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Research Article

Comparative Investigation Of Antidiabetic Effect Of Aquous Extract Of Murraya Koenigii, Aloe Vera And Their Combination In Alloxan Induced Albino Rats

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ABSTRACT

Diabetes mellitus is a metabolic disease characterized by altered cellular metabolism. A number of folk medicine are being used by diabetic patients to control diabetes. This experiment was performed to compare the Antidiabetic effect of aquous extract of Murraya koenigii, Aloe vera and their combination with reference to Metformin (150 mg/kg body weight) as a standard drug in alloxan induced diabetic male albino rats. In addition to that the experiment was also conducted to measure biochemical parameters such as serum creatinine, albumin and urea, serum glutamate oxaloacetate and pyruvate transaminase (SGOT and SGPT) and also to the change in body weight after administration of these extracts. After 28 days during the treatment with these extracts following observations were made. M. koenigii when administered orally 300 mg/kg body weight significantly ($p>0.05$) decreased blood glucose level (109.2 ± 6.14), Aloe vera when administered orally 300 mg/kg body weight significantly ($p>0.05$) decreased blood glucose level (112.2 ± 7.83) and combination of these two when administered orally 300 mg/kg body weight significantly ($P>0.05$) decreased blood glucose level (101.8 ± 3.86) which showed synergistic effect compared to reference drug Metformin when administered orally 150 mg/kg body weight significantly ($p>0.05$) decreased blood glucose level (86.6 ± 4.27). From this study it was concluded that Curry leaves (M. koenigii) and Aloe vera is a potent Antidiabetic drugs and the combination of these two drugs showed synergistic effect in lowering the blood glucose level as Metformin. The administration of diabetic rats with plant extracts for 28 days periods showed a remarkable alteration in lowered blood glucose levels that are close to normal values reported in control group (88.8 ± 3.76) For all the parameters it was observed that the aq. extracts of MK, AV and their combination have diminishing effect on severity of diabetes.

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INTRODUCTION

A number of herbal plants are used in many countries to treat and control diabetes mellitus. The hypoglycemic action of these herbal plants is being investigated.[1] Diabetes mellitus is a very common chronic metabolic disease where there is a gradual deterioration of organs in the body and is associated with multi organ complications. Alloxan is a diabetogenic agent used in diabetes research to induce insulin reduction.[2] The number of diabetic patients in the world had been estimated as 150 million. This number is predicted to double of its number by 2025.[3] The picture in India is much more alarming. The current estimation shows that there are 6.2 crore people with diabetes and this number is likely to raise upto 8.5 crore by 2030. India has now been declared by WHO as the 'Diabetes capital of the world'. [4] A wide research work on diabetes leads to a number of synthetic oral Antidiabetic (hypoglycemic) agents like thiazolidinedione's, biguanides, and sulphonylureas being used to treat and control diabetes mellitus. But all synthetic oral hypoglycemic drugs have serious side effects associated with their long term uses. On the other hand, traditional herbal plants along with their various biological constituents have been used effectively by the communities since long time to treat and maintain high blood glucose level. Several natural products such as polysaccharides, glycosides alkaloids, saponins, terpenoids, Flavonoids are isolated from these medicinal plants and are being reported to have or possess anti-diabetic activities. In addition, herbal drugs are extensively used for the various diseases treatment due to their effectiveness, minimal side effects and relatively low cost. From the very ancient times, some of these herbal medicinal plants preparations have been used in the treatment of diabetes mellitus. Therefore in the treatment of diabetes so many herbal folk medicines were used. The active compounds of these folk medicine plays an important role in the diabetes mellitus management. Alloxan (2,4,5,6 tetraoxypyrimidine) is an oxygenated derivative of pyrimidine & a cyclic urea analog which was reported to produce permanent diabetes in experimental animals such as rabbits, rats. It is a well Known diabetogenic agent that is used to induce type- 2 diabetes in experimental animals. [5, 6] One of the most potent methods to induce experimental diabetes is chemical induction by Alloxan. [7] Murraya koenigii is belongs to Family Rutaceae and is commonly known as "Curry Patta" in Hindi & is widely used as a condiment and spice in India & other tropical countries and is a native plant of India, Sri Lanka & other south Asian countries as well. It is found usually almost everywhere in the Indian subcontinent. Various parts of this plant have been used in folk medicine or traditional medicine for the treatment of traumatic injury, rheumatism, and snake bite and it has been reported to have antioxidant, anti-diabetic, & anti-dysenteric activities. Curry leaf is traditionally used as, anti-dysenteric and a stimulant for the diabetes mellitus management.[8] Aloe vera is a member of the Liliaceae family, which has about 360 species. Aloe vera is a cactus-like plant that grows in hot, dry climate[9] Aloe vera has been used for medicinal purposes in several cultures like Greece, Egypt, India, Mexico, Japan, and China since ancient time.[10] The pharmacological actions of Aloe vera, as evidenced by in-vitro and animal studies, include anti-inflammatory and anti-arthritic activity, and antibacterial and hypoglycemic effects. The therapeutic claims made for Aloe vera range over a broad list, as do the pharmacological activities associated with it.[11] The primary aim of this study is to investigate the anti-hyperglycemic effects of aqueous extracts of Murraya koenigii (curry leaves) and aloe vera juice, both individually and in combination. The study also aims to compare these effects with those of

