International Journal of Agriculture Sciences
ISSN: 0975-3710 & E-ISSN: 0975-9107, Volume 15, Issue 2, 2023, pp.-12211-12216.
Available online at https://bioinfopublication.org/pages/jouarchive.php?id=BPJ0000217

Review Article

ANEMIA IN ANIMALS- AN OVERVIEW

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Received: February 02, 2023; Revised: February 25, 2023; Accepted: February 26, 2023; Published: February 28, 2023

Abstract: Anemia is most common and frequently diagnosed complication caused due to many etiological factors, some of which are life-threatening in veterinary practice. Anemia could be a disease by itself or could be consequential to several other factors. It is a syndrome affecting all animals especially with neoplasia and other clinical conditions like Bacterial, Viral, Parasitic & Fungal infections, auto- immune diseases, deficiency diseases, toxicities, etc. Clinical manifestations of anemia depend on the severity, duration (short or long-term), and the underlying cause of the illness. Classification of anemia is based on various factors. Assessment and diagnosis of anemia can be made on the basis of history, clinical manifestations, laboratory findings and bone marrow examination. Therapeutic management of anemia is usually based on the etiology behind it. Considering the importance and the high prevalence of anemia in animals it has been reviewed here.

Keywords: *Anemia, Classification of anemia, Etiological factors, Thrombocytopenia*

Citation: Moregaonkar S.D., *et al*., (2023) Anemia in Animals- An Overview. International Journal of Agriculture Sciences, ISSN: 0975-3710 & E-ISSN: 0975-9107, Volume 15, Issue 2, pp.- 12211-12216.

Academic Editor / Reviewer: Dr Gowher Gull Sheikh, Dr A K Shrivastava

Introduction

Anemia is defined as a condition in which the body has a decreased number of circulating erythrocytes (RBCs), or decrease in hemoglobin concentration. It can also be defined as reduction in number of the erythrocytes in an animal for that particular age, species, breed, and geographic location. It can either be due to decreased production of erythrocytes or haemoglobin or increased destruction of red blood cells (RBCs). It is therefore associated with reduction in red cell count or volume of red cell. Anemia is suggested clinically by pale or white mucous membranes, exercise intolerance, tachycardia, weakness, tachypnoea, possible systolic murmurs and collapse (Smith and Sherman, 2009). Clinical manifestations of anemia depend on the severity, the duration (short or long-term), and the underlying cause of the illness. Anemias due to bleeding or the destruction of existing red blood cells are usually regenerative. Anemias that are caused by a decrease in the hormone that stimulates red blood cell production or by an abnormality in the bone marrow are nonregenerative. The causes of nonregenerative anemias are mainly nutritional deficiencies, chronic & inflammatory diseases, kidney diseases, or bone marrow diseases.

The major causes of anemia were hepatic & renal dysfunction, infectious & systemic diseases and nutritional deficiencies. The course and outcome of anemia is mainly dependent upon the degree and severity affecting the blood components and it could be life threatening or at other side it may be asymptomatic and even difficult to detect clinically. Therefore, one should give more attention towards predisposing causes, exciting causes, diagnosis and prevention of anemia rather than treating it.

Anemia in Animals

The term anemia come from Greek word, An-without, emia-blood. Later in early 19th century anemia word comes from modern Latin. Anemia is the qualitative (Hb concentration) or quantitative (No. of circulating erythrocytes) decrease of the erythrocytes in the circulation, for an animal of a particular age, species, breed and geographic location. Anemia is a reduction in number of the erythrocytes in an animal for that particular age, species, breed, and geographic location [1]. Runnells *et al*. (1976) [2] stated that the anemia as a quantitative or qualitative decrease in the amount of blood in an individual and anemia may be local or general. Jones and Hunt (1983) [31] described anemia as a reduction below normal of the number of erythrocytes and/ or hemoglobin concentration per unit volume of blood. Brar (2002) [3] stated that on CBC examination decrease in hemoglobin value, packed cell volume and erythrocytes per microliter indicates anemia. Radostits *et.al*., (2007) [4] defined anemia as deficiency of circulating RBCs per unit volume of blood. Anemia is defined physiologically as a diminished capacity of the blood to provide tissues with enough oxygen for healthy metabolic processes [5]. Local anemia is called as "ischemia". It is an absolute oligocythemia, which could occur due to two factors, a) Production of erythrocytes is low, but destruction is normal *i.e*., Dys-haemopoietic anemias. b) Production of erythrocytes is normal but the destruction is excessive *i.e*., Hemolytic or hemorrhagic anemias. Anemia is a "syndrome" which can be caused by many etiological factors.

Classification of Anemias

Different authors have classified anemia differently. However, considering all, we have formulated the best way of classifying anemia as under; Anemia can be classified as-

1. Morphological classification of anemia

- Macrocytic normochromic anemia
- Macrocytic hypochromic anemia
- Normocytic normochromic anemia
- Normocytic Hypochromic anemia
- Microcytic normochromic anemia
- Microcytic hypochromic anemia

2. Classification of anemia based on etiology

- Blood loss anemia- Extravascular (hemorrhagic) and Intravascular (hemolytic)
- Dys-haemopoietic anemia (Nutritional deficiency anemias)
- Hereditary anemia: Generically determined defects in erythropoiesis

3. Classification of anemia according to a bone marrow response
Basically, it is further classified as, i. Regenerative ii. Non-regenerative
Other types are, i. Toxic inhibition, ii. Aplastic anemia, iii. Myelopthistic anemia

4. Broadly an anemia can be classified on the basis of A) Bone marrow response B) Size of RBC and Hemoglobin concentration and C) On etiology. Further, in anemia, haematological and biochemical alterations provide important indices for its diagnosis. They classified anemia in four categories as, Blood loss anemias- due to surgery, trauma, defects in clotting mechanisms, viral infections, rupture of vascular malignant tumour etc. Hemolytic anemias-Per-acute or chronic blood loss. Nutritional deficiency anemias-most commonly due to, mineral deficiency including iron, Cu, Co, due to Vit B_{12}, Niacin deficiency etc. Anemia due to bone marrow depression.

