

EFFECT OF DIFFERENT DOSES OF ALOE VERA VERSUS INDOMETHACIN ON SODIUM AND WATER RETENTION IN HEALTHY RATS

Abdul Qudoos Arain¹, Mazhar Hussain², Sadia Chiragh³

¹ Department of Pharmacology and Therapeutics, Federal Medical and Dental College, SZABMU-PIMS, Islamabad - Pakistan.

² Department of Pharmacology and Therapeutics, Sheikh Zayed Medical College Rahim Yar Khan - Pakistan.

³ Department of Pharmacology and Therapeutics, Post Graduate Medical Institute, Lahore - Pakistan.

Address for Correspondence:
Dr. Mazhar Hussain

Assistant Professor of Pharmacology, Department of Pharmacology and Therapeutics, Sheikh Zayed Medical College, Rahim Yar Khan - Pakistan.

Email: mazharhussain214@gmail.com

Date Received:

January 17, 2017

Date Revised:

June 18, 2017

Date Accepted:

June 28, 2017

ABSTRACT

Objective: To compare the dose dependent effect of chloroform extract of aloe vera gel on sodium and water retention in rats with indomethacin and control.

Methodology: This randomized controlled trial was conducted at Post Graduate Medical Institute, Lahore in which thirty male Sprague Dawley rats were divided in to five groups. First group was control (CTL), second was given indomethacin 3 mg/kg (INDO) while groups AVL, AVM, and AVH were given 200, 400 and 800 mg/kg doses of Aloe vera gel extract orally for a period of 28 days. Body weight, blood haematocrit level, total sodium excretion and serum sodium and potassium concentrations were measured. SPSS version 17 was used for data analysis.

Results: Comparison of treatment groups with control at end of study showed significant weight gain in group INDO and AVH (p value <0.001). INDO and AVH groups also showed a significant decrease in total sodium excretion (p value 0.001 and 0.015). The mean haematocrit decreased insignificantly in groups INDO and AVH, while serum sodium level increased insignificantly in group INDO. Highest serum potassium level was observed in group INDO (p value 0.001) and the lowest was found in group AVH but difference was not significant as compared to control.

Conclusion: High dose of aloe vera gel extract causes significant sodium and water retention in healthy rats, which was still less than that of indomethacin.

Key Words: Aloe vera, Indomethacin, Sodium retention, Water retention

This article may be cited as: Arain AQ, Hussain M, Chiragh S. Effect of different doses of aloe vera versus indomethacin on sodium and water retention in healthy rats. *J Postgrad Med Inst* 2017; 31(3): 286-91.

INTRODUCTION

Herbal medicine is often regarded as unregulated treatment because plant material contains numerous chemical ingredients, some of which have unidentified actions. So it is difficult to fix the exact dose of herbal medicine for particular ailments. Moreover the plant extract by one method shows different therapeutic and toxic effect when compared to that prepared by another method¹. In spite of all that still 75-80% of the world population especially of developing countries relies on herbal medicine for primary health care and the use of herbal remedies exceeds two to three times more as compared to conventional medicine because of people belief that they are natural, safe, easily available, acceptable, afford able and reliable². Hence the safety and standardization of herbal medicine requires both

pre-clinical and clinical evaluation of toxicity and post marketing pharmacovigilance³.

Non-steroidal anti-inflammatory drugs (NSAIDs) are widely prescribed and used over the counter by a significant number of people because of their success in treating acute and chronic painful inflammatory conditions. However, these medicines are expected to have various side effects which may be serious and life threatening⁴. NSAIDs are associated with a relatively high incidence of dose dependent adverse effects which include renal, gastro-intestinal and cardio-vascular toxicities. NSAIDs are also associated with hyperkalemia, sodium & fluid retention, edema and hypertension^{5,6}. Considering these multiple adverse effects there is need for a trial herbal medicine like aloe vera because of its analgesic and ant-inflammatory properties similar as NSAIDs⁷.