Metformin, a standard anti-diabetic medication, in alloxan-induced diabetic rats. Additionally, the study will evaluate the impact of these treatments on liver functions (assessed by SGOT and SGPT levels), renal functions (assessed by creatinine, serum albumin, and urea levels), and body weight changes.

MATERIAL AND METHODS

The study was conducted at the Institute of Pharmacy, Bundelkhand University, Jhansi, to investigate the comparative efficacy of aqueous extracts of Murraya koenigii leaves, Aloe vera juice, and their combination, with Metformin serving as the standard drug. The research was performed on alloxan-induced diabetic male albino rats to evaluate anti-hyperglycemic activity and the effects on liver and renal functions, as well as body weight changes. The following procedure were adopted for conducting this study.

Drug, Chemicals and Reagents

Alloxan monohydrate (chemdyes corporation, Rajkot, Gujrat) Metformin, Accu check active Glucometer and strips (Suman Medicals, Jhansi) All other chemicals were provided by the central store, Institute of pharmacy, Bundelkhand University Jhansi.

Collection and authentification of Plant Materials

The fresh leaves of Aloe vera and Curry neem were collected from local region of Bundelkhand, Jhansi Uttar Pradesh, India, and authenticated at Botanical department Ayurvedic College Jhansi, Uttar Pradesh, India. The voucher specimen was deposited in the departmental herbarium for future reference.

Preparation of plant extracts

The fresh leaves of Aloe vera were collected, washed with distilled water and shadow dried. The shadow dried leaves of aloe vera were subjected to pulverization to get coarse powder. Aqueous extract was made by dissolving it in distilled water using by mortar and pestle. The dose was initially made to 300 mg/kg body weight for oral administration.[12] Murraya koenigii leaves aqueous extract was prepared by maceration method. About 200 g of leaf powder was subjected to cold maceration with chloroform : water in a conical flask for 7 days at room temperature. The flask was plugged with absorbent cotton at the mouth of flask and shaken periodically. It was filtered through a muslin cloth and the collected filtrate was refiltered through Whatmann filter paper to get the clear filtrate. The filtrate was concentrated to dry residue by shade drying it for 30 days. This extract was labeled as MKAE and the selected dose was 300 mg/kg body weight for oral administration.[13]

Preparation of Metformin

Metformin solution was prepared by dissolving 150 mg of Metformin pure powder in 10 ml of distilled water to attain a concentration of 15 mg/ml, labeled as MET and its dose was selected as 150 mg/kg body weight.[14]

Animals

Adult healthy male albino rats (Wistar Strain) weighing 100 – 200g were selected. They were kept at departmental animal house in standard polypropylene cages and maintained under controlled room temperature (22±20C) and humidity (55±5%). All the animals were provided with commercially available rat normal pallet diet and water ad-libitum. Approval for the study protocol was granted by the Institutional Animal Ethical Committee of Institute of pharmacy, Bundelkhand University, Jhansi, Uttar Pradesh, India (Reg. No. 716/GO/Re/S/02/CPCSEA).

Phytochemical Screening

The individual extract was subjected to qualitative phytochemical screening for the presence of some chemical constituents such as steroids, Phenolics, fixed oil, alkaloid, glycoside, Saponin, Flavonoids, tannins and carbohydrate.

Induction of Diabetes

Overnight fasted albino rats will be made diabetic by injecting alloxan monohydrate (in ice cold normal saline) intraperitoneally at a dose of 120 mg/kg weight. After that the rats will be keep a side for 4 hrs and then 10% glucose solution will be placed in the cages for 24 hrs. Measure the FBS concentration after 72 hr of alloxanization. Animals with blood glucose level above 250 mg/dl will be considered to be diabetic and were used in the study. [15, 16, 17]

Experimental Work

Animals will be maintained in clean polypropylene cages with 12 hr light & 12 hr dark cycle at temp of 27-29 0C & relative humidity of 60 ± 5. They will be given standard pellet diet and water ad-libitum throughout the course of the study. The study will be carried out in accordance with the guidance by committee constituted for the purpose of experiment animals.