5. Moreno Chulilla (2009) [29] classified anemia in to, On the basis of Pathogenesis- Hypo-regenerative and regenerative types. Red Cell Morphology- Microcytic, Normocytic or Macrocytic. Clinical Presentation- Acute or Chronic

6. Sanap *et al*., 2011 classified human anemias in to, ACD- Anemia of Chronic Diseases or cases, BFD- Vit B_{12} or Folic acid Deficiency anemia, ARD- Anemia of Renal Diseases or Disorder, APA- Aplastic Anemia, IDA- Iron Deficiency anemia, THAL- Thalassemia, CD or SSD- Combined Deficiency or Sickle Cell Anemia, Severity of anemia -i) Severe, ii) Moderate, iii) Mild types

I. Morphological Classification of Anemia
1. Macrocytic and normochromic anemias
This type of anemia which denotes the presence of immature red cells in the circulation (macrocyte). The size of erythrocyte is bigger than normal (macrocyte) with usual color. *Causes*: Results from deficiency of Vit. B_{12}. Deficiency of Erythrocyte Maturation Factor (EMF). Deficiency Cobalt, folic acids and niacin. In gastric disorders: which reduces the absorption of EMF and Vit. B_{12}. In Hepatic insufficiency: in this condition EMF is not stored in the liver. *Consequences*: The deficiency of above factors results in to depressed synthesis of the RNA and DNA and causes arrest of maturation of prorubricytes and metarubricytes. Depressed DNA synthesis which causes delayed maturation of nucleus, but hemoglobin concentration reaches to normal and this type of the anemia results.

2. Macrocytic and Hypochromic anemia
In this erythrocyte are bigger in size with low hemoglobin concentration. It is seen in the regenerative phases after hemorrhages. *Causes*: The hemorrhages may be due to- Trauma, Surgical bleeding, Parasites, Epistaxis.

3. Normocytic Normochromic anemia
These anemias are the most frequently encountered anemias in animals. They result from depression of erythropoiesis and are therefore often referred to as aplastic anemias or hypoplastic anemia. The size of the RBC is normal and hemoglobin concentration is also normal. *Causes*: Neoplastic diseases, irradiation & certain toxicities produce this type of anemia. Acute or sub-acute systemic diseases. Suppression of the bone marrow activity. Chronic hemorrhages. Neoplastic conditions. Deficiency of Vit.B6 and prothrombin. Ionization, Irradiation, chemical poisoning, bracken fern poisoning, trichloroethylene extracted soya bean poisoning etc. Sulphonamide and chloramphenicol toxicity.

4. Normocytic and hypochromic anemia
In this type of anemias there is reduced concentration of the hemoglobin. *Causes*: Dietary deficiency of iron, copper, ascorbic acid, pyridoxine, nicotinic acid, riboflavin, thyroxin.

5. Microcytic and normochromic anemia
In this type the RBCs are smaller than the normal size with normal staining properties. *Causes*: Chemical poisoning e.g., mustard nitrogen, antibiotic toxicity. Chronic interstitial nephritis- due to low production of erythropoietin. Worm infection *e.g., Hemonchus, oesophagostomium*. Chronic infection *e.g.*, Tuberculosis, brucellosis. Ionizing radiations.

6. Microcytic and hypochromic anemias
In this anemia, RBC is smaller in size than normal and also having low concentration of hemoglobin. These are classical iron deficiency anemias or "Tired blood". *Causes*: Dietary deficiency of iron, copper, manganese, cobalt, ascorbic acid, pyridoxine, nicotinic acid, riboflavin and thyroxin. Anemia due to chronic blood loss. Nutritional deficiency of copper and pyridoxine also produce microcytic hypochromic anemia. Copper is necessary for the utilization of iron in the production of hemoglobin.

II. Classification of Anemia Based on Etiology
1. Blood Loss Anemia
A. Extravascular (Hemorrhagic) Anemia
Normally, there is a balance between blood production and blood loss. But, in this condition blood loss is greater than production. Hemorrhagic anemia results from severe hemorrhages. In this condition there is extravascular destruction of erythrocytes. *Factors*: The hemorrhagic anemias are classified depending on the following factors: The amount of blood loss. The rate of blood loss. The diet controlling the balance between blood loss and production *i.e*., sufficient quantity of iron.
Blood picture: The blood picture in this type of anemia, initially is Normochromic Normocytic (when the blood production and blood loss is balanced), then it turns to macrocytic normochromic (when bone marrow working at faster rate) and lastly it is microcytic hypochromic. Types: The various types of hemorrhagic anemias are:
1) Acute hemorrhagic anemia:
Etiology: *Severe injury*- Sweet clover poisoning ingestion of moldy sweet clover hay or silage- dicoumarol. -It inhibits the conversion of thrombin into prothrombin and inhibits the clotting mechanism.
Warfarin poisoning- Braken fern poisoning: In this condition there is an acute thrombocytopenia, which is the direct cause of the hemorrhages. It produces the thiamine deficiency, (bracken fern contains the thiaminase). It is seen in ulceration in stomach in pig, bleeding abomasal ulcers in cattle, bovine enzootic hematuria, in Coccidiosis in poultry and Haemonchosis & Ancylostomiasis- in all

