytochrome b5 deficiency, cytochrome b5 reductase deficiency or hemoglobin M.[1] The gene regulating the synthesis of cytochrome b5 reductase has been localized to chromosome 22q13qter. A number of mutations have been identified.[13]

In our patient, a deficiency of the cytochrome b5 reductase is the cause of cyanosis as evidenced by a low enzyme level.

Clinical manifestations of methemoglobinemia reflect the reduction in oxygen-carrying capacity. The clinical hallmark is unexplained cyanosis and decreased oxygen saturation via pulse oximetry despite adequate ventilation and increased inspired oxygen concentration.[12,14] This manifestation was seen in our patient. The spectrum of skin pigmentation depends on the level of methemoglobin in the blood, the higher the methemoglobin level the more expressive the cyanosis.[12] In general, a fraction of methemoglobin fewer than 15% causes only a pale, grayish or bluish pigmentation of the skin which is frequently overlooked.[12]

An additional clinical sign among patients with methemoglobinemia is the presence of dark brown colored blood. Varying degrees of brownish discoloration of the blood occurs depending on the levels of methemoglobin.[12,14] As levels of methemoglobin increase, the patient evolves with a reduction in the level of consciousness, respiratory depression, shock, and death. Levels of methemoglobin above 70% are usually fatal.[12] Table 6 shows the correlation of methemoglobin and its clinical manifestations.

At birth, our patient manifested with dusky skin complexion. Based on the table, it is estimated that she has a 3-15% methemoglobin level. On consult, the patient manifested with visible cyanosis and dark colored blood associated with dyspnea on exertion. A 30% fractional methemoglobin level may be present. [13]

Several methods are available for detecting the presence of methemoglobinemia and assessing the severity of the disease. Arterial blood gas paired with oxygen saturation by pulse oximetry and serum methemoglobin levels are clinically utilized to make a diagnosis. In methemoglobinemia, arterial blood gas will show a high partial pressure of O2 (PaO2) with normal hemoglobin saturation (SaO2), with values well above those indicated by pulse oximetry.[12] In comparison, children with cyanotic heart disease who receive supplemental oxygen have a low partial pressure of oxygen and low calculated oxygen saturation. Methemoglobinemia is strongly suggested when there is clinical cyanosis in the presence of calculated normal arterial PaO2 as obtained by arterial blood gases.[4]

The gold standard in the diagnosis of methemoglobinemia is the use of a pulse co-oximeter[11] Unlike a pulse oximeter, which measures light absorbance at two wavelengths (oxyhemoglobin and deoxyhemoglobin), a co-oximeter measures light absorbance at four different wavelengths which correspond to the absorption characteristics of deoxyhemoglobin, oxyhemoglobin, carboxyhemoglobin, and methemoglobin. In patients presenting with cyanosis of uncertain cause, co-oximetry measurements are a valuable diagnostic tool.[15] The high cost of these machines is a hindrance to its availability. [11]

Measurement of the level of cytochrome b5 reductase activity or cytochrome b5 is recommended to distinguish cytochrome b5 reductase deficiency from cytochrome b5 deficiency.[4] These assays are not available in the local market. The author was able to coordinate to an international institution to determine the enzyme assay of our patient. The cytochrome b5 reductase enzyme level of our patient is 1.4 U/g Hb which is low compared to the reference value of 6.6-13.3 U/g Hb. This result is consistent with cytochrome b5 reductase deficiency.

Congenital methemoglobinemia secondary to NADH-cytochrome b5 reductase deficiency is usually benign and does not necessitate any treatment. However, for cosmetic reasons, treatment might be necessary.[1,4,5] Treatment options include methylene blue and ascorbic acid.[1,5,16]

Ascorbic acid is a potent antioxidant and reducing agent. It has direct reducing action on methemoglobin rather than restoration of the normal enzymatic reduction mechanism.[5] Daily oral treatment with ascorbic acid (200-500 mg/day in divided doses) gradually reduces the methemoglobin to

approximately 10% of the total pigment and alleviates cyanosis as long as therapy is continued. Some studies recommended the use of 300 to 600 mg of ascorbic acid given orally three times a day.[4,11,15] In this case report, our patient was started on ascorbic acid at 200 mg daily. There was improvement in skin discoloration and oxygen saturation measured via pulse oximetry from 89% pre-treatment to 90- 92%.

