

AMELIORATION OF ALUMINIUM FLUORIDE TOXICITY BY ALOE VERA ON BLOOD PARAMETERS IN MALE ALBINO RATS

Kusum Kushwah*, Akansha Rao and Dharmendra Pratap Singh

Department of Zoology, Agra College, Agra. DR. Bhimrao Ambedkar University, Agra (U.P.) India

* Author Email: drkusumkushwaha@gmail.com

ABSTRACT:

It is suspected that drinking water containing fluoride (F) and aluminum (Al) results in negative health effects in the human and animals. Its toxic effect in the especially on blood, brain, liver, and kidney. In this investigation, the effect of AlF_3 blood disease was noted. Albino rats were treated either with WHO recommended or slightly higher F and Al levels in drinking water. Treatment solutions contained 0.05–10.0 mg/L of F, and 0.08–10.0 mg/L of Al, and the treatment period was 45 days. Aloe vera gel (Aloes) is a member of the family Liliaceae that is used as herbal or medicine in many cultures for different purposes. The present study was designed to observe the role of Aloe vera gel on blood parameters in rats. The present study was designed to assess the ameliorative effect of Aloe vera gel on aluminium fluoride induced toxic changes in blood parameters of albino rats. The present study was carried out 60 male albino rats that were randomly divided into three groups consisting of 20 rats in each group. Aluminium fluoride was gavages per orally using distill water as vehicle 200mg/kg b. w. to group II and Aloe vera 300mg/kg b. w. in distilled water was given along with AlF_3 to group III for 45 days to study of ameliorative effects group I is a control group. Ten rats from each group were sacrificed at sixteenth and forty sixth day night interval. Entire changes were observed in blood cells. The haematological parameters TEC, Hb. Conc., MCV significant decrease treated with AlF_3 , and TLC, PVC, MCH, MCHC and ESR significant increase after 45 days treated with AlF_3 compared with the control group. Treated with AlF_3 along with Aloe vera gel all parameters significant increased compared with the control group. These results showed that Aloe vera gel have haematoprotective effects in experimently induced AlF_3 rats. This study concluded that Aloe vera gel has haematoprotective effect against aluminium fluoride induced haematotoxicity.

Keywords: Aloe vera, Albino rats, Anemia, blood parameters, Aluminium fluoride and Aloe vera.

Introduction

Fluoride is an important component but one that could risk health issues when fluoride contained in drinking water at a concentration more than one ppm or in the area where a huge quantity of fluoride is released because of the burning of F loaded coal [1]. Fluoride cause deleterious effects in organ such as

brain, liver and kidney [2] by passing by the cell membrane [3]. F deposits in bone, organs where it could negative impact hematopoiesis caused in bone marrow [4]. In same as a site of actively metabolism, the liver is susceptible to F poisoning [5]. However, it's been suggested that adjustments due to fluoride exposure arise in advance in blood in comparison to other tissues and organs [6].

Previous research revealed that fluoride has destructive outcomes on haematological parameters [7] in rats. Fluoride is a general constituent of fluid present in body, soft tissue organ, bones and dental cavity. Fluorine is an essential element to body of human and plays a key character in the protection and are treatment raise exposure to fluoride might occur from natural or sources of industries and from wrong uses of fluoride containing dental cavity care component. While, F_l have a capacity to administrate in the animals if the exposure persisted over duration, even at slow concentration.

Fluoride is a ubiquitous environment compound and broadly industrial pollutant released into the nature by a mixture of natural and anthropogenic processes. In less conc. F_l have been reported beneficial for teeth and bone growth F_l compound were extensively used in industries, agriculture and chemicals like cleansing products, reddened. As a observation every year these was 1000 of reports related to poisoning due to highly effects of F_l composition of dental material and suicidal exposure to chemicals contained at house, and laboratories. Long term exposure of elevated fluorine doses found that adverse effects like dental and mental disturb situation, while large quantity doses of causes serious pathological changes.

Aluminium is an abundant element on the Earth crust. It is utilize elevated due to its excellent properties. It have been no any one of known biological role, when administrate in the body it could be induce various biological disorder like hepatotoxicity, neurotoxicity [8], bone disease and lack of haemoglobin [9] Al have effect on hematopoiesis [10] and that elevated stages in serum of patient suffer from hemodialysis were conducted with impaired erthropoiesis and deficiency of iron [11]. It can be induced in our body by many vaccines, mostly in pandemic causes, to make the more stability and effective impact of vaccines [12].