2) Chronic hemorrhagic anemia
Etiology: Blood sucking worms: In Cattle and sheep: Hemonchus, Fasciola, Bunostomum. In Horses: strongyles, In Dog: Ancylostomas, Ectoparasites: ticks, lice and flies, Protozoa: coccidiosis in dog, Hemorrhagic diseases: chronic bovine hematuria, Gastrointestinal ulcers and vascular tumors

3) Purpura and hemorrhagic diseases
Purpura is an accumulation of blood under the skin due to spontaneous rupture of capillaries. Hemorrhages results even due to mild damage.to capillary endothelium. *Causes*: Purpura is a syndrome and the causes may be broadly classified as under,
a) Vascular disorders: *Purpuric infections (Bacterial and viral)*: symptomatic purpura is found in various diseases characterized by petechial hemorrhages. E.g., H. S., Anthrax, etc. in these bacterial toxins causes injury to blood vessels. While in case of viral diseases, e.g., ICH and hog cholera, the virus multiplies in the endothelial cells and cause damage to the endothelium. Allergic purpura, also known as purpura hemorrhagica, can result in strangulations, fistulous withers, poll-evil, and guttural pouch emphysema after an infectious toxaemia. Edema of subcutis, petechial hemorrhages, etc. are seen. The defect is seen in the vascular endothelium due to development of the allergy, resulting in increased capillary permeability. *Congenital purpura*: It is seen in fetus. Iso-agglutinins formed against platelets in the mother pass into the fetus via placenta and produce thrombocytopenia. *Senile purpura*: Generally, not seen in animals, but can be rarely seen in undernourished animals. The vessels are get easily damaged as there is no subcutaneous fat and the skin is very much atrophied. *Vitamin C deficiency*: Due to deficiency of vit. C there is increased capillary permeability and capillary fragility since, cement substance of capillary wall is not synthesized.

But this condition is rare in animals. Impaired absorption and deficiency of Vit. K.
b) Impaired clotting mechanism: Thrombocytopenia**:** *1) Idiopathic or primary* thrombocytopenia; probably due to the auto antibodies against platelets are present. *2) Secondary thrombocytopenia*: Damage to bone marrow: by chemicals like nitrogen mustard, benzol; individual sensitivity to drugs like oxytetracycline, sulphonamides, streptomycin; Animal toxins like zootoxins, snake venom; physical agents like radiations, heat stroke; or septic diseases. Myelopthistic replacements *e.g.*, leukemia (Myelo- BM, Pthisia- wasting), Hypersplenism, Aplastic anemia, Bracken fern poisoning.

Other coagulating defects:
Hemophilia: It is a condition in which coagulation of blood does not occur after injury and it ends in fatally. It is inherited defect. Prothrombin deficiency causes of prothrombin deficiency are; *Liver diseases*: in liver diseases, fibrinogen, factor V, prothrombin, factor VII & factor IX are not synthesized, which are required for coagulation of blood. *Impaired absorption and deficiency of vitamin k*: Diseases of liver and intestines interferes in the absorption of vitamin k. Bile is required for the absorption of the vitamin k, so in hepatitis the deficiency of vitamin k occurs.
Poisoning by di-coumarin, warfarin and sweet clover poisoning: The di-coumarin is depresses the activity of vitamin k and so depresses the formation of the prothrombin, factor VII, factor IX and factor X. The cattle and sheep suffer mostly, sometimes it is seen in pets and swine. *Presence of circulating anticoagulants*: In anaphylactic shock in dogs, large amount of heparin is liberated resulted in bleeding. Heparin prevents the conversion of prothrombin to thrombin. Some snake venom possesses the coagulants so bites of such snakes may result in fatal bleeding. *Unknown etiology*: *Moldy corn poisoning*: In cattle and swine the moldy corn produces hemorrhages in various parts of the body. Together with necrosis of hepatic parenchyma and renal epithelium. Abortion in pregnant cows. Lesions include centrilobular necrosis, cloudy swelling and fatty degeneration of the renal tubular epithelium with glomerular atrophy and necrosis of some tubular epithelium. *Epistaxis in horses*: In some families of horse epistaxis occurs due to strenuous exercise. This is due to non-sex-linked character, the walls of blood vessels are very thin and so rupture whenever distended during great exertion (racing).

B. Intravascular (Hemolytic) Anemia
In this condition intravascular destruction of erythrocyte occurs. Anemia is normochromic and macrocytic becoming hypochromic and microcytic as the iron stores are used up. In this condition hemoglobin is break up into heme and globin. The iron of the heme is stored by the RE cells for the future use. The pigment part is excreted as chole-bilirubin and urobilinogen. The protein moiety is broken down into amino acids in liver which are again used for the synthesis of the hemoglobin again. In some hemolytic anemias there is faster rate of breakdown of Hb, so it results into; *Jaundice*: With increased bile pigments in the blood, feces and urine
Hemosiderosis: Increased storage of iron in the form of hemosiderin crystals, Hemoglobinuria. *Causes*: Causes of HA can be categorized in to, a. Infectious Diseases, b. Poisoning/ toxicities, c. Heredity, d. Immunological, e. Others, Further causes vary with the species of animal.

