EFFECT OF ALOE VERA (*Aloe barbadensis* Miller) EXTRACT ON KIDNEY FUNCTIONS AND SOME HAEMATOLOGICAL PARAMETERS IN WISTAR RATS

ABSTRACT

The aim of this study was to determine the effect of oral administration of aloe vera extract on kidney functions and haematological parameters in Wistar rats. Twenty adult male albino rats weighing between 150g – 200g were used for the study. They were randomly divided into four groups, namely. Group I (control), II, III and IV were administered 50, 100 and 150mg/kg aloe vera extract respectively for 21 days. Blood samples (2ml), were obtained by cardiac puncture into EDTA bottles to determine; Red blood cells, White blood cells, Haemoglobin, Mean cell volume, Mean cell haemoglobin, Mean cell haemoglobin concentration and Platelets. Another 2ml were collected into plain sample bottles without anticoagulant to determine; Blood urea nitrogen, Creatinine, Total protein and Serum albumin. Data obtained were subjected to analysis of variance using SPSS version 20 and Microsoft 2019 to plot graphs of means. The results obtained revealed that oral administration of aloe vera extract to Wistar rats significantly increased ($p<0.05$) RBC and MCHC in rats. RBC values obtained were $8.09 \pm 0.20 \times 10^{12}$/L in group 2 (50mg/kg aloe vera extract), $7.91 \pm 0.27 \times 10^{12}$/L in group 3 (100mg/kg aloe vera extract) and $7.54 \pm 0.22 \times 10^{12}$/L in group 4 (150mg/kg aloe vera extract). WBC, PCV, MCH, MCV did not vary significantly ($p>0.05$) from those of the control group. In conclusion, oral administration of aloe vera extract up to 150mg/kg body weight did not have any compromising effect on the haematological and Kidney function indices in Wistar rats.

Keywords: Aloe vera, renal function, haematology, Wistar rats.

INTRODUCTION

Aloe species are increasingly being incorporated into different cosmetic products, health drinks, foods, and beverages due to the abovementioned beneficial biological activities of the phytochemicals found mainly in the leaves. Polysaccharides, flavonoids, carbohydrate, coumarins, tannins, chromones, alkaloids, anthraquinones, organic compounds, pyrones, phytosterols, anthrones, sterols, vitamins, proteins, and mineral components are the phytochemicals found in the Aloe plant (Banik and Sharangi, 2019). Some of these phytochemicals, however useful, have been linked to harmful effects, many researchers have discovered potential toxicity and risks associated with many plants and vegetables, including hepatotoxicity, nephrotoxicity, and cancer. (Haq, 2004). Haematology parameters such as red blood cell count, haemoglobin concentration and derived haematology indices, e.g Mean cell volume (MCV), Mean cell haemoglobin (MCH), Mean cell haemoglobin concentration (MCHC) are important blood marker for diagnosis of various blood diseases (Channa *et al*., 2014). The major objective of this study is to evaluate the effect of aqueous aloe vera extract on kidney functions and haematological parameters in male Wistar rats.

MATERIALS AND METHODS

Animals and Management

A total of twenty (20) male Wistar rats weighing 150g-200g was used for this study. The Wistar rats were purchased from the

small animal unit of National Veterinary Research Institute (NVRI), Vom, Plateau State. Upon arrival, the animals were kept in cages in the Animal Care Unit, Department of Physiology, Faculty of Basic Medical Sciences, Bingham University, karu. The animals were fed with pelletized grower's mash, with wheat flour as binder and water *ad libitum*. The Wistar rats were allowed to acclimatize to the laboratory environment for the period of one week before commencement of the experiment.

Plant Collection, Storage and Extraction

Fresh Aloe vera leaves was purchased from Sarius Palmetum and Botanical Garden, Maitama, Abuja FCT, Nigeria. The leaves were thoroughly washed in clean water and air-dried in the laboratory. The dried material was pulverised into a meal using a mortar and pestle. A known quantity of the aloe vera meal was extracted with distilled water. The powdered leave was weighed and poured into a 2000ml conical flask in which 1000ml of distilled water was added. The mixture was kept for 12 hours with constant agitation at 30 minutes interval. The extract was filtered out using Whatman No.1 filter paper. The filtrate was concentrated in *vaccuo.* The semi-solid extract obtained was stored in a refrigerator for further use.