The use of ascorbic acid to treat toxic methemoglobinemia is not recommended. Some studies suggested that high dose ascorbic acid may be used at 10 grams intravenously every 6 hours for the treatment of symptomatic methemoglobinemia in patients without renal insufficiency when methylene blue is not available.[4] A retrospective chart review done in a large tertiary care pediatric hospital in Argentina involving 5 patients less than 18 years of age with confirmed methemoglobinemia significant enough to cause cyanosis and functional hypoxia. These patients were treated with 100% oxygen and high dose intravenous ascorbic acid. There were no side effects noted in all patients. There was marked improvement within 24 hours. None of the patients had any additional episodes of cyanosis. This retrospective study concludes that ascorbic acid can be used in the absence of methylene blue.[17] Concerns about kidney stone formation with ascorbic acid therapy remain unproven, although high-dose therapy may be associated with some risk. [4]

In case of life-threatening and toxic methemoglobinemia, definitive treatment is methylene blue through intravenous route. Methylene blue is a thiazine dye which has dose-dependent antiseptic and oxidizing properties. It serves as a cofactor for the enzyme NADPH-methemoglobin reductase utilized by the body when normal physiologic reductive pathways are overwhelmed. Methylene blue is administered intravenously initially at 1-2 mg/kg to treat toxic methemoglobinemia. It is administered as one percent solution over a five-minute interval and should not exceed 7mg/kg. It is in itself toxic and can cause dyspnea, chest pain, and hemolysis. This dose may be repeated at 1mg/kg every 30 minutes as necessary. An oral dose can be administered at 100-300 mg orally per day as maintenance therapy.[11] Methylene blue is not devoid of toxicity and its chronic administration can cause urinary tract irritation. It also colors the urine blue or green.[18] However, methylene blue may not always be accessible in our setting.

A treatment option for methemoglobinemia is the administration of riboflavin at a dose of 400mg once daily. Riboflavin utilizes the alternate pathway of methemoglobin reduction. The clinical experience with the use of Riboflavin in the management of methemoglobinemia is very limited. [4,16]

In patients with severe methemoglobinemia unresponsive to methylene blue, hyperbaric oxygen therapy and exchange-transfusion are recommended.[5]

Patients with hereditary methemoglobinemia like our patient are advised to avoid exposure to oxidizing agents that induce methemoglobinemia. Any further increase in their baseline methemoglobin level may be life-threatening.[4]

The prognosis of patients with congenital methemoglobinemia depends on the type of the disorder. Type I recessive congenital methemoglobinemia is a benign condition and is the most common form.[4,12,18,19]The life expectancy in this type is not lower than the general population. Pregnancies may develop normally and have not been complicated due to the disorder. Type II is much less common. It is associated with neurological manifestations that would usually begin in infancy. 11 Due to the benign nature of symptoms and the absence of any neurological manifestations, our patient has type I recessive congenital methemoglobinemia.

It is recommended that families of an affected child undergo enzymatic and molecular genetic testing. Prenatal diagnosis of type II disease has been performed successfully in some cases. It has a recurrence rate of 25%. [20] It is necessary to emphasize the importance of family counselling and referral to support groups or organizations to uphold the primary concerns and welfare of individuals with rare disorders. [21,22]

SUMMARY, CONCLUSION AND RECOMMENDATION

We reported a case of a ten year old female with progressive cyanosis unresponsive to oxygen supplementation. A low cytochrome b5 reductase assay confirmed the diagnosis of congenital methemoglobinemia. Ascorbic acid treatment at 200mg daily gradually improved clinical manifestations and oxygen saturation. Prevention of exposure to oxidizing agents is warranted to prevent life-threatening complications. The importance of history, clinical clues and a high index of suspicion are indispensable in arriving to a correct diagnosis even in a limited setting.