2. Infectious Diseases:
Inflammatory diseases are reported to be a common cause of anemia. Protozoal infections: e.g., babesiosis, theileriosis, Nagana (a disease of cattle, antelope, and other livestock in southern Africa, characterized by fever, lethargy, oedema, & caused by trypanosome parasites transmitted by the tsetse fly), trypanosomiasis, anaplasmosis and eperythrozoonosis. *Bacterial infections*: *e.g.*, leptospirosis, *Bacillary hemoglobinuria, staphylococci and streptococci*, *Clostridium hemolyticum*, Ehrlichiosis (*Haemobartonella canis*) In dog. Viral infection: Equine Infections Anemia, Chicken infectious anemia, feline leukemia virus (FeLV) and feline immunodeficiency virus (FIV). *Poisonings*: By potassium & sodium chlorates-in cattle. *Onion poisoning*: The toxic principle of the onion is the n-propyl disulphide. The carcass smell of onions, hemolytic anemia with hemoglobinuria and icterus. *Poisoning by castor seeds*: Ricin in castor seeds produces haemolysis and so ingestion of large quantities of castor seeds results in hemolytic anemia. Poisoning by rape, kale plants: etc.: It usually occurs due to accidental ingestion of these plants in animals. One particularly important toxic substance present in rape and kale crops is a haemolytic anemia factor, S-methylcysteine sulfoxide (SMCO). SMCO is converted by bacterial fermentation in the rumen to dimethyl disulfide, which causes haemolysis. Chronic copper poisoning can occur. But usually seen in sheep. Lead also produces acute hemolytic anemia due to severe red cell destruction. Lead inhibits a variety of red cell enzymes, including several enzymes of porphyrin metabolism and pyrimidine-5'-nucleotidase. Arsenic may cause hemolysis by interacting with sulfhydryl groups, severe red cell destruction, and hemolytic anemia. Many red cell enzymes are inhibited by copper, and it also speeds up the oxidation of intracellular glutathione (GSH). *Phenothiazine poisoning (Drug sensitivity)*: In light doses, it causes the hemolysis. Hemolytic anemia produced by phenazopyridine is often associated with “bite cells” and “blister cells.” Hemolytic anaemia can also be brought on by animal poisons, such as those produced by insects, spiders, and snakes. Snake bite also causes the hemolytic anemia. The snake venom contains a leocithinase, which act on lecithin & converts into lyolecithin (in vitro), which is highly hemolytic.

3. Immunoogical Causes: *Autoimmune hemolytic anemia*: antibody is made against one's own RBC's (Icterus neonatorum (In foals). ST. in calves and piglets. Similar to Erythroblastosis fetalis (In human). Maternal IGG crosses the placenta and attaches to fetal RBC's (In calves). Abnormal auto-antibodies in malignant tumors, collagen diseases cause autoimmune hemolytic anemia- spontaneous agglutination of erythrocytes. Incompatible blood transfusion, in this presence of hemolysins in the plasma - causes the hemocysis. Injection of blood products (Rare in animals). Immune-mediated conditions are the most frequent causes of hemolytic anaemia in dogs (IMHA). Red blood cells are destroyed when the body no longer recognises them as its own and produces antibodies against circulating red blood cells. *Hereditary*: Genetically determined defects in erythropoiesis. This type of anemia is uncommon in animals. *Autoimmune hemolytic anemia*: antibody is made against one’s own RBCs. *Erythroblastosis fetalis*: maternal IgG crosses the placenta and attaches to fetal RBCs
Other Causes: Hypersensitivity of certain drugs: sulphanilamide, quinine. para-amino-salicylic acid & certain anti-pyretic drugs may cause hemolytic anemia in cattle. Phenothiazine (in horses). Treatment with long acting oxytetracycline. Napthalene moth balls when ingested accidentally by the pet dogs. *Water intoxication*: Excessive drinking of the cold water when the calves (rarely in older cattle). *Heat*: It has been known for over a hundred years that heating blood to temperatures above 47°C (117°F) rapidly produces visible damage to erythrocytes. These observations explain the severe hemolytic anemia which occurs in patients with extensive burns.

Immune Mediated Haemolytic Anemia
In dogs, the most common cause of hemolytic anemia is immune mediated. Anemia of this kind can develop spontaneously or as a result of tumors, infections, medications, or immunizations. Red blood cells are destroyed when the body no longer recognizes them as its own and produces antibodies against circulating red blood cells. Immune-mediated hemolytic anemia in dogs typically causes jaundice, fever, and maybe an enlarged spleen. They may have slow-moving, mild symptoms with no outward indicators of pain or they may suddenly experience acute crisis. According to the symptoms, your veterinarian will customize the treatment. The use of needless medication therapy will be stopped and any underlying infections will be treated. When necessary, blood transfusions can be added to intravenous fluid therapy. In order to limit the oxidation of red blood cells, immune system-suppressing medications are also administered.
Canines with immune-mediated hemolytic anemia run the danger of having a blood clot fragment that has broken free from a blood clot block a blood vessel in their lungs (a condition known as pulmonary thromboembolism). Although there is no known underlying cause, supportive therapy with fluids and blood transfusions may lower the risk. Fluids are crucial to maintain healthy kidneys and to shield them from the high levels of circulating bilirubin (the reddish yellow pigments in blood that cause jaundice).

Anticoagulant medicine may also be used if there is a high risk of blood clot formation. A dangerous medical disorder known as immune-mediated hemolytic anemia results in mortality in 20% to 75% of affected animals. An increased risk of death may be indicated by fast reductions in red blood cell counts, moderate to high white blood cell counts, irregular bruising, and excessive clotting. One of the most prevalent immune-mediated illnesses in dogs is idiopathic immune-mediated hemolytic anaemia (IMHA) [6,7]. In the 1960s, the first case series of 19 dogs with IMHA was described. Five of the 19 dogs perished following recurrences and another six during the initial haemolytic event [8]. The mortality of IMHA is still high despite various studies conducted since then [9-14].