In our previous study we showed that low dose (200mg/kg body weight) of Aloe vera extract did not cause any significant sodium and water retention in healthy rats and was considered to be safe⁸, but one study showed that excess dosage of aloe vera may cause electrolyte imbalance, including low potassium level⁹. Low serum potassium level can lead to cardiac arrhythmias and muscle weakness.

The problem addressed by the present study is whether aloe vera produces a similar effect like that of NSAIDs regarding electrolyte imbalance in the body and to resolve the controversies about aloe vera dose by using aloe vera extract up to a dose of 800mg/kg. This study was done to study the effect of aloe vera when given in higher doses especially on sodium and water retention in the body and comparing with that of indomethacin effect. The results would help in considering the safety of this useful herbal medicine as well as determining the exact dosage that does not cause any serious side effects like that caused by popular NSAIDs. Not just the patients but also the health professionals are going to benefit from the results of this research as there is a need to combine the effectiveness of both complementary as well as traditional allopathic medicine to contribute positively to better healthcare in the future. Since aloe vera is a popular ingredient in many home remedies for inflammation, wound healing and other common ailments, the patients need to be aware of its safety.

METHODOLOGY

Adult healthy male Sprague Dawley rats of 7-8 weeks age, weighing 252-271g were purchased from National Institute of Health (NIH), Islamabad. They were kept at 22°C – 24°C with natural light-dark cycle and given standard rat chow obtained from NIH and mineral water (Nestle) after estimation of sodium concentration¹⁰. After one week period of acclimatization they were divided into five groups each containing six rats. The first group served as control (CTL) was given 0.5ml of distilled water, second group (INDO) was given indomethacin 3mg/kg while third (AVL), fourth group (AVM) and fifth (AVH) were given aloe vera gel extract 200mg/kg, 400 and 800 respectively. Doses were given orally by gavage as a single morning dose.

The chloroform extract of aloe vera gel was prepared as described previously⁸. Gummy crude extract weighing 65 g was obtained from 65 kg of mucilaginous gel. Dose of aloe vera gel extract for each rat was weighed individually and dissolved in 0.5ml of distilled water at time of administration. Indomethacin solution was prepared by dissolving 25 mg of the drug in 8.33 ml of distilled water (3 mg were present in 1 ml). At time of administration it was diluted, such that each rat received 0.5 ml solution.

Each group was labeled and kept in separate cage during a study period of twenty-eight days. Weight of rats was measured every week on digital weighing scale to adjust the dose. On day 0, 14 and 28 urine, feces and blood samples were collected. Urine and feces samples were collected for 24 hour by keeping rats in individual cages. Urine volume was measured, feces were weighed, their sodium concentrations were measured and total sodium excretion calculated. Blood sample was collected by cardiac puncture¹¹ and divided into two portions for estimation of haematocrit by hematology analyzer (NIHON-KOHDEN) and separation of serum for estimation of sodium and potassium concentration by flame photometry. No visible adverse effects of aloe vera were noted in all study groups and tolerability profile of drug was quite good. All rats completed the study and were analyzed in their allocated group. There was no mortality reported in any of the group.

All the grouped data were statistically evaluated with a computer software program (SPSS, Science, Chicago, USA/version 17). Data was expressed as mean \pm SD. Student's t-test (paired) was used for comparison between different times in each group. One-way analysis of variance (ANOVA) was used with post hoc Tukey's test to assess the effect of different doses of aloe vera gel extract and indomethacin on electrolytes and water retention. The level of significance was set at $p < 0.05$.

RESULTS

The results showed increased body weight in all groups over time with p values ≤ 0.01 . At the end of study period, mean body weight was significantly higher in INDO and AVH group as compared to CTL, AVL and AVM with p value 0.001. Group INDO showed the highest weight gain with significant difference from AVH having p value 0.004 (figure 1).