Authentication

Authentication of aloe vera was carried out with the botanical name *Aloe barbadensis Miller* from the family of Liliaceae with specimen number 28563

Experimental Design

Animal Grouping

The male Wistar rats were randomly grouped into four (4) groups of five (5) rats each as follows:

Group I: This served as a control group with a total number of five rats (n = 5). The rats were fed with pelletized grower's mash and water only for three weeks.

Group II: Group II had a total number of five (n= 5) Wistar rats. The rats were fed with pelletized grower's mash, water and 50mg/Kg body weight of aloe vera crude extract for three weeks.

Group III: There were a total number of five (n= 5) Wistar rats. The rats were fed with pelletized grower's mash, water and 100mg/Kg body weight of aloe vera crude extract for three weeks.

Group IV: This was the last experimental group with a total number of five (n= 5) Wistar rats. The rats in this group was fed with pelletized grower's mash, water and 150mg/Kg body weight of aloe vera crude extract for three weeks.

Sample Collection and Preparation

All the animals were sacrificed at the end of the experiment. The rats were anaesthetized at the time of sacrifice by being placed in a sealed cotton wool-soaked chloroform inhalation jar. Blood sample was collected directly from the heart via cardiac puncture from each Wistar rat and the whole blood was collected into both EDTA and plain bottles. The sample in plain bottles was centrifuged and the serum was used to determine: total protein, serum albumin, blood urea nitrogen and creatinine respectively. While the sample collected in EDTA bottles was used to determine the various haematological parameters: RBC, WBC, Platelets, Hb, MCHC, MCH, Differential WBC and PCV.

Statistical Analysis

Data obtained were analyzed using One Way Analysis of Variance (ANOVA) using SPSS version 20 was and the results obtained were presented as mean ± standard error of mean (SEM) and an appropriate post hoc test of multiple comparison was used to determine

the statistical significance, p values (P< 0.05) were considered statistically significant.

RESULTS AND DISCUSSION

Haematological parameters of Wistar albino rats administered varying levels of Aloe vera extract orally

Haematology refers to the study of the numbers and morphology of the cellular elements of the blood and the use of these results in the diagnosis and monitoring of disease conditions in both human and animals (Merck Manual, 2012). Results revealed that all haematological indices evaluated were not statistically significant (p>0.05) at the various levels of aloe vera extract administration in the study except, red blood cells (RBC) and mean cell haemoglobin concentration (MCHC). Packed cell volume (PCV) of the rats though not significantly influenced (p>0.05) by Aloe vera extract administration, were higher than those of the control group. PCV was highest (39.36 ± 1.00%) in group 2 (50mg/kg) and decreased progressively non-significantly (p>0.05) through group 4 (150mg/kg). Packed Cell Volume (PCV) which is also known as haematocrit or erythrocyte volume fraction (EVF), is the percentage (%) of red blood cells in blood (Purves *et al.*, 2003). Isaac *et al*. (2013) stated that PCV is involved in the transportation of oxygen and absorbed nutrients to target tissues. Consequently, increased PCV shows a better transportation and thus results in an increased primary and secondary polycythemia. Rats in groups 3 (100mg/kg) and 4 (150mg/kg) had PCV values of 38.25 ± 2.00% and 36.70 ± 1.20% respectively, while rats in the control group without aloe vera extract recorded the least PCV value of 31.30 ± 4.67%. PCV values obtained in this study were below the range values of 41.75 ± 2.17% – 42 ± 4.32% reported by Ekanade *et al*. (2015) for Wistar rats administered aqueous extract of Aloe vera. The values are however within the range of 18-48 and 10-47% recommended by Delwatta *et al*. (2018), for male and female rats respectively. This result however disagrees with that of Ekanade *et al.* (2015) who reported increase in the PCV of rats administered with the extract of A. vera. This implies that the different doses of aloe vera extract administered did not impact the PCV of the rats negatively in the course of the study. Since PCV is involved in the transport of oxygen and absorbed nutrients (Isaac *et al.* 2013), the rats hence, did not suffer from nutritional inadequacy and oxygen deprivation.