Etiology: Breed and sex predispositions: Breed predisposition and familial occurrence suggest that a genetic component contributes to the susceptibility for IMHA. The recognition of foreign proteins or self-proteins by the major histocompatibility complex (MHC) proteins is one of the key events in the development of immune-mediated disease [15]. Canine IMHA is associated with both susceptible and protective dog leucocyte antigen (DLA) haplotypes which, interestingly, are associated with different effects in specific breeds [30]. The presence of autoreactive T-cells in dogs with IMHA supports the view that MHC molecules are candidate susceptibility genes for IMHA ([16]. The distribution and frequency of both DLA alleles and haplotypes vary substantially between breeds as a result of selective breeding [17-18]. An increased incidence of IMHA has been observed in female [19] and neutered dogs. *Clinical Signs:* IMHA can occur at any age. Although most reports describe an onset after the first year [20-21]. The majority of dogs with IMHA develop anaemia quickly, often in as little as three days. Most dogs experience non-specific symptoms including lethargy and appetite loss, which in 15-30% of instances are also accompanied by vomiting and diarrhoea. Red urine and yellow to orange discoloration of the faeces are more specific symptoms of hemolysis. The physical examination reveals clinical signs caused by anemia, such as, tachycardia, tachypnoea, steep pulse, pale mucous membranes, and systolic murmur. Fever is a common clinical sign [22]. Petechiation as a result of concurrent severe thrombocytopenia is reported incidentally (2-5% of cases) and may be due to concurrent immune-mediated thrombocytopenia (ITP). Cranial abdominal organomegaly, due to splenomegaly and hepatomegaly, is found in up to 40% of cases.

Complete blood count

At the time of presentation, most dogs have severe anemia with a haematocrit of 12-14%, but some dogs may show a more chronic disease course and have a higher haematocrit [23]. Tissue oxygenation is severely impaired at a haematocrit below 10% [24] and can result in severe exercise intolerance, tachypnoea, and tachycardia. Pronounced leucocytosis with a left shift is a common laboratory feature at presentation, or leucocytosis may develop in dogs with leukopenia or normoleukaemia at presentation. Monocytosis is present in about 50% of cases [25]. *Diagnosis: Diagnostic testing*: The laboratory diagnosis of IMHA rests upon the demonstration of an immune-mediated mechanism for the haemolysis. The direct Coombs' test is still the main method used to demonstrate anti-erythrocyte antibodies, alternative methods, however, include flow cytometry [26]. Spherocytosis can be the result of immune-mediated erythrophagocytosis. Spherocytosis is generally accepted as pathognomonic for IMHA. Autoagglutination is generally accepted as a diagnostic criterion for IMHA [27,28].

Mortality: The death rate of canine IMHA may be as high as 80%, and most deaths occur in the first 2 weeks after diagnosis. As many as half of these deaths are due to thromboembolism in the lung, liver, spleen, or in multiple organs, which was found in 50-80% of cases.

Risk factors for mortality: The most important being variables related to DIC, inflammation, and liver & kidney failure. Tissue hypoxia due to the often-severe anemia that develops during IMHA has been suggested to play a central role in the development of pathology.Multivariate models suggest that liver and renal failure, DIC, and inflammation contribute independently to the risk of death.

Dyshemopoitic Anemia (defect in the formation of erythrocytes): These are the nutritional deficiency anemias. The anemias occurred due to decreased production of erythrocytes or hemoglobin are grouped in this category.

The defect may lie in the formation of stroma protein or in the formation of hemoglobin. The porphyrins are the substances required for the synthesis of heme. The defective synthesis of the heme gives rise to excessive number of porphyrins and leads to the porphyrinuria and porphyria. Congenital porphyria occurs in the pigs and bovines. The pigment is photosensitive, when deposited in tooth gives rise to condition "pink tooth". In bones causes the "osteohemochromatosis". In porphyrinuria it imparts the red color to urine when exposed to light. The affected animals may suffer from photodynamic photosensitization. *Causes*: The following are the etiological factors responsible for the causing the dys-hemopoitic anemia. Diminished Stroma Protein Formation: The blood picture is usually macrocytic and normochromic. The bone marrow is megaloblastic showing numerous megaloblasts and giant metamyelocytes.

***a) Dietic deficiency of extrinsic factors*:**

i. Deficiency of cobalt and vitamin B_{12}: In ruminants the vitamin B_{12} is synthesized by the ruminal microflora, with the help of cobalt. So, deficiency of cobalt leads to decreased production of vitamin B_{12} which is required for synthesis of RNA and DNA. So, deficiency of vitamin B_{12} causes arrest of maturation of prorubricytes and metamyelocytes. So, depressed DNA synthesis leads to delayed nuclear maturation, but hemoglobin synthesis is not affected. When hemoglobin synthesis reaches certain concentration in the erythrocytes, the nucleus leaves them and so macrocytes results giving rise to macrocytic anemia.

ii. Deficiency of folic acid: folic acid is also required for the maturation of prorubricytes and metamyelocytes. So, deficiency also leads to delayed maturation of the erythroblasts and it leads to the macrocytic anemia.

b) Deficiency of intrinsic factor: The gastric mucosa contains certain enzymes which helps in the absorption of vitamin B_{12}. But, in gastric diseases the enzyme synthesis is impaired ultimately leading to the less absorption of vitamin B_{12} and in turns it leads to anemia.

c) Failure to store the erythrocyte maturation factor (Vit.B_{12}): The vitamin B_{12} is also known as Erythrocyte Maturation Factor (EMF) and is stored in the liver. In hepatic diseases the storage of vitamin B_{12} does not occur and leads to the anemia.

d) Failure to use the EMF: The failure of mobilization the EMF from liver and failure to utilization of EMF also give rise to the macrocytic anemia. It is also called as the achrestic anemia.

e) Hypopituitarism: The hypopituitarism leads to defective metabolism of the carbohydrate, so there is less production of the energy, so lack of energy reduces the rate of erythropoiesis which in turn leads to anemia.

