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Vol. 7(48), pp. xxxx-xxxx, 28 February, 2014 DOI 10.5897/AJPP2013.4010 ISSN 1996-0816© 2013 Academic Journals Copyright© 2014 Author(s) retain the copyright of this article http://www.academicjournals.org/AJPP

African Journal of Pharmacy and Pharmacology

Full length research paper

Effect of oral supplementation of *Aloe vera* extract on haematology indices and immune cells of blood in rabbit

Amjad Ali Channa¹*, Izhar Hyder Qazi², Saeed Ahmed Soomro³, Atta Hussain Shah⁴, Jameel Ahmed Gandahi⁵, Roshan Ali Korejo¹, Imtiaz Ahmed Shah¹, Nawab Ali Kalhoro¹ and Band-eali Khaskeli¹

¹Faculty of Animal Husbandry and Veterinary Sciences, Sindh Agriculture University, Tandojam, Pakistan. ²Department of Veterinary Anatomy and Histology, Faculty of Bio-Sciences, Shaheed Benazir Bhutto University of Veterinary and Animal Sciences, Sakrand, Pakistan.

³Department of Physiology and Biochemistry, Sindh Agriculture University, Tandojam, Pakistan.
⁴Department of Animal Products Technology, Sindh Agriculture University, Tandojam, Pakistan.
⁵Department of Anatomy and Histology, Sindh Agriculture University, Tandojam, Pakistan.

Received 1 January, 2014; Accepted 24 April, 2014

Aloe vera is being widely used in herbal medicine as antibacterial, antiviral, antifungal, and antiinflammatory agent. It is also used as topical application for many skin diseases; its antidiabetic effect in rats is also reported. Looking at its enormous medicinal effects, the present study was conducted to investigate the effects of oral supplements of *Aloe vera* extract on hemotology indices and immune cells of rabbit. Total of twenty (n=20) rabbits were selected and divided into 4 group's i-e A, B, C and D with n=5 in each group. Group A was kept as control while B, C and D were given 400, 500 and 600 mg/kg of *Aloe vera* extract, respectively daily for 21 days. Blood samples were collected from central ear vein transferred into test tubes containing ethylene diamine tetra acetate (EDTA) on 0 day, 7th day, 14th day and 21st day and analysed for complete blood count (CBC). Results revealed that oral supplementation of *A. vera* extract significantly increase mean corpuscular haemoglobin (MCH), mean corpuscular volume (MCV) and lymphocytes at level (< 0.05) and decrease neutrophils in normal domestic rabbits. Results of this study concluded that *A. vera* has hematopoetic and immunomodulatory effects; thus its extract can be used for the treatment of anemia and immune deficiency problems.

Key words: Aloe vera, biological active, anaemia, immunodeficiency.

Introduction

Aloe vera, derived from an Arabic word "Alloeh" means shinning bitter substance and vera means 'true' (Ombrello, 2008). It is a succulent cactus like perennial plant originated from arid climates of North Africa (Akinyele et al., 2007). There are about 275 species worldwide (Nandal and Bhardwaj, 2012). *A. vera* contains 99.5% water and 0.5% solids(Hamman, 2008). It contains 200 chemical compounds out of which 75 are biological

*Corresponding author. E-mail: brainboss@ymail.com. Author(s) agree that this article remain permanently open access under the terms of the <u>Creative Commons Attribution</u> License 4.0 International License active, that is, aloin, *Aloe* polysaccharide (Jun et al., 2005), reducing sugars, organic acids, enzymes, metallic cations and lecithin (Eshun and He, 2004). Due to antibacterial properties, *A. vera* is used against commonly found bacteria including *Staphylococcus, Streptococcus, Klebsiella, Pseudomonas, Escherichia coli, Salmonella*, etc., (Lawrence et al., 2009). It is also used to treat many viral diseases and enhance immune system by acting as effective antiviral agent against human immunodeficiency virus (HIV) and hepatitis (Rabe et al., 2005). Research has investigated that *A. vera* have positive influence on central nervous system, especially on the ependymal cells at ventricular zone (Rengin et al., 2008).

Besides antibacterial and antiviral effects, *A. vera* has antifungal, antineoplastic activities and widely used for topical applications as an antiaging agent (Ivan, 2006). *A. vera* significantly influences the function of liver and pancreas by influencing aspartate amino transferase (AST) and alanine transferase (ALT) (Iji et al., 2010). It has been reported that treatment of normal as well as diabetic rats with *A. vera* significantly reduces its blood sugar level indicating that it has antidiabetic effects (Rehman et al., 2011).

Keeping in view the clinical importance of *Aloe vera*, the present study is conducted with objectives to determine: (a) effects of oral supplementation of *A. vera* on haematology indices and immune cells in blood of rabbits, (b) the safe and effective therapeutic dose of *A. vera* through oral route of administration.