Table 1: Haematological parameters of Wistar albino rats administered varying levels of Aloe vera extract orally

PARAMETERS	TREATMENT GROUPS			
	GROUP I (Control)	GROUP II	GROUP III	GROUP IV

		(50mg/kg aloe vera extract)	(100mg/kg aloe vera extract)	(150mg/kg aloe vera extract)
Packed Cell Volume (%)	31.30 ± 4.67	39.36 ± 1.00	38.25 ± 2.00	36.70 ± 1.20
White Blood Cell (x $10^{9/}$L)	4.05 ± 0.76	5.84 ± 0.58	6.28 ± 1.34	4.58 ± 0.54
Red Blood Cell (x $10^{12/}$L)	6.44 ± 0.85	8.09 ± 0.20	7.91 ± 0.27	7.54 ± 0.22
Haemoglobin (g/dL)	10.70 ± 15.72	13.68 ± 3.68	13.60 ± 6.10	12.85 ± 4.21
Mean Corpuscular Volume (fL)	48.40 ± 1.32	48.73 ± 0.09	48.76 ± 0.59	48.73 ± 0.33
Mean Corpuscular Haemoglobin (Pg)	16.48 ± 0.46	16.80 ± 0.50	17.10 ± 0.19	17.00 ± 0.15
Mean Cell Haemoglobin Concentration (g/dL)	341.75 ± 1.93	344.50 ± 0.50	355.38 ± 2.88*	349.75 ± 1.93
Platelet (x10^9)	400.25 ± 8.73	286.25 ± 7.09	425.88 ± 13.88	423.00 ± 58.19

Values are represented as mean ± standard error of mean.

Red blood cells (RBC) of the rats increased ($p<0.05$) with oral administration of aloe vera extract, with the highest value been observed in treatment group 2 (50mg/kg), followed closely by rats in group 3 (100mg/kg) and 4 (150mg/kg) respectively. RBC values observed in this study were similar in treatment groups 2 (8.09 ± 0.20 x 10^{12}/L), 3 (7.91 ± 0.27 x 10^{12}/L) and 4 (7.54 ± 0.22 x 10^{12}/L) and differed ($p<0.05$) from those of the control group that had mean RBC value of 6.44 ± 0.85 x 10^{12}/L. This result agrees with the report of Ekanade *et al*. (2015) who also observed significant differences in RBC and haemoglobin and, is within the range of 6–8 g/dL as reported by Wikivet (2012). Archibong *et al.* (2018) also reported significant increase in RBC of rats administered aloe vera gel. Etim *et al*. (2013) earlier suggested that red blood cells serve as a carrier of haemoglobin. According to Isaac et al. (2013) red blood cell is involved in the transport of oxygen and carbon dioxide in the body. Thus, a reduced red blood cell count implies a reduction in the level of oxygen that would be carried to the tissues as well as the level of carbon dioxide returned to the lungs (Etim *et al*., 2013). The higher RBC values observed in this study indicates that aloe extract did not affect the oxygen carrying capacity of the blood.

Mean white blood cells of the Wistar rats administered aloe vera extract in this study did not vary ($p>0.05$) from those of the control group, hence, had similar values of 4.05 ± 0.76, 5.84 ± 0.58, 6.28 ± 1.34 and 4.58 ± 0.54 x 10^9/L) for treatment groups 1, 2, 3 and 4 respectively. The major functions of the white blood cell and its differentials are to fight infections, defend the body by phagocytosis against invasion by foreign organisms and to produce or at least transport and distribute antibodies in immune response (Etim *et al.*, 2013). Consequently, animals with low white blood cells are exposed to high risk of disease infection, while those with high counts are capable of generating antibodies in the process of phagocytosis and

have high degree of resistance to diseases (Soetan *et al*., 2013). The values of WBC are within normal ranges of 4,400–14,800 and 3,600–14,500 per/mm3 for male and female rats reported by Dewatta *et al.* (2018), therefore, the immune system of the rats was not compromised by doses of aloe vera extract administration.