Total sodium excretion increased in CTL group while group INDO showed a significant decrease from day 0 to 14 and from day 0 to 28 with p values 0.036 and 0.001 respectively. In all other groups, there was no significant change recorded over time. Total sodium excretion was significantly low in group INDO at day 14 as compared to CTL and at day 28 as compared to group CTL, AVL and AVM. It was low in group AVH as compared to group CTL at day 28 only (figure 2).

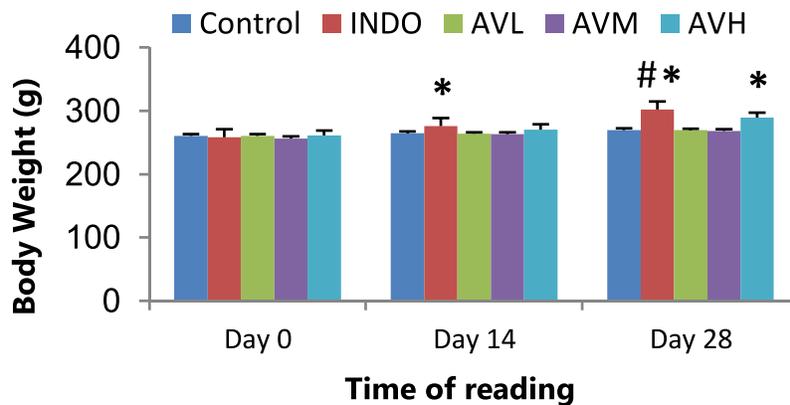
The hematocrit decreased significantly in group INDO from day 0 to 14, from day 0 to 28 and from day 14 to 28 with p values 0.003, < 0.001 and 0.004 respectively. In group AVH, the decrease was significant from day 0 to 14, from day 0 to 28, and from day 14 to 28 with p values 0.026, 0.007 and 0.007 respectively. In all other groups there was no significant change recorded over time in the value of hematocrit. Difference between groups was not significant at end of study (figure 3).

Serum sodium level increased significantly in group INDO from day 0 to 14, from day 0 to 28 and from day 14 to 28 with p values 0.009, 0.004 and 0.015 respectively. There was no significant change recorded over time for other groups. Difference between groups was not significant at end of study (figure 4).

The potassium level increased significantly in group INDO over time from day 0 to 14, from day 0 to 28, and from day 14 to 28 with p values 0.010, <0.001 and

<0.001 respectively. In group AVH, the decrease was significant from day 0 to 28 and from day 14 to 28 with p values 0.001 and 0.045 respectively. There was no significant change recorded over time for all other groups. Group AVH had lowest serum potassium level at day 28 but difference was not significant from group CTL, AVL and AVM. Group INDO had highest serum potassium level at day 28 and difference was significant from group CTL, AVL, AVM and AVH (figure 5).

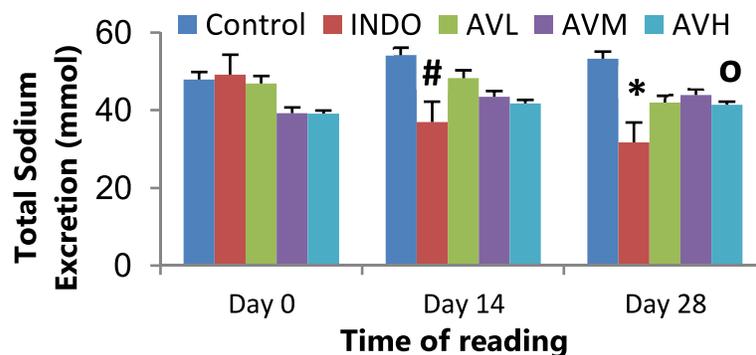
Figure 1: Effect of Aloe vera gel extract on body weight (mean \pm SE) of healthy rats (n=6)