With the broadly uses of biochemical findings and biological results, it could be beneficial to detected the toxicity of metal effected the injuries in organ cause by it. And in daily live interact with environment both human and animal and exposed of chemical and heavy metal like Al and their compounds. The toxicity of AlF₃ many of the procedure can be found one of them was herbal/medicinal plant treatment. Utilization of medicinal plant treatment free of cost anybody could be used and take the benefits of their properties Author also be advised that utilization of Al materials reduced in daily needs also help to decrease the toxicity of metals.

Aloe vera (*Aloe barbadensis* Miller) is one of the extreme valuable plant in the Liliaceae family and its use recent time in medical, beauty and health properties. *Aloe vera* contains more than seventy five nutrients, twenty minerals, eighteen amino acids and two hundred active compounds, and twelve vitamins and compound such as aloin, antrokihon, famodin and barbaloin [13], [14], [15]. This *Aloe vera* plant has a broad variety of pharmacological effects such as wound healing, skin lesions, antibacterial and antiviral and others effects have been credited to this plant. *Aloe vera* gel has equally been exhibited to have antioxidant capacities in human and animals. It demonstrates the capacity of radical scavenging, which is more

important than the capacity tocopherol [16], [17]. The compound Aloe includes enzymes, antioxidant, fat and water soluble, polysaccharids, organic acid and phenolic compounds. *Aloe vera* gel is excellent for health. *Aloe vera* helps in naturally detoxifying body. Aloe contains vitamin-C, E B9 and B12. It also holds minerals like copper, potassium, sodium, magnesium, selenium, zinc and manganese etc. aloe gel have been centenaries for its beauty, health and medicinal properties (NCCIH).

So it could be appears that toxicity of AlF_3 is dose dependent. Our study aimed to evaluate the possible pathological alteration in rat haematological parameters. In this study evaluation was done of blood parameters of albino rats and these were accumulated to aluminium fluoride solution for duration of selected doses. And cured this toxicity by *Aloe vera* gel [18].

The blood is fluid connective tissue, provides the body's cells with oxygen and removes carbon dioxide, transports nutrients and hormones, regulates body temperature, platelets clot blood at sites of injury, brings waste products to the kidney and liver, red blood cells are the most numerous living cells in blood, and white blood cells protect the body from pathogens. It helps in many physiological and metabolic functions of body. The chief function of the blood is to the transport of oxygen and food materials to all the cells of body and the removal of a excretory products from the body with the help of excretory organs. The maintenance of blood pH within normal limits (7.35-7.45), called acid-base homeostasis, is a complex synergy involving three organs as well as chemical buffers in blood and blood cells (erythrocytes).

Albino rat will be used for this study because they are physiologically similar to humans. They are gentle and easily caged animals, they can easily be handled, they do not transmit disease to humans and they grow quickly and therefore results can be seen in a few weeks [19].

Keeping these points in view, the present study was undertaken to show the toxic effect of aluminum fluoride on haematological parameters and protection by *Aloe vera* in albino rats.

MATERIALS AND METHODS

Experimental model Animals

60 Adult male albino rats of weighing between 150-200mg. the animals were kept from the animal house of the school of life science khandari campus, Agra. All animals were preserved under a suitable environment at 20°C with 12 hours dark and 12 hours light cycle and 50-70% humidity. All rats were maintained on good quality diet and water was given ad libitum.

Experimental Protocol

Albino rats divided into three groups, I group-Control group, II group is treated with aluminium fluoride (200mg/kg body weight) [20] , and III group is treated with aluminium fluoride along with *Aloe vera* gel (300mg/kg body weight) [21]. For 45 days. All treatments were given orally with a gavage tube.

Group I animals served as control group (untreated). To group II animals treated with aluminium fluoride (AlF_3) perched from (Indian Biologicals) was administered at a dose of 200mg/kg b. w. for 45 days.

To group III animals treated with aluminium fluoride along with *Aloe vera* (300mg/kg b. w.) for 45 days. Animals (albino rats) were observed at least twice a day, for medically symptom and signs of toxicity. Their

diet and water consumption was noted after every 24 hours, and the animal body weight was balance at the termination of the experiment (45 day treatment).