MATERIALS AND METHODS

A. vera leaves were used in this study. Fresh leaves of A. vera were splitted into two parts after removing spiny margins; white transparent pulp was scraped out and weighed. Hundred grams (100 g) of A. vera was blended and boiled in 200 ml of distilled water for 10 min, after boiling it was cooled at room temperature and filtered with muslin cloth. The extract thus obtained was stored in a refrigerator at 4°C until use. A total of 20 male rabbits of an average body weight: 1.4 kg were used in this study. Rabbits were kept at animal house under control environment. The rabbits were given 0.25 kg rice and 0.25 kg green grass, especially Alpha alpha and water was given as ad-libitum. Rabbits were divided into four groups, that is, A, B, C and D consisting of 5 in each group. Group A was kept as control, while groups B, C, and D were given A. vera extract orally at the dose of 400, 500 and 600 mg/kg body weight, respectively daily for 21 days. Blood was collected before treatment day 0, and then days 7, 14, and 21st post treatment. Blood was collected from central ear vein, and transferred to test tubes containing anticoagulant (EDTA: Ethylene diamine tetra acetate), and brought to Post Graduate Laboratory of Veterinary Physiology, Sindh Agriculture University, Tandojam for CBC analysis.

RESULTS

Mean corpuscular hemoglobin (MCH)

As shown in Figure 1, MCH of rabbits that received oral

supplementation of *A. vera* extract increased from day 0 to 21st post treatment. MCH of groups A, B, C and D was 15.42, 15.34 14.94 and 15.24 pg and 15.46, 21.18, 21.14 and 21.54 pg day 0 and 21st day post treatment, respectively. Results revealed gradual increase in MCH values of treatment group. The statistical analysis of mean MCH values of treatment and control group indicated significantly (P<0.05) higher values in treatment than MCH values of the control groups.

Mean corpuscular volume (MCV)

Results shown in Figure 2 indicate gradual increase in values of MCV in the blood of rabbits in treatment group. MCV of groups A, B, C and D were 58.48, 58.558, 58.48 and 58.54 cu and 58.56, 65.72, 65.04 and 65.66 cu, on day 0 and day 21st, respectively. Statistical analysis showed significantly higher (P<0.05) MCV of rabbit's blood in treatment group than control.

Neutrophils

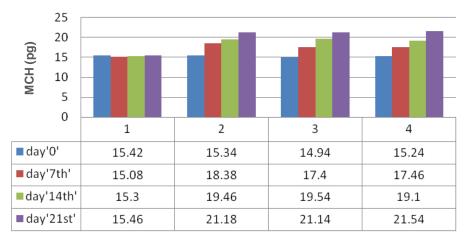
The neutrophil counts in the blood of rabbits in treatment group, that is, B, C and D significantly decreased (P<0.05). As shown in Figure 3, the neutrophil count of groups A, B, C and D was 45.94, 45.46, 45.5 and 45.38 and 45.76, 29.18, 24.52 and 23.38 on day 0 and 21st day. There was significantly lower (P<0.05) neutrophil in groups B, C, D as compare to control group A.

Lymphocytes

Oral supplementation of *A. vera* significantly increased lymphocyte in treatment groups B, C, and D, that is, 43.66, 43.86, 43.68 and 57.84, 60.32, 61.74 (Figure 4) from day 0 to 21st post treatment, respectively.

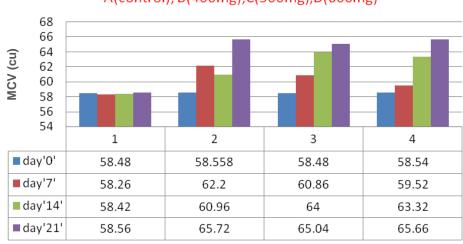
DISCUSSION

Haematology parameters such as red blood cell count, hematocrit, hemoglobin concentration and derived hemotology indices, e.g MCV, MCH and MCHC are important blood marker for diagnosis of various blood diseases. The excessively decreased values of these parameters indicated different types of anaemia in human as well as animals caused by loss of blood through hemorrhage, bone marrow disease, iron deficiency, vitamin B12 deficiency, or folic acid deficiency, etc. Various therapeutic medicines are used for the treatment of blood diseases. Nowadays, remedies through herbal medicines (alternative medicines) have been more demanding due to their natural source and less side effects. The results of this study have indicated that *A*.



MCH of group A(control), B(400mg), C(500mg), D(600mg)

Figure 1. Mean corpuscular haemoglobin (MCH) on various days.

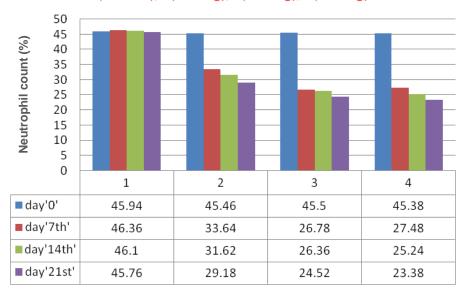


MCV of group A(control), B(400mg),C(500mg),D(600mg)

Figure 2. Mean corpuscular volume (MCV) on various days.