* p-value \leq 0.001 compared to CTL, AVL, AVM;

p-value \leq 0.01 compared to AVH

Figure 2: Effect of Aloe vera gel extract on total sodium excretion (mean \pm SE) of healthy rats (n=6)



* p-value \leq 0.001 compared to CTL; # p-value \leq 0.01

compared to CTL; ^O p-value \leq 0.05 compared to CTL

Figure 3: Effect of Aloe vera gel extract on haematocrit (mean \pm SE) of healthy rats (n=6)

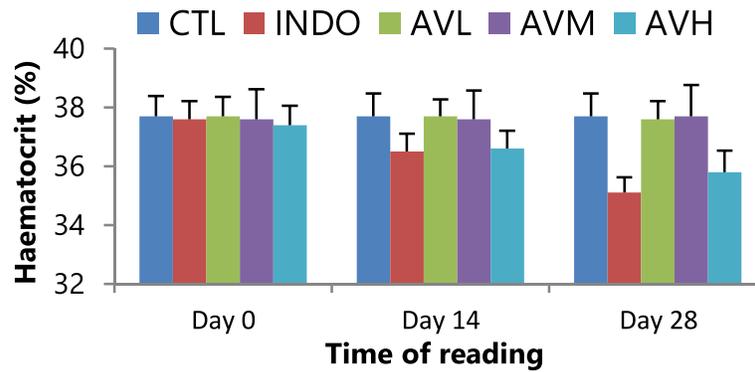


Figure 4: Comparison of serum sodium among all study groups; results are expressed as mean \pm SE (n=6)

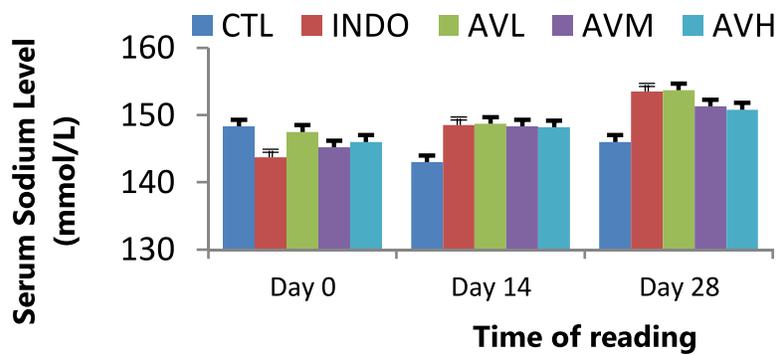
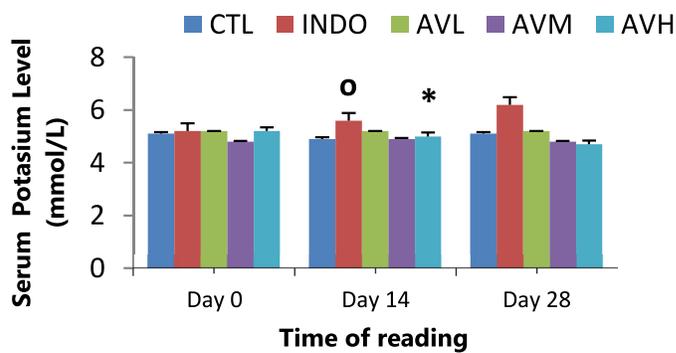


Figure 5: Comparison of serum potassium among all study groups; results are expressed as mean \pm SE (n=6)



^O p-value \leq 0.05 compared to CTL, AVM; ^{*} p-value \leq 0.001 compared to CTL, AVL, AVM, AVH

DISCUSSION

Our previous study using chloroform extract of aloe vera gel in a dose of 200 mg/kg in healthy rats revealed no significant effect on sodium and water retention except slight fall in blood hematocrit and increase in serum sodium concentration⁸, which indicated for further research using higher doses of extract. In this study 200, 400 and 800 mg/kg doses of extract were used. Aloe vera extract has been used in 100-600 mg/kg dose in rat models of different diseases^{7,12,13}. Indomethacin, the most potent inhibitor of prostaglandin synthesis and causing more fluid retention was used for comparison¹⁴.