At the end of each treatment albino rats were weighed on an animal weighing balance and sacrificed by heart puncture for the haematological parameters tests and kept at room temperature for one hour.

Collect blood samples in the EDTA vials for haematological parameters analysis. All vial used in this research naming properly for avoid any confusion.

Drugs and Chemicals

Aluminium fluoride (AlF_3) used in this study. AlF_3 is a colour less, odor less and inorganic compound. AlF_3 formshydrates $\text{AlF}_3 \cdot x\text{H}_2\text{O}$. Its density is 3.10 g/cm, solubility in water 5.6 g/L and melting point 1,290°C. (Wikipedia). This chemical was of analytical grade and purchased from Indian Biologicals, Baghmujafarkhan, Agra, U.P. India. *Aloe vera* (*Aloe barbadensis miller*) its chemical formula is $\text{C}_{16}\text{H}_{13}\text{NO}_3$. Collected from botanical garden in our university.

Preparation of *Aloe vera* gel for experiment

Fresh leaves of *Aloe vera* plant were collected from local garden daily and washed properly. Using a sharp knife and cuts the *Aloe vera* leaves in small sliced gently scaped into an electric blender for gel separation than its gel is collected in a beaker and filtered by muslin cloths. After this process, the doses of *Aloe vera* are ready to treat to the albino rats.

Haematological parameters were analyzed

TEC (Total Erythrocyte Counts), TLC (Total Leucocyte Count), Hb. Conc. (Haemoglobin Concentration), PCV (Packed Cell Volume), MCV (Mean Corpuscular Volume), MCH (Mean Corpuscular Haemoglobin) MCHC (Mean Corpuscular Haemoglobin Concentration), ESR (Erythrocyte Sedimentation Rate). The tested chemicals (AlF_3) altered the haematological parameters.

DATA ANALYSIS

Outcomes are expressed Mean \pm S.Em. the difference between treated and control groups were calculated by Student 't' test (using SYSTAT software program version 7.0) and significant values were compared with standard values at the respective degree freedom.

RESULT

Changes in TEC, Hb. Conc., and MCV these values are significantly decrease and TLC, PVC, MCH, MCHC and ESR significantly increased. This study reveals the effects of chronic administration of aluminium fluoride on the haematological parameters of the male albino rats. Although oral administration of the AlF_3 toxicity increased the TLC, PVC, MCH, MCHC and ESR when compared with the control group.

Table I and graph I-reveals the toxic effects of the aluminium fluoride on the blood parameters of the male albino rats. Although oral administration of the AlF_3 toxicity increased in the PVC and, significantly decreased in TEC, Hb. Conc. and MCV. Treated with *Aloe vera* all parameters smoothly increased. TEC after 15 days treated with aluminium fluoride highly significant ($p < 0.01$) decreased and treated with *Aloe vera* gel along with AlF_3 , significantly ($p < 0.05$) increased when compared with the control group. And after 45 days treated with AlF_3 highly significant ($p < 0.01$) increased and treated with *Aloe vera* gel along with

AlF₃ very highly significant ($p < 0.001$) increased when compared with the control group. TLC treated with AlF₃ after 15 days significant ($p < 0.05$) increased and treated with AlF₃ along with *Aloe vera* gel significant ($p < 0.05$) increased when compared with control group. After 45 days treated with AlF₃ highly significant ($p < 0.01$) increased and treated with *Aloe vera* gel along with AlF₃ highly significant ($p < 0.01$) increased when compared with control group. Hb. Conc. treated with AlF₃ after 15 days significant ($p < 0.05$) decreased and treated with *Aloe vera* along with AlF₃ significant ($p < 0.05$) increased compared with control group. Treated with AlF₃ after 45 days highly significant ($p < 0.01$) decreased and treated with AlF₃ along with *Aloe vera* gel highly significant ($p < 0.01$) increased when compared with the control group.

Table I: Beneficial effects of *Aloe vera* in blood parameters (TEC, TLC, Hb%, and PVC) of Albino rat after Aluminum fluoride intoxication.