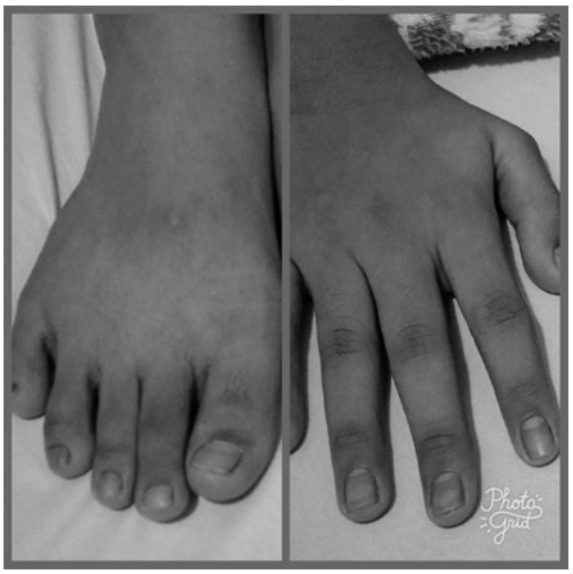
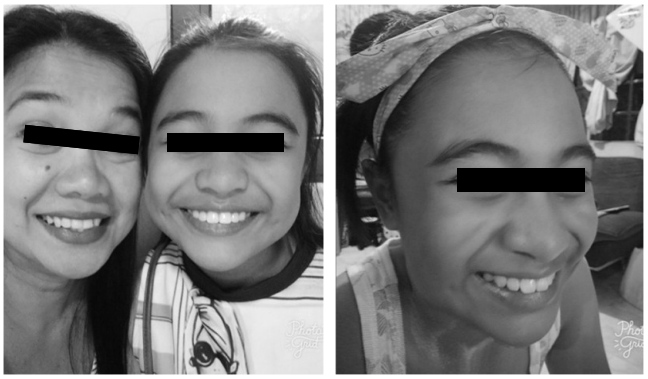


Figure 5: Picture of the right foot and hand of the patient presenting with cyanoitc nailbeds, absence of clubbing with a note of dusky skin complexion

APPENDIX

Figures 1,2,3,4: Pictures of the patient from infancy to childhood showing the progression of dusky skin complexion



Figures 6 and 7: Picture of the patient showing the gradual resolution of cyanosis

Table 3: Differential Diagnoses of Cyanosis as to Peripheral Cyanosis and Central Cyanosis

Peripheral Cyanosis	Central Cyanosis
Low cardiac output states Shock Left ventricular failure Hypovolemia	A. Decreased arterial oxygen saturation 1. High altitude (> 8000ft) 2. Impaired pulmonary function a. Hypoventilation b. Impaired oxygen diffusion c. Ventilation-perfusion mismatching - Pulmonary embolism - Acute respiratory distress syndrome - Pulmonary hypertension d. Respiratory compromise - upper airway obstruction - pneumonia
Environmental exposure (cold) Arterial occlusion Thrombosis Embolism Vasospasm (Raynaud's phenomenon) Peripheral Vascular Disease	B. Anatomic shunts 1. Pulmonary arteriovenous fistula and intrapulmonary shunts 2. Cerebral, hepatic, peripheral arteriovenous fistula 3. Cyanotic congenital heart disease
Venous Obstruction Redistribution of blood flow from extremities	C. Abnormal hemoglobin 1. Methemoglobinemia a. Hereditary b. Acquired 2. Sulfhemoglobinemia 3. Mutant hemoglobin with low oxygen affinity