The results of present study showed significant increase in body weight, decrease in total sodium excretion and insignificant decrease in hematocrit with high (800 mg/kg) dose of chloroform extract of aloe vera gel as compared to control, but less than that of indomethacin. Administration of stabilized aloe vera gel for 14 days did not produce any effect on body weight of male or female mice as compared to controls¹⁵. A study on healthy boys of 8-13 year old, receiving 100 ml of aloe vera juice twice daily for 7 days, demonstrated no effect on urine sodium excretion¹⁶ but significant decrease in serum sodium concentration was exhibited with aloe vera juice in a quantity of 1 ml daily for 28 days in a study on healthy Sprague Dawley rats¹⁷. Mechanism of fluid retention by NSAIDs is by renal COX-2 inhibition¹⁸. Aloe vera gel produces anti-inflammatory activity through multiple mechanisms, COX inhibition being one of them^{19,20}.

Indomethacin-treated group exhibited the highest level of serum potassium concentration at the end of the experiment. This parameter was found to be the lowest in the group that received Aloe vera extract 800 mg/kg but difference from control was not statistically significant. Another study also demonstrated insignificant decrease in serum potassium level with aloe vera juice in rats¹⁷. Hyperkalemia with NSAIDs is due to reduced potassium excretion possibly because inhibition of prostaglandin synthesis results in decreased renin secretion, which in turn reduces the level of aldosterone in blood²¹. Secondly, less sodium is available for exchange with potassium in cortical collecting duct (CCD). On the other hand, aloe vera reduced the level of potassium in blood. One possible explanation may be its aldosterone like action. As aldosterone is responsible for regulating total body potassium homeostasis, an increase in its production causes renal potassium loss through CCD principal cell potassium secretion. In this study high dose of aloe vera extract caused low serum potassium and it also caused some fluid retention and decreased sodium excretion, which resembles the effect of aldosterone. Sterols present in aloe vera may be responsible for this action²².

In summary, aloe vera extract did not cause sodium and water retention when given orally in 200 and 400 mg/kg doses however it had some fluid retaining activity when given in 800 mg/kg dose to healthy rats but still less than that of indomethacin. Effect on serum potassium concentration was insignificant reduction, which is opposite to that of indomethacin. The effect of different doses of aloe vera on heart failure and hypertension has not been studied in the present research, which may be seen as a weakness of the study. Aloe vera may possibly make the condition worse by causing sodium and water retention. For future studies, it is recommended that aloe vera be tried as a therapeutic agent for inflammatory conditions in smaller doses. The safe dose that was established through present study should be used for further research.

CONCLUSION

The results from the present study consider more than 400 mg/kg oral dose of aloe vera extract as being unsafe due to mild sodium and water retention. Administration of aloe vera in hypertensive and high risk patients needs to be further investigated.

REFERENCES

1. Hunter J. Applying evidence based medicine when prescribing herbal products. *OA Alternat Med* 2013; 1:21.
2. Gunjan M, Naing TW, Saini RS, Ahmad AB, Naidu JR, Kumar I. Marketing trends and future prospects of herbal medicine in the treatment of various diseases. *World J Pharma Res* 2015; 4:132-55.
3. Debbie S, Graeme L, Pierre D, Elizabeth W, Kelvin C. Pharmacovigilance of herbal medicine. *J Ethnopharmacol* 2012; 140:513-8.
4. Koffeman AR, Valkhoff VE, Celik S, W't Jong J, Sturkenboom MC, Bindels PJ et al. High risk use of over the counter non steroidal anti inflammatory drugs. *Br J Gen Pract* 2014; 64:e191-8.
5. Harirforoosh S, Asghar W, Jamali F. Adverse effect of non steroidal anti inflammatory drug: an update of gastrointestinal, cardiovascular and renal complications. *J Pharm Pharm Sci* 2013; 16:821-47.
6. Snowden S, Nelson R. The effect of non steroidal anti inflammatory drugs on blood pressure in hypertensive patients. *Cardiol Rev* 2011; 19:184-91.
7. Devaraj A, Karpagam T. Evaluation of anti inflammatory activity and analgesic effect of Aloe Vera leaf extract in rats. *Int Res J Pham* 2011; 2:103-10.
8. Arain AQ, Hussain M, Muneer A, Chiragh S. Effect of chloroform extract of Aloe vera jell on sodium and water retention in rats. *Biomedica* 2016; 32:88-92.
9. Baretta Z, Ghiotto C, Marino D, Jirillo A. Aloe-induced hy-