S.No.	Parameters	No. of Albinorat	Period (days)	Control Group		Treated-I (AlF ₃)		Treated-II (AlF ₃ + <i>Aloe vera</i>)	
				Mean	±S.Em.	Mean	±S.Em.	Mean	±S.Em.
1	TEC	10	15	6.50	0.20	5.25	0.19**	7.35	0.22*
	Millios/CCM	10	45	7.76	0.50	8.96	0.16**	4.53	0.43***
2	TCL	10	15	5.09	0.16	5.73	0.20*	5.76	0.23*
	Th/cumm	10	45	7.31	0.43	9.12	0.43**	8.83	0.21**
3	Hb. Conc.	10	15	13.87	0.210	13.2	0.19*	14.34	0.17*
	gm/dl	10	45	14.72	0.35	9.87	0.275	11.88	0.17**
4	PVC	10	15	40.93	0.26	39.48	0.19***	41.46	0.22*
	%	10	45	42.08	0.34	40.27	0.37***	42.99	0.21**

S.Em. = Standard Error of Mean, *** = Very Highly Significant ($p < 0.001$), ** = Highly

Significant ($p < 0.01$), * = Significant ($p < 0.05$).

FIGURE I: The effect of AlF₃ and combined with *Aloe vera* and AlF₃ for 45 days in the blood parameters (TEC, TLC, Hb%, and PVC).

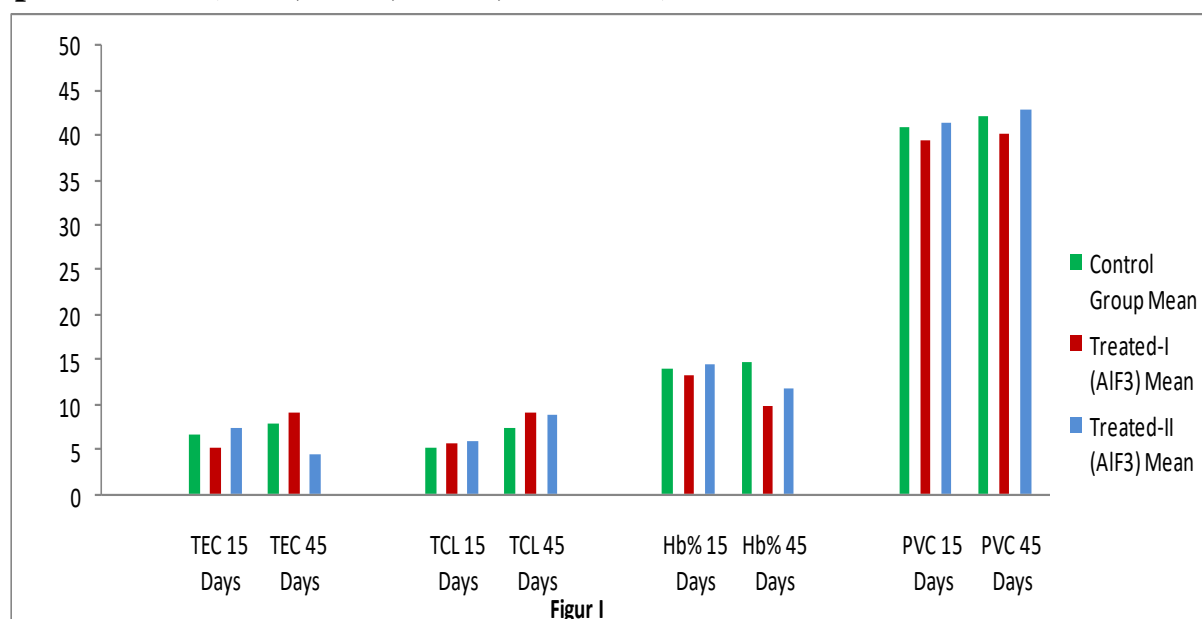


FIGURE I: Effects of *Aloe vera* for 15 and 45 days on the (1), TEC, (2), TLC, (3), Hb% and (4), PVC in albino rats treated with AlF₃. TEC, Hb%, PVC significantly decreased after 15 days and TLC increased. After 45 days TEC, TLC, smoothly increased and Hb% and PVC decreased when compared with the control group.

Table II: Beneficial effects of *Aloe vera* in blood parameters (MCH, MCV, MCHC and ESR) of Albino rat after Aluminum fluoride intoxication.