Table 4: Classification of Hemoglobin Disorders

Quantitative Disorders of Globin Chain Synthesis	Qualitative disorders of globin structure: structural variants of hemoglobin
A. Beta Thalassemia B. Alpha Thalassemia C. De novo and acquired a-thalassemia	Sickle cell disorders Hemoglobin with decreased stability (unstable hemoglobin variants) Mutants causing congenita Heinz body hemolytic anemia Acquired instability—oxidant hemolysis: Drug induced, G6PD deficiency Hemoglobin with altered oxygen affinity High/ increased oxygen affinity states: - Fetal red cells - Decreased RBC 2,3 BPG - Carboxyhemoglobinemia - Structural variants Low/ decreased oxygen affinity states: Increased RBC 2,3 BPG Structural variants Methemoglobinemia Congenital methemoglobinemia Structural variants Cytochrome b5 reductase deficiency 2. Acquired (toxic) methemoglobinemia E. Posttranslational modifications Nonenzymatic glycosylation Amino-terminal acetylation Deamination Amino-terminal carbamylation

Table 5: Different Drugs and Chemical Capable of Inducing Methemoglobinemia

Drugs capable of Inducing Methemoglobinemia		Chemical Agents Capable of Inducing Methemoglobinemia	
Acetaminophen	Phenacetins	Acetanilide	Automobile exhaust fumes
p-Aminosalicylic acid	Oral hypoglycemics	Alloxan	Burning wood and plastic
local anesthetics	Metoclopromide	Anilines	Nitrates
Benzocaine	Nitrates	Aminophenol	Potassium nitrate
Bupivacaine	Silver nitrate	Benzene	Sodium nitrate
Lidocaine	Nitroglycerine	Bivalent copper	Nitrites
Prilocaine	Nitroprusside	Chlorates	Naphthalene
EMLA	Nitrites	Chromates	Nitrophenol
Anticonvulsants	Nitrofurantoin	Dimethyl sulfoxide	Nitrobenzene
Valproic acid	Nitric oxide	Dinitrophenol	Toluidine
Phenytoin	Nitrous oxide	Phenol	
Antimalarial drugs	Piperazine	Fumes	
Chloroquine	Rifampin		
Primaquine	Sulfonamides		
Quinacrine	Sulfasalazine		
Methylene blue	Sulfamethoxazole		
Dapsone	Sulfadiazine		

Table 6: Percentage of Methemoglobin level with Associated Signs and Symptoms

Fractional Methemoglobin level (%)	Signs and symptoms
<3 (normal)	None
3-15	Frequently none Grayish skin
15-30	Cyanosis Chocolate-brown blood
30-50	Dyspnea Headache Fatigue, weakness Diziness, syncope SpO_2 ~85%
50-70	Tachypnea Metabolic acidosis Cardiac arrythmias Seizures CNS depression Coma
>70	Death

REFERENCES

1. Clinical Gate. (2018). Cyanosis. [online] Available at: https://clinicalgate.com/cyanosis-2/ [Accessed 2 Mar. 2018].
2. Uptodate.com. (2018). UpToDate. [online] Available at: https://www.uptodate.com/contents/etiology-and-evaluation-of-cyanosis-in-children [Accessed 2 Mar. 2018].
3. Hiremath, G. and Kamat, D. (2018). Diagnostic Considerations in Infants and Children with Cyanosis.
4. Uptodate.com. (2018). UpToDate. [online] Available at: https://www.uptodate.com/contents/clinical-features-diagnosis-and-treatment-of-methemoglobinemia [Accessed 2 Mar. 2018].
5. Da-Silva, S., Sajan, I. and Underwood, J. (2003). Congenital Methemoglobinemia: A Rare Cause of Cyanosis in the Newborn--A Case Report. PEDIATRICS, 112(2), pp.e158-e161.

6. Turner-white.com. (2018). Cite a Website - Cite This For Me. [online] Available at: http://www.turner-white.com/pdf/brm_Hem_V5P3.pdf [Accessed 2 Mar. 2018].

7. Forget, B. and Bunn, H. (2018). Classification of the Disorders of Hemoglobin.

8. Marengo-Rowe, A. (2018). The Thalassemias and Related Disorders.Sarnaik, S. (2005). Thalassemia and related hemoglobinopathies. The Indian Journal of Pediatrics, 72(4), pp.319-324.