Diminished Hemoglobin Formation

The blood picture may be normocytic hypochromic or microcytic hypochromic, depending upon stage of anemia. *Deficiency of iron*: Iron is required for the synthesis of heme. The milk of pigs is deficient of the iron, so piglets suffer from anemia. The ingestion of phosphorous and phytic acid with iron, from insoluble complexes of iron which are excreted through the faeces and leads to iron deficiency and further which leads to the anemia. In growing animals, in pregnancy and in race horses there is increase demand of oxygen and which leads to the iron deficiency. *Deficiency of copper*: Copper acts as a catalyst in the utilization of iron in hemoglobin formation. The microcytic anemia results in the iron deficiency. *Deficiency of pyridoxine*: Pyridoxine is required for the utilization of iron during heme synthesis. *Deficiency of ascorbic acid (vitamin C)*: Vitamin C is required for the reduction of Fe+++ (ferric ion) to Fe++ (ferrous ion) state which is easily absorbed. The ascorbic acid also required for the synthesis of folic acid.

Deficiency of nicotinic acid: Nicotinic acid is concerned in synthesis of pyridine nucleotide which takes part in cell respiration. So, deficiency of nicotinic acid interferes with the respiration of immature red cells, this is seen in dogs and pigs.

Deficiency of riboflavin: Riboflavin is concerned with the metabolism and arrangement of the amino acids in the globin molecule, so deficiency leads to the impaired hemoglobin, seen in dogs. *Deficiency of thyroxin*: The thyroxin is required for the metabolism of fats and carbohydrates. Impaired thyroxin production results in the decreased metabolism of the CHO and fat, so leads to decrease production of the energy, so decrease in synthesis of hemoglobin rate. In this normocytic or macrocytic anemia is seen. This condition seen in myxedema, hypopituitarism, etc.

4. Classification of Anemia by Bone Marrow Response:

Role of Erythropoietin: For erythropoiesis, a humoral substance, the erythropoietin, appears to be of great importance. The granular cells of the juxtaglomerular apparatus appear to be important source of erythropoietin, which is probably a glycoprotein. It is present in plasma, urine and milk. Erythropoietin stimulates the differentiation of the bone marrow stem cells to rubriblast. It governs the rate of hemoglobin synthesis.

Synthesis and regulation: Its secretion is controlled by oxygen content of renal arterial blood. Hypoxia is stimulus for erythropoietin secretion. This is reason for the polycythemia found in high altitude disease. Anemia of chronic renal disease (CKD) may mostly due to decreased erythropoietin production by the damaged kidneys. Androgens, adrenal corticoids, thyroxine and growth hormone stimulate erythropoiesis. The first three probably act directly by stimulating erythropoietin production, while the last has a direct effect on marrow, stimulating erythropoiesis. Estrogens depress erythropoiesis, probably by competing with erythropoietin production or by competing with it in it action on stem cells.

Regenerative Anemia: It is also called as responsive anemia. Regenerative anemia can be caused by a hemorrhage or hemolysis (the rupture or destruction of blood cells). Bone marrow responding and there is presence of good number of reticulocytes in circulation. Findings that denote regeneration are polychromasia, reticulocytosis and hypercellular bone marrow.

Non- Regenerative Anemia: Bone marrow do not respond to anaemic state and reticulocytes are absent in circulation. Bone marrow examination is indicated; it may reveal the pathophysiologic mechanism. Polychromasia and reticulocytosis are absent.

Toxic Inhibition of BM: The blood picture in this type of the anemia is microcytic normochromic. This is non regenerative type of anemia. The marrow is normal and active but unable to utilize the hematinic so regenerative form does not occur. *Causes*: Chemical poisons: *e.g*., Nitrogen mustard (cytotoxic), folic acid metabolites, streptomycin, sulphonamides, chloromycetin, bismuth, gold, arsenic, benzyl, hair dyes, insecticides. *Chronic interstitial nephritis*: In this condition, there is uremia which suppresses the production of the erythropoietin in kidneys and also the erythropoietic cells. *Oesophagostomiasis*: This worm causes the pimply gut condition in intestine and depresses the absorption, in turn leads to different deficiencies resulting in anemia. *Chronic infections*: In chronic infections like TB, brucellosis and rheumatic fever, there is hypoferremia with reduction in serum iron binding capacity. Along with this hypocupremia also seen. In these conditions there appears to be some abnormalities in hemoglobin synthesis for there is increased excretion of coproporphyrins. *Ionizing radiations*: Ionizing radiations are injurious to the hemopoietic tissues. After exposure to radiations the spleen and lymphoid tissue becoming soft and shrunken. After exposure there is decrease in the granulocytes number very rapidly. Hemorrhages occur due to thrombocytopenia and damage to the vascular endothelium.

Aplastic Anemia: Aplasia term came from English and Greek word, aplasia. In English a- without and In Greek, plasia- formation *i.e*., without formation. It is due to incomplete or faulty development of an organ or part. This occurs due to aplasia of bone marrow, where there is utter inactivity. The anemia seen is normochromic and normocytic. No regenerative forms are present. Causes/Types: Aplastic anemia may be divided into: *Primary or idiopathic*: This is rare.