S.No.	Parameters	No. of Albino rat	Period (days)	Control Group		Treated-I (AlF ₃)		Treated-II (AlF ₃ + <i>Aloe vera</i>)	
				Mean	±S.Em.	Mean	±S.Em.	Mean	±S.Em.
5	MCH (fl)	10	15	23.05	0.22	25.18	0.37**	24.01	0.27*
		10	45	25.22	0.44	27.96	0.21***	26.84	0.24**
6	MCV (pg)	10	15	68.51	0.4	70.04	0.54*	69.36	0.27*
		10	45	72.26	0.68	68.41	0.27***	74.42	0.22**
7	MCHC (g/dl)	10	15	33.88	0.28	35.11	0.14**	35.05	0.27*
		10	45	33.51	0.22	37.56	0.25***	36.40	0.28**
8	ESR (mm/h)	10	15	4.98	0.26	5.85	0.31*	5.76	0.19*
		10	45	6.81	0.48	8.53	0.34**	8.30	0.21**

S.Em. = Standard Error of Mean, *** = Very Highly Significant ($p < 0.001$), ** = Highly Significant ($p < 0.01$), * = Significant ($p < 0.05$).

FIGURE II: The effect of AlF₃ and combined with *Aloe vera* and AlF₃ for 45 days in the blood parameters (MCH, MCV, MCHC and ESR).

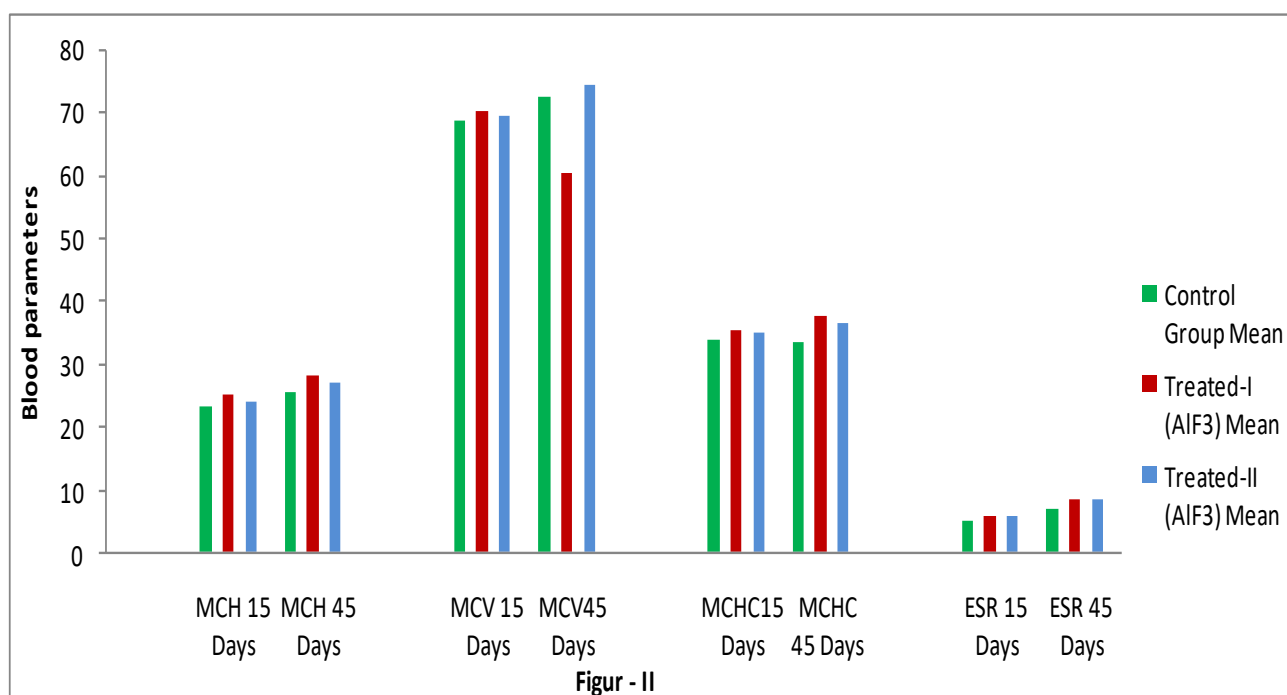


FIGURE II: Effects of *Aloe vera* for 15 and 45 days on the (1), MCH, (2), MCV, (3), MCHC and (4), ESR in albino rats treated with AlF₃. MCH, MCV, MCHC and ESR significantly increased after 15 days. After 45 days mch, MCHC and ESR smoothly increased and MCV significantly decreased, comparison with control group.