9. Rehman, H. U. (2001, September). Methemoglobinemia. Retrieved October 07, 2017, from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC1071541/

10. Methemoglobinemia: a systematic review of the ... (n.d.). Retrieved October 7, 2017, from https://www.bing.com/cr?IG=889C181E05E64CF89C8197A3E4FFB537&CID=07B90B5DED1E60CB2BE70049EC1861C4&rd=1&h=0Dy2J6axGfbRK0jx6ZH1nVLjuGJl_rP6CkSP3ch3PEg&v=1&r=https%3a%2f%2fwww.researchgate.net%2fpublication%2f51677330_Methemoglobinemia_a_systematic_review_of_the_pathophysiology_detection_and_treatment&p=DevEx,5063.1

11. Nascimento, T., Pereira, R., Mello, H. and Costa, J. (2018). Metemoglobinemia: do diagnóstico ao tratamento.Percy, M. J., Mcferran, N. V., & Lappin, T. R. (2005). Disorders of oxidised haemoglobin. *Blood Reviews, 19*(2), 61-68. doi:10.1016/j.blre.2004.02.001 [11].

12. Percy M, Lappin T. Recessive congenital methaemoglobinaemia: cytochrome b5reductase deficiency. 2018.

13. Percy MJ, Lappin TR. Recessive congenital methaemoglobinaemia: cytochrome b5 reductase deficiency [Internet]. British Journal of Haematology. Blackwell Publishing Ltd; 2008 [cited 2018Mar2]. Available from: http://onlinelibrary.wiley.com/doi/10.1111/j.1365-2141.2008.07017.x/full

14. Da-Silva S, Sajan I, Underwood J. Congenital Methemoglobinemia: A Rare Cause of Cyanosis in the Newborn—A Case Report [Internet]. Pediatrics. 2018 [cited 2 March 2018]. Available from: http://www.bing.com/cr?IG=C8D771609A8C44068DA4C1C0A7D2A78A&CID=1D5AE638EE4D6F860DBAED2CEF4B6E5A&rd=1&h=H77VhtAHFetZX2xkVRTG6g-lHkzmubkEDCUypOtM6Lk&v=1&r=http%3a%2f%2fpediatrics.aappublications.org%2fcontent%2f112%2f2%2fe158&p=DevEx,5068.1

15. Nelson Textbook of Pediatrics, 2-Volume Set, 20th Edition. (n.d.). Retrieved October 07, 2017, from https://elsevier.ca/product.jsp?isbn=9781455775668

6. Ascorbic acid for the treatment of methemoglobinemia: the experience of a large tertiary care pediatric hospital. [Internet]. Medscape.com. 2018 [cited 2 March 2018]. Available from: http://www.medscape.com/medline/abstract/24914501

7. Percy MJ, Lappin TR. Recessive congenital methaemoglobinaemia: cytochrome b5 reductase deficiency [Internet]. British Journal of Haematology. Blackwell Publishing Ltd; 2008 [cited 2018Mar2]. Available from: http://onlinelibrary.wiley.com/doi/10.1111/j.1365-2141.2008.07017.x/full Ulph, F. (2013). PINSA (Provision of Information about Newborn Screening Antenatally): The Provision of antenatal Information for the NHS Newborn Bloodspot Screening Programme (NBSP). Http://isrctn.org/>. doi:10.1186/isrctn70227207

18. Mansouri A, Lurie A. Methemoglobinemia. 2018.

19. Umbreit J. Methemoglobin—It's not just blue: A concise review. 2018.

20. Ulph, F. (2013). PINSA (Provision of Information about Newborn Screening Antenatally): The Provision of antenatal Information for the NHS Newborn Bloodspot Screening Programme (NBSP). Http://isrctn.org/>. doi:10.1186/isrctn70227207

21. Philippine Society for Orphan Disorders [Internet]. NORD (National Organization for Rare Disorders). [cited 2018Mar2]. Available from: https://rarediseases.org/organizations/philippine-society-for-orphan-disorders/