Secondary: Exhaustion: Occurs due to chronic hemorrhages e.g., gastric or intestinal ulcers, blood sucking worms, deficiency of vitamin C, K and prothrombin. Toxic: e.g., chemical poisons, ionizing radiations, etc. when exposed at higher doses for longer duration. *Metabolic*: In piglets which are born from the sow which is suffered from protein malnutrition at pregnancy. *Myelopthistic (myelo- + phthisis- wasting or replacement of BM or exaution of BM) Anemia*: Myelo-phthisic anemia is a normocytic, normochromic anemia that occurs when normal marrow space is infiltrated and replaced by nonhematopoietic or abnormal cells. In this condition there is replacement of the bone marrow by other tissues. An anemia in which the blood-forming elements of the bone marrow are unable to reproduce normal blood cells In this disease immature forms of granulocytes are found in the peripheral blood; it is also known as leuco-erythroblastic anemia. *Causes*: This condition seen in; Tumors, specific toxins, granulomatous disorders, lipid storage diseases, and primary myelofibrosis. Secondary metastasis of other tumors- lymphatic leukemia in dogs and cats. Osteodystrophies in which the myeloid tissue is replaced be connective tissue. Primary tumors of the RE system- Nieman- pick disease, Hodgkin's disease.

Assessment of Anemic Cases

On the basis of, A. Clinical Signs of Anemia: B. Clinical Examination in Anaemic Cases.

Diagnosis of Anemia: As with any disease, the diagnosis rests on information gained from historical, physical and laboratory findings. *History*: The following may be significant, Drug administration, Exposure to toxic chemicals,

Family or herd occurrence, Recent transfusions or colostral ingestion, Age at onset.

Physical Findings: Clinical signs suggesting the presence of anemia and these includes; Pale mucus membrane, Weakness, loss of stamina and exercise intolerance, Tachycardia and polypnea particularly after exercise, Hypersensitivity to cold, Heart murmur caused by reduced viscosity and increased turbulence of the blood, Shock if one third of blood is lost in a short period of time, Icterus, hemoglobinuria, hemorrhage or fever, depending on the pathophysiologic mechanism involved. Signs are less marked if onset is gradual and animal can adapt to the decreased erythrocyte mass.

Laboratory Findings: PCV is the easiest, most accurate method for detecting anemia. Its results should be interpreted with the knowledge of the hydration status and any alteration by splenic contraction. Hb concentration and RBC count may be used. Other laboratory procedure includes; Erythrocyte indices which are helpful in classification of anemia. Peripheral blood examination of RBC morphology- for morphological classification of anemia.

Reticulocyte counts: For regenerative & non-regenerative anemias. Assessment of IMHA- three criteria-i) presence of in-saline agglutination or ii) significant positive titre by Coombs test and iii) The presence of significant Spherocytosis.

Peripheral blood smear examination: for parasitic anemia (parasites occur within or on the cell surface- hemoprotozoal diseases). *Hemobartonella felis*, *Anaplasma marginale*, *Babesia canis, B. equi, B. caballi* and *Eperythrozoon suis* are most commonly encountered). ***Serum biochemical assessment***: General health parameters, STP, LFT, KFT, icterus index.

Serum micromineral assessment: Iron, copper, cobalt, zinc, Folic acid and $VitB_{12}$ estimation. For parasitic anemias (due to helminthic and other parasites)- you will have to collect and process the faecal samples collected from suspected cases.

Microbiological assessment: for bacterial and viral diseases (for haemolytic and haemorrhagic anemia) associated with anemia

Bone Marrow Examination

For both regenerative & non-regenerative types and for detection of other bone marrow abnormalities. Bone marrow evaluation is indicated when peripheral blood abnormalities are detected. The most common indications are persistent neutrophilia, unexplained thrombocytopenia, poorly regenerative anemia. Bone marrow examination in anemias is performed primarily to evaluate the erythropoietic response. Erythropoiesis is reduced in depression anemias yielding an elevated myeloid: erythroid ratio. In Veterinary medicine, bone marrow aspirates are done more frequently than core biopsies. Aspirates are easier, faster and less expensive to perform than are core biopsies. Bone marrow core biopsies require special needles that cut a solid core of material, which is then placed in fixative, decalcified, embedded, sectioned, stained and examined microscopically by a pathologist. Core biopsy sections provide a more accurate way of evaluating marrow cellularity and examining for metastatic neoplasia than do aspirate smears, but cell morphology is more difficult to assess.

Sites of Bone marrow biopsy: Ilium, proximal femur, proximal humerus, sternum and proximal ribs.

Morphological identification of cells: When marrow particles examined microscopically, they contain blood cell precursors, vessels, reticular cells, macrophages and plasma cells. Fat is dissolved away during alcohol fixation. Megakaryoblasts are the earliest recognizable cell having single nucleus and deeply basophilic cytoplasm. Erythroid series rubriblasts, prorubricytes are seen.