There was no significant difference ($p>0.05$) in the levels of haemoglobin in the blood of the rats in this study, however, higher numerical values were observed in rats administered aloe vera extract than those without aloe vera extract administration. Haemoglobin, although statistically similar among the different treatment groups, increased non-significantly ($p>0.05$) with aloe vera extract administration with the highest value (13.60 ± 6.10 g/dL) been observed in group 3 while the lowest haemoglobin value (10.70 ± 15.72 g/dL) was observed in rats in the control group. Haemoglobin of rats in this study are within the normal range of 13.70 – 17.6 g/dL recommended by Giknis and Clifford (2008) and 13.88±0.89 - 16.31±0.68 g/dL by Ekanade *et al.* (2015) *for* Wistar rats. Haemoglobin is the iron-containing oxygen-transport metalloprotein in the red blood cells of all vertebrates (Maton *et al*., 1993) with the exception of the fish family, channichthyldae (Sidell and O' Brien, 2006) as well as tissues of invertebrates. The physiological function of haemoglobin is to transport oxygen to tissues of the animal for oxidation of ingested food so as to release energy for the other body functions as well as transport carbon dioxide out of the body of animals (Etim *et al*., 2013)

Mean cell volume (MCV) of Wistar rats was not statistically influenced ($p>0.05$) by oral administration of aloe vera extract in the current study. MCV values were 48.40 ± 1.32, 48.73 ± 0.09, 48.76 ± 0.59 and 48.73 ± 0.33 fL for treatment groups 1, 2, 3 and 4 respectively. MCV values were similar to those of Ekande *et al.* (2015) for rats administered aqueous extract of Aloe vera. Delwatta *et al.* (2018) recommended MCV range of 29.41-123.07 fL and 15.15-119.44 fL for male and female rats respectively. Ofem *et al.* (2015) also observed non-significant difference in MCV of rats administered aloe vera gel. The statistical analysis of mean MCH values of treatments and control group indicated no significant difference ($p>0.05$) in this study. The values obtained ranged between 16.48 ± 0.46 – 17.10 ± 0.19 Pg. Mean cell haemoglobin concentration (MCHC) of Wistar rats administered aloe vera extract varied significantly ($p<0.05$) and was highest (355.38 ± 2.88 g/L) in treatment group 3 (100mg/kg) and lowest (341.75 ± 1.93 g/L) in control group (without aloe vera extract). Treatment groups 2 (50mg/kg) and 4 (150mg/kg) had MCHC values of 344.50 ± 0.50 g/L and 349.75 ± 1.93 g/L respectively. MCHC of rats in this study were within the normal range of 33.20 – 37.9g/L recommended by Giknis and Clifford (2008). Delwatta *et al.* (2018) also suggested MCHC of 25.41-80.55 and 21.16-95.00g/L for male and female rats respectively.

Peters *et al.* (2011), previously stated that PCV, haemoglobin and MCH are major indices for evaluating circulatory RBCs, and are significant in the diagnosis of anaemia and also serve as useful indices of the bone marrow capacity to produce red blood cells in mammals. Channa *et al.* (2014), also opined that MCV, MCH and MCHC are important blood markers for diagnosis of various blood diseases. They also added that 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. Again, Chineke *et al.* (2006) also posited that high PCV reading indicates either an increase in number of Red Blood Cells (RBCs) or reduction in circulating plasma volume while MCH and MCHC indicate blood level conditions. A low level is an indication of anaemia (Aster, 2004).

Blood platelets are implicated in blood clotting. Low platelet concentration suggests that the process of clot-formation (blood clotting) will be prolonged resulting in excessive loss of blood in the case of injury (Etim *et al*., 2013). Platelets value did not show significant variation ($p>0.05$) with oral administration of aloe vera extract to Wistar rats in the current study. Platelets value observed in the study were 400.25 ± 98.73, 286.25 ± 7.09, 425.88 ± 13.88 x and 423.00 ± 58.19 x 10^9/L for treatment groups 1, 2, 3 and 4 respectively.

Previous investigations found that A. vera extract improved immune cells and complement system, active anti-bacterial agent and, improves ventricles key component of producing cerebrospinal fluid (Shahid, 2014). The non-significant differences observed in most of the haematological parameters suggest that aloe vera extract at the various doses did not compromise the health of the rats in the study.