PVC treated with aluminium fluoride after 15 days very highly significant ($p < 0.001$) decreased and treated with *Aloe vera* gel along with AlF₃ significant ($p < 0.05$) increased when compared with control group. After 45 days treated with AlF₃ very highly significant ($p < 0.001$) decreased and treated with *Aloe vera* gel along with AlF₃ highly significant ($p < 0.01$) increased when compared with control group.

Table II and graph II showed the effect of chronic administration of aluminium fluoride on the haematological parameters of the male albino rats. Although oral administration of the AlF_3 toxicity increased in the MCH, MCHC and ESR, and MCV decreased treated with aluminium fluoride, and treated with *Aloe vera* all parameters smoothly increased.

MCV treated with aluminium fluoride after 15 days significant ($p < 0.05$) increased and treated with *Aloe vera* gel along with AlF_3 significant ($p < 0.05$) increased when compared with control group. After 45 days treated with AlF_3 very highly significant ($p < 0.001$) decrease and treated with *Aloe vera* gel along with AlF_3 highly significant ($p < 0.01$) increased comparison with control group.

MCH treated with aluminium fluoride after 15 days highly significant ($p < 0.01$) increased and treated with *Aloe vera* gel along with AlF_3 significant ($p < 0.05$) increased when compared with control group. After 45 days treated with AlF_3 very highly significant ($p < 0.001$) increased and treated with *Aloe vera* gel along with AlF_3 highly significant ($p < 0.01$) increased when compared with control group.

MCHC treated with aluminium fluoride after 15 days highly significant ($p < 0.01$) increased and treated with *Aloe vera* gel along with AlF_3 significant ($p < 0.05$) increased when compared with control group. After 45 days treated with AlF_3 very highly significant ($p < 0.001$) increased and treated with *Aloe vera* gel along with AlF_3 highly significant ($p < 0.01$) increased when compared with control group.

ESR treated with aluminium fluoride after 15 days ESR values significant ($p < 0.05$) increased and treated with *Aloe vera* gel along with AlF_3 significant ($p < 0.05$) increased comparison with control group. After 45 days treated with AlF_3 highly significant ($p < 0.01$) increased and treated with *Aloe vera* treated group highly significant ($p < 0.01$) increased comparison with control group.

DISCUSSION

Haematological study is a very important diagnostic tool in pathological field for diseases examine, and has also been establish valuable to detector stress due to environmental infective. Our study results showed that acute exposure of albino rats to AlF_3 induced toxicity. AlF_3 toxicity also affects on reduction in food consumption and water, body weight [22] and functional integrity of the vital organ, and alterations in hematological physiology and increase in weight of kidney and liver may be attribute to inventive of the body's habituated mechanisms to combat systemic AlF_3 toxicity, but the haematological parameters suggested toxic effects of the chemicals.

The present study suggested reduction in the values of TEC, Hb Conc., MCV (AlF_3 treatment) and TLC, PCV, MCH, MCHC and ESR significantly increased. Our outcomes are in accordance with those of others. TEC, Hb Conc. significantly decreased [23], [24], [25] and MCV [26]. TEC, Hb Conc., and MCV, all parameters treated with *Aloe vera* gel slightly significantly increased our outcomes similar to others research papers [17].

Aloe vera has also been notably used in the treatment of many diseases such as gout, arthritis, peptic ulcer and dermatitis as well as treatment of burns [27]. The fresh *Aloe vera* gel, juice and formulated products have long been used for cosmetic and medical purposes and normal health [28], [29]. Higher Hb. Conc. as well as PCV and RBC values in the rats that received the aloin gel is an representation that the aloin gel

stimulated incremented erythropoiesis in those albino rats, even though its strength as a haematic is low, in view of the fact that the effects on RBCs count and PCV were not pronounced.

The polysaccharides, which are the prime components of the *Aloe vera* gel, have also been noted to stimulate erythropoiesis [30], [31]. Deficiency of thiamine therefore outcomes in thiamine deficiency megaloblastic anemia syndrome [32]. The haematoprotective action of the *Aloe vera* plant were preservation of the blood parameters through the antioxidant properties of the aloin. Moss and Hathway (1964)[33] suggested permeability of the erythrocytes membrane to pollutants, which may decrease life span and production of erythrocytes due to harm of erythrocytic tissue [34].