Application of research: Study of classification of anemia

Research Category: Veterinary and Animal Sciences

Acknowledgement / Funding: Authors are thankful to Department of Pathology, College of Veterinary and Animal Sciences, Parbhani, 431402, Maharashtra Animal and Fishery Sciences University, Nagpur, 440006, India

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Research project name or number: Review study

Author Contributions: All authors equally contributed

Author statement: All authors read, reviewed, agreed and approved the final manuscript. Note-All authors agreed that- Written informed consent was obtained from all participants prior to publish / enrolment

Study area / Sample Collection: College of Veterinary and Animal Sciences, Parbhani, 431402

Cultivar / Variety / Breed name: Nil

Conflict of Interest: None declared

Ethical approval: This article does not contain any studies with human participants or animals performed by any of the authors.
Ethical Committee Approval Number: Nil

References

[1] Vegad J.L. and Swamy M. (2010) *A textbook of Veterinary Systemic Pathology. 2nd Edn. Lucknow, IBDC publishers Uttar Pradesh, India*, 266.
[2] Runnells R.A., Monlux W.S., Monlux A.W. (1976) *Principles of Veterinary Pathology. 7th edn., Scientific Book Agency, Calcutta*, 1309-1325.
[3] Brar R. S. (2002) *Veterinary clinical diagnosis by laboratory Methods, 1st edn. Kalyani publications, Ludhiana*, 12-69.
[4] Radostits O. M., Gay C.C., Hinchcliff K.W. and Constable P.D. (2007) *Veterinary Medicine. A text book of the diseases of cattle, horses, sheep, pig and goats. 10th Edn. W.B. Saunders Co. Ltd. London,* 414-416.
[5] Hoff brand A.V. and Pettit J.E. (Ed). (1993) *Essential Hematology*, 3rd Edn, 428- 439.
[6] Giger U. Regenerative anemia caused by blood loss or hemolysis. In, Ettinger JE, Feldman EC, (2005) *Textbook of Veterinary Internal Medicine. St. Louis (MO), Elsevier Saunders*, 1886-1907.
[7] Balch A., & Mackin A. (2007) *Compend*., 29(4), 217-225.
[8] Lewis M., Schwartz R.S., Gilmore C.E. (1965) *Ann N. Y. Acad. Sci*., 124(1), 178-200.
[9] Reimer M.E., Troy G.C., Warnick L.D. (1999*) J. Am. Anim. Hosp. Assoc*., 35(5), 384-391.
[10] Burgess K., Moore A., Rand W., Cotter S.M. (2000) *J. Vet. Intern. Med*., 14(4), 456-462.
[11] Grundy S.A., Barton C. (2001) *J. Am. Vet. Med. Assoc*., 218(4), 543-546.
[12] Weinkle T.K., Center S.A., Randolph J.F., Warner K.L., Barr S.C., Erb H.N. (2005) *J. Am. Vet. Med. Assoc*., 226(11), 1869-1880.
[13] Piek C.J., Junius G., Dekker A., Schrauwen E., Slappendel R.J., Teske E. (2008) *J. Vet. Intern. Med.*, 22(2), 366-373.
[14] Piek C.J., van Spil W.E., Junius G., Dekker A. (2011) *BMC Vet. Res*., 7, 15.
[15] McDevitt H.O. (2000) *Annu Rev. Immunol*., 18, 1-17.
[16] Corato A., Shen C.R., Mazza G., Barker R.N., Day M.J. (1997) *Vet. Immunol. Immunopathol*., 59(3-4),191-204.
[17] Angles J.M., Kennedy L.J., Pedersen N.C. (2005) *Tissue Antigens*, 66(3),173-184.
[18] Kennedy L.J., Barnes A., Short A., Brown J.J., Lester S., Seddon J., Fleeman L., Francino O., Brkljacic M., Knyazev S., *et al*., (2007a) *Tissue Antigens*, 69(Suppl. 1), 272-288.
[19] Kennedy L.J., Barnes A., Short A., Brown J.J., Lester S., Seddon J., Happ G.M., Ollier W.E. (2007b) *Tissue Antigens*, 69(Suppl. 1), 289-291.
[20] Carr A.P., Panciera D.L., Kidd L. (2002) *J. Vet. Intern. Med*., 16 (5),504-509.
[21] Klag A.R., Giger U., Shofer F.S. (1993) *J. Am. Vet. Med. Assoc*., 202 (5),783-788.
[22] Scott-Moncrieff C. (2001) *J. Am Anim Hosp Assoc*., 37(1),11.
[23] Mellett A.M., Nakamura R.K., Bianco D. (2010) *J. Vet. Intern. Med*., 25(1), 71-75.
[24] Day M.J. (1999) *Vet. Immunol. Immunopathol*., 69(2-4),215-224.
[25] Cain S.M. (1977) *J Appl Physiol*., 42(2), 228-234
[26] Thompson M.F., Scott-Moncrieff J.C., Brooks M.B. (2004) *J. Am. Anim. Hosp. Assoc*., 40(6),446-454.
[27] Morley P., Mathes M., Guth A., Dow S. (2008) *J Vet Intern Med*., 22(4), 886-892.
[28] Couto C.G. Anemia. In, Nelson R.W. (2009) *Small animal internal medicine. St. Louis (MO), Mosby Elsevier,* 1215.
[29] Day M.J. (2008) *Immune-mediated haematological disease. In, Clinical immunology of the dog and the cat. Day MJ, editor. London, Manson Publishing Ltd*., 94-120.
[30] Ganti A Sastry, Veterinary Clinical Pathology, CBS Publishers & Distributors Pvt. Ltd., Delhi, 1-25.
[31] Chulilla J.A.M., Colás M.S.R., & Martín M.G. (2009) *World Journal of Gastroenterology, WJG*, 15(37), 4627.
[32] Kennedy L.J., Barnes A., Short A., Brown J.J., Seddon J., Fleeman L., Brkljacic M., Happ G.M., Catchpole B., Ollier W.E. (2007c) *Tissue Antigens*, 69(Supp1), 292-296.
[33] Thomas Carlyle Jones and Ronald Duncan Hunt (1979) *Veterinary Pathology, 4th Edn; Reprinted, K.M.Varghese Company, Bombay*, 1309-1336.