- pokalemia in a patient with breast cancer during chemotherapy. *Ann Oncol* 2009; 20:1445-6.
10. Horwitz W, Latimer GW. Eds: Official methods of analysis. 18th Ed. Gathersburg AOAC Int; 2005.
 11. Diehl KH, Hull R, Morton D, Pfister R, Rabemampianina Y, Smith D et al. A good practice guide to the administration of substances and removal of blood, including routes and volumes. *J Appl Toxicol* 2001; 21:15-23.
 12. Dutta S, Roy I, Lakshmikanth RN. Effect of Aloe vera supplement on phenytoin induced behavioral abnormalities in rats. *World J Pharm Pharm Sci* 2014; 3:1084-94.
 13. Manjunath K, Bhanu Prakash G, Subash KR, Tadvi NA, Manikanta M, Umamaheswara Rao K. Effect of Aloe vera leaf extract on blood glucose level in alloxan induced diabetic rats. *Natl J Physiol Pharm Pharmacol* 2016; 6:471-4.
 14. Schellack N, Schellack G, Fourie J. A review of nonsteroidal anti-inflammatory drugs. *S Afr Pharm J* 2015; 82:8-18
 15. Sehgal I, Winters WD, Scott M, Kousoulas K. An in vitro and in vivo toxicologic evaluation of a stabilized Aloe vera gel supplement drink in mice. *Food Chem Toxicol* 2013; 55:363-70.
 16. Kirdpon S, Kirdpon W, Airarat W, Thepsuthammarat K, Nanakom S. Changes in the urinary compositions among children after consuming prepared oral doses of Aloe vera (*Aloe vera* linn). *J Med Assoc Thai* 2006; 89:1199-205.
 17. Saka WA, Akhigbe RE, Popoola OT, Oyekunle OS. Changes in serum electrolytes, urea and creatinine in Aloe vera treated rats. *J Young Pharm* 2012; 4:78-81.
 18. Rahman S, Malcoun A. Nonsteroidal antiinflammatory drugs, cyclooxygenase-2, and the kidneys. *Prim Care* 2014; 41:803-21.
 19. Sahu PK, Giri DD, Sing R, Pandey P, Gupta S, Shrivastava AK et al. Therapeutic and medicinal uses of Aloe vera: A review. *Pharmacol Pharm* 2013; 4:599-610.
 20. Gupta VK, Malhotra S. Pharmacological attribute of Aloe vera: Revalidation through experimental and clinical studies. *An Int Quarterly J Res Ayurveda* 2012; 33:193-6.
 21. Krishnan SK, Lepor NE. Acute and chronic cardiovascular effects of hyperkalemia: new insights into prevention and clinical management. *Rev Cardiovasc Med* 2016; 17:S9-21.
 22. Bayati Zadeh J, Moradi Kor N. Component and application aloe vera plant in medicine. *Int J Adv Bio Biomed Res* 2014; 2:1876-82.

CONTRIBUTORS

AQA conceived, designed and did statistical analysis & editing of manuscript. MH and SC Helped in data collection and manuscript writing. SC did review and final approval of manuscript. All authors contributed significantly to the submitted manuscript.