When TEC decreased than decreased red blood cells, haemoglobin and packed cell volume (PCV) as a result of decreased rates of erythrocytes production and increased rates of erythrocytes destruction or loss. A low value of RBC count also displays a vit. B6, B12 and folate deficiency. It's also cause internal bleeding, malnutrition and kidney diseases (www.nhs.uk). Decrease the TEC may be due to bleeding, anemia, myeloma, bone marrow failure, erythropoietin, leukemia and hemolysis.

Hb. Concentration-Anemia is a problem of not having enough healthy RBCs or haemoglobin to carry O₂ to the body's tissues. Having anemia can cause tiredness, weakness and shortness of breath. Acute anemia can results from various causes, including hemorrhage, trauma, hemolysis and other acute conditions.

A high level of MCV means our RBCs are larger than normal RBCs, a condition called macrocytosis. Some people with high MCV have anemia. A high MCV can be associated with MDS, on uncommon type of cancer. It also affects cells in the bone marrow. High MCV, such as liver disease, and vitamin deficiency are for more common.

TLC, PCV, MCH, MCHC and ESR parameters are significantly increased after 15 and 45 days.

TLC – decrease TLC a lower – than normal TLC , known as leucopenia, may be autoimmune disorders, associated with viral infection and bone marrow problems. PVC (hematocrit) level that is higher than the normal range can be a sign that the body is making too many RBC (Red Blood Cells). That can be caused by congenital heart disease, lung disease, heart failure and polycythemia. MCH - A high level of MCH means red blood cells are larger comparison than normal. This is called macrocytosis. Higher MCH score are generally a sign of macrocytic anemia. MCHC – A high MCHC measurements is known as hyperchromia. It suggests that we have a higher conc. of hemoglobin in red blood cells. Cause of high MCHC includes some types of anemia, over action thyroid and liver disease.

ESR- If an ESR test outcomes shows that RBCs sink faster than normal RBCs, it may mean medical condition causing inflammation.

In this study albino rats administered with the *Aloe vera* gel along with AIF₃ had increased value of the TEC, TLC, Hb. Conc., PCV, MCV, MCH, MCHC and ESR were observed in albino rats administrated with the aloin for 45 days. This increase in PCV was not due to Hb. Conc. because there was a generalized increment in TEC and TLC cells, but can be credited to stimulation of haematopoiesis related to the outcome of the red blood cell indices, can be used further incremented MCV, MCH and MCHC, which appeared that immature red blood cells were present in indication, circulation of stimulation of production of immature erythrocytes,

to known as reticulocytes. In morphologically, reticulocytes are characterized by increment in the size of red blood cells in circulation and it is usually noted as the initial response to stimulation of the haematopoietic system [35].

TLC (total leucocytes count) on the other hand showed significant increases, particularly in albino rats administered with the gel for 45 days. This result obtained for this outcome, it can be suggested that *Aloe vera* gel may contain bioactive compounds which are effective of mobilizing all blood cells types into circulation or stimulate haematopoiesis out coming incremented production and release of blood cells into the circulation. A significant decrease in Hb. Conc., was noted which was reflective of the decline in haematocrit and red blood cells counts and agree with the outcomes [24].

This result obtained for this outcome, it can be suggested that *Aloe vera* gel mat contain bioactive compound which are effective of mobilizing all blood cells type into circulation or stimulate haematopoiesis out coming in incremented production and release of blood cells in to the circulation [36]. A significant decrease in Hb. Conc. was noted which was reflective of the decline in haematocrit and red blood cells counts and agree with the outcomes [24].

Reports also have noted that F induced disorders in hematopoetic organs in mice/rats [37] and in humans hematopoetic progenitor cells [4]. In this study the rats on exposure of fluoride (F) noted a significant positive correlation between F level and erythrocyte indices.

CONCLUSION

Aluminium fluoride have adverse effects on animals and human health. The study concluded that the administration of AlF_3 to albino rats at a dose of 200mg/kg b. w. daily for a time period of 45 days is capable to inducing haematological (TEC, TLC, Hb. Conc., PCV, MCV, MCH, MCHC and ESR) parameters. The use of *Aloe vera* combined with AlF_3 was discovered to reduce the harmful effects of AlF_3 in the mentioned parameters. Our results also have shown that *Aloe vera* is effective in preventing changes in haematological parameters in albino rats, since *Aloe vera* is protective against haematotoxins, cheap and easily available, it can be used easily in our daily life.

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