Biochemical indices of Wistar albino rats administered varying levels of Aloe vera extract orally

There were no significant variations ($p>0.05$) in serum biochemical indices of Wistar rats administered oral doses of aloe vera extract except creatinine. The blood urea level was not statistically influenced ($p>0.05$) but were increased non-significantly with aloe vera extract administration to rats compared to the control. Values observed were 8.48 ± 0.72, 9.99 ± 0.43, 11.35 ± 0.93 and 10.66 ±1.00 mmol/L for treatment groups 1 (control), 2 (50mg/kg), 3 (100mg/kg) and 4 (150mg/kg) respectively. Treatment group 3 (100mg/kg) had highest non-significant value of urea, followed by rats in group 4 (150mg/kg). The mean blood urea levels in this study were below the range of 17.26-45.00 and 12.33-77.6 mmol/L recommended by Delwatta *et al.* (2018) for male and female rats respectively and 12.30 – 24.60 mmol/L recommended by Giknis and Clifford (2008). This hence, suggests that the kidneys of the rats were negatively impacted in the course of the study. However, a non-significant improvement was observed with aloe vera extract administration.

Creatinine decreased significantly ($p<0.05$) in rats with aloe vera extract administration compared to the control group. Rats in groups 2 (50mg/kg) and 4 (150mg/kg) had similar creatinine values of 0.20 ± 0.00 g/dL while rats in group 1 (control) and 3 (100mg/kg) had close values of 0.28 ± 0.03 mg/dL and 0.25 ± 0.03 mg/dL respectively. Creatinine levels of the rats were within the value range of 0.20 – 0.60 suggested by Giknis and Clifford (2008). This implies minimal damage to the kidney since urea and creatinine are important markers of kidney function