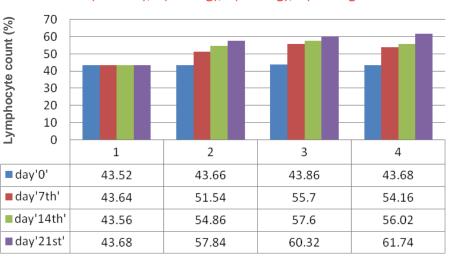
vera leaf extract significantly increase (<0.05) in MCH and MCV values. The erythropoietin effect of A. vera extract of hemopotetic cells of bone marrow have been reported by Iji et al. (2010). Similar findings have been reported by Hamman (2008), who has attributed the effect of increasing MCH to essential vitamins, e.g. riboflavin, thiamine and folic acid; and essential and non essential amino acids in A. vera, that are required for synthesis of haemoglobin. Research has also been investigated that this increase in erythropoiesis is due to the presence of polysaccharides in A. vera leaf gel (Ni et al., 2004). It has been investigated that these hemopoietic properties of A. vera is only due to the presence of thiamine that is responsible for formation of α-ketoglutarate dehydrogenase and pyruvate dehydrogenase complex in Krebb's cycle (Oishi et al., 2002).

This enhancement in MCV also reveals aloe polysaccharide that can initiate the formation of cells. These results are contrary to Iji et al. (2010), who investigated no change occurs in MCV by chronic administration of *A. vera*. This also contradict the study of Taiwo et al. (2005) who had investigated that *A. vera* can cause tissue necrosis, heart, liver and kidney damage in fish In this study, it was found out that neutrophils count significantly decreases in rabit blood treated with varying doses of *A. vera*. However, studies are being conducted to investigate the use of *A. vera* for treatment of cancer and blood disease associated with low white blood cell count, that is, leukaemia. It has been reported that



Neutrophil count of group A(control), B(400mg), C(500mg), D(600mg)

Figure 3. Neutrohil count on various days.



lymphocyte count of group A(control), B(400mg), C(500mg), D(600mg

Figure 4. Lymphocyte count on various days.

veterinarians use *A. vera* for the treatment of cancer and feline leukaemia in animal patients. *In vitro* studies by British Cancer Research, UK has identified the presence of a compound in *A. vera*, named di 2-ethyl hexyl phthalate (DEHP), which stop the development of leukaemia cells in test tubes. Thus, the findings of these studies are very much important and the mechanisms involved in reducing neutrophil count should be further investigated.

A. vera has immunomodulatory effect that has been observed in the results of this study. Treatment of varying doses of A. vera extract to rabbits significantly increased (P<0.05) the total lymphocyte concentration. Lymphocytes are important blood cells that play a role in developing humoral and T cell immune response against foreign antigens, that is, bacterial, viral, etc. It is thought that a molecule in the aloe gel, known as acemannan, stimulates the body to produce disease-fighting white blood cells, particularly macrophages. Macrophages engulf and digest unwanted substances, such as bacteria and viruses.

It has been investigated that when any foreign protein enter the body in response, some cytokines produced enhance cell mediated and antibody immune response (leung et al., 2004; Im et al., 2005). Research has investigated that A. vera has initiated phagocytic activity of recticuloendothellial system (Im et al., 2005). It has been investigated that it can enhance cellular as well as humoral immunity by proliferation of myeloid and erythrocyte colony forming cells, macrophage colony forming cells and pleuri-potent hemopoietic cells (Boudreau and Beland, 2006). It has been investigated that it significantly improves B lymphocyte that is responsible for formation of both immunoglobulin's M and G (Yates et al., 1992). It has also been investigated that A. vera can increase CD^4 and CD^8 receptor of T lymphocytes (Ghasem et al., 2011). Thus, increase in lymphocyte concentration might be due to the presence of low molecular weight proteins and glycoproteins that have mitogenic effect causing proliferation of immune cells, that is, lymphocytes.

Results of these studies also revealed that *A. vera* has dose dependent effects. It is observed that doses of 500 and 600 mg were slightly more beneficial than 400 mg at the level of p<0.05 (Figures 2 and 4). Our findings are in accordance with that of Iji et al. (2010). They reported that *A. vera* leaf extract is dose dependant.

Conclusion

The present research work investigated that the oral administration of *A. vera* extract showed significant increase in MCH and MCV level, therefore it was concluded that liquid extract of *A. vera* plant may be useful in the treatment of iron deficiency anaemia. It may be used as immune-stimulant. It is believed that it will be helpful in further investigation.

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