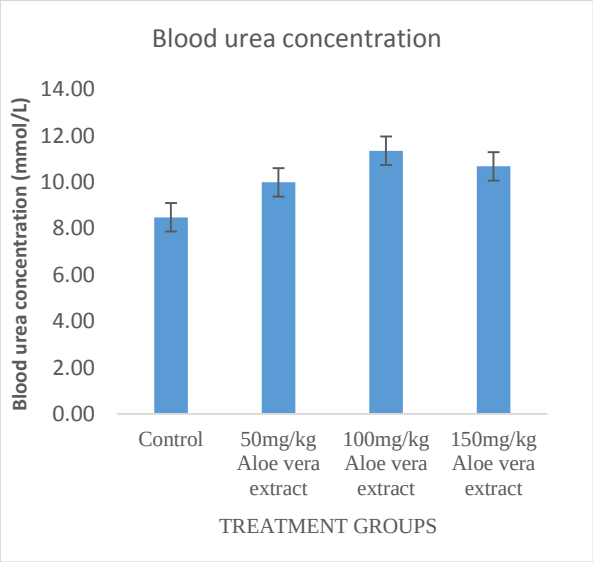

Figure 1. Effect of aloe vera extract on blood urea concentration in Wistar

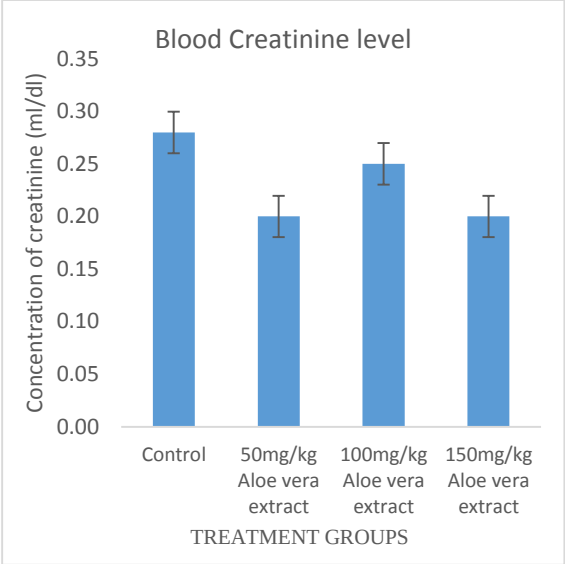

Figure 2. Effect of aloe vera extract on creatnine concentration in Wistar rats

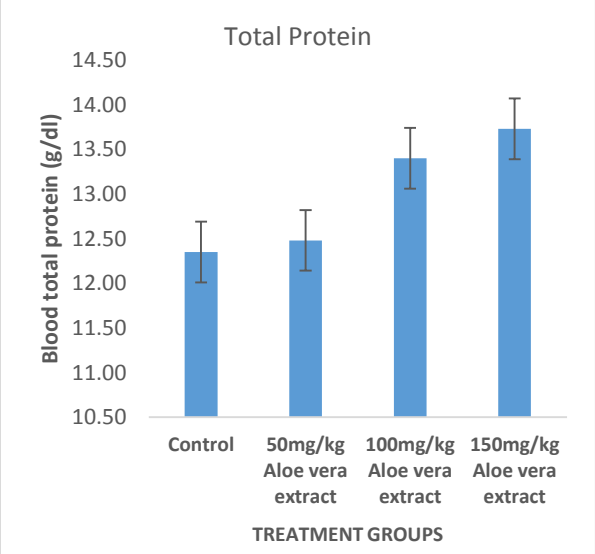

Figure 3. Effect of aloe vera extract on total protein concentration in Wistar

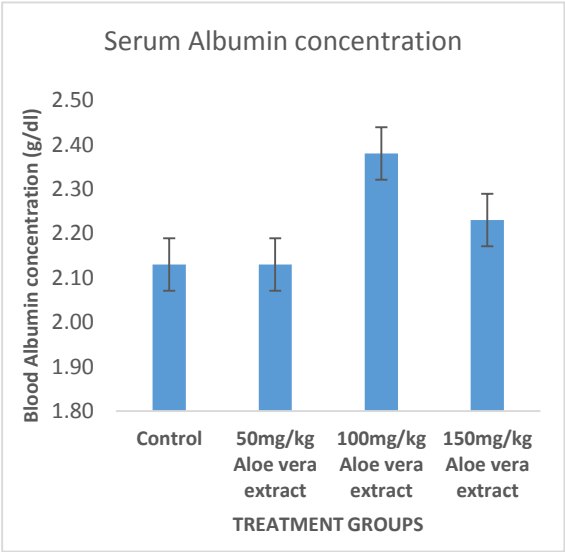

Figure 4. Effect of aloe vera extract on albumin concentration in Wistar rats

Total protein and albumin were not statistically influenced ($p>0.05$) in this study. Total protein values in the study were 12.35 ± 0.78, 12.48 ± 0.42, 13.40 ± 0.51 and 13. 73 ± 0.88 g/dL for treatment groups 1 (control), 2 (50mg/kg), 3 (100mg/kg) and 4 (150mg/kg) respectively while albumin values ranged from 2.13 ± 0.05 g/dL in the control group to 2.38 ± 0.03 g/dL in group 3 (100mg/kg). Groups 2 (50mg/kg) and 4 (150mg/kg) had values of 2.13 ± 0.13 g/dL and 2.23 ± 0.10 g/dL respectively. Shahid (2014) reported significant difference in total protein and albumin at higher doses of aloe vera extract

administration, above values in the current study. He also observed that aloe vera extract at higher dose of 200 – 400mg/kg causes gradual changes in the means of total protein, albumin, globulin, urea, and creatinine in different groups. Serum total protein is a marker of the synthetic function of the liver and a valuable guide to assess the severity of liver damage (Osigwe *et al.* (2017). Serum total protein in the current study were above the normal range of 5.2 – 7.1 g/dL suggested by Giknis and Clifford (2008) and the normal ranges of 5.6–7.6 and 3.8–4.8 g/dL reported by Wikivet (2012) and 5.2–7.1 and 3.4–4.8g/dL reported by Mary and Charles (2008). Low or high total protein is an indication of liver disorders and malnutrition (Augustine *et al.*, 2020), hence the results in this study based on the total protein values obtained, indicates adverse effects on the liver of the rats. This damage may be unconnected to doses of aloe vera extract administration since the control group had similar elevated serum total protein. It is therefore suggested that this observed possible liver damage may be nutritional rather than treatment induced.

CONCLUSION

The study showed that the oral doses of aloe vera extract administered to Wistar rats in the study improved the haematological indices and had no compromising effect on liver and kidney functions in this study. This is evident by the non-significant differences observed in key kidney and liver function parameters. It is therefore concluded that, oral ingestion of aloe vera extract up to 150mg/kg can improve the haematological indices in Wistar rats and has no adverse effect on kidney function markers in rats. However, prolong ingestion is discouraged as they may cause possible increase in liver and kidney function markers.

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