Grouping

30 male albino Wistar rats were randomly divided into six groups (n=5 per group)

Group I (Normal Control):

Rats without any treatment.

Group II (Diabetic Control):

Alloxan-induced diabetic rats without any treatment.

Group III (Metformin Treatment):

Diabetic rats treated with Metformin 150 mg/kg.

Group IV (Murraya koenigii Treatment):

Diabetic rats treated with aqueous extract of Murraya koenigii 300 mg/kg.

Group V (Aloe vera Treatment):

Diabetic rats treated with aqueous extract of Aloe vera 300 mg/kg.

Group VI (MK + AV):

Diabetic rats treated with a combination of Murraya koenigii extract and Aloe vera juice 300 mg/kg.

The blood glucose concentration of the animals will be measured at the beginning of the study and measurements will be repeated on 3rd ,7th,11th, 15th, 19th, 23th and 28th day of experiment. Change in body weight will be observed by measuring weekly changes in body weight. At the end of the experimental period, the animals were fasted overnight and then sacrificed by cervical decapitation. Blood was collected in tubes containing EDTA for the estimation of creatinine, albumin and urea as biochemical markers of kidney functioning [18-19] and determination of SGOT and SGPT as biochemical markers of liver functioning. [20-21]

Statistical Analysis

All the data are expressed as Mean ± SD. The anti-diabetic potential was analyzed by one-way analysis of variance (ANOVA). A P value of < 0.05 was considered as statistically significant.

RESULTS

The experiment was carried out for the study of comparative efficacy of herbal drug preparation of Curry neem leaves extract, Aloe vera juice and its combination with a standard drug Metformin as blood glucose lowering agent in rats. Attempts were also made to investigate the effects of these herbal preparation and their combination and reference drug on creatinine, albumin and urea as biochemical markers of kidney functioning, determination of SGOT and SGPT as biochemical markers of liver functioning and effect on body weights in rats. To perform the experiment 30 Rats (albino Wistar male rats) were randomly divide into six groups. Group I was kept as normal control without giving any treatment Group II as a diabetic control, Group III were treated with Metformin, Group IV were treated with aq. Extract of Murraya koenigii, Group V were treated with aqueous extract of Aloe vera and Group VI were treated with combination of MK and AV. All the control and treated were closely observed during 28 days of treatment.

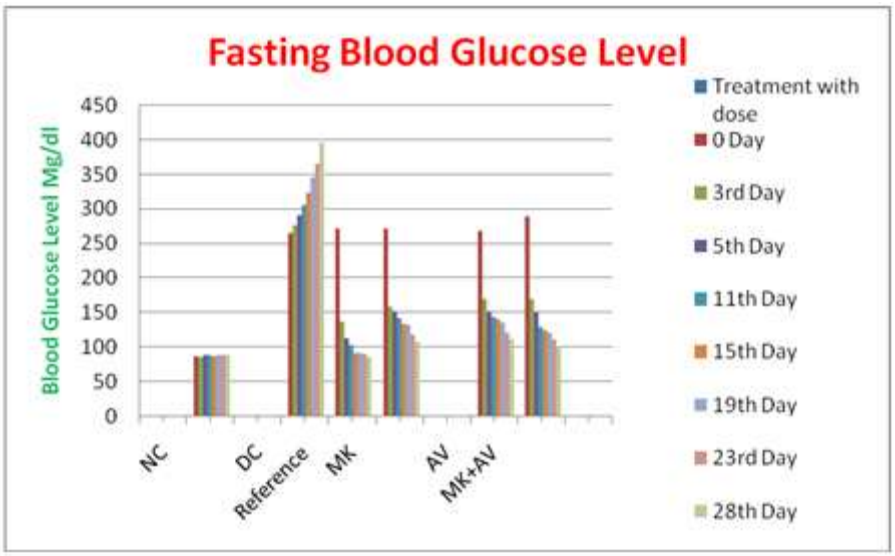
Effect on blood glucose level

The aq. extracts of MK, AV and combination of MK+AV showed significant Antidiabetic activity in lowering blood glucose level. After 28 days of treatment the effectiveness of extracts was found to be in order of MK+AV > MK > AVJ. The results are shown in table 1.

Table 1: Effect of Metformin, Curry neem aq. extract, Aloe vera Aq. Extract and combination of (MK + AV) on blood glucose level (mg/dl) in rats (n=5)

Groups	Treatment with dose	Day 0	Day 3	Day 7	Day 11	Day 15	Day 19	Day 23	Day 28
I	**NC**	87.4 ± 5.98	86.4 ± 3.97	88.6 ± 5.46	89.8 ± 3.31	87.2 ± 3.54	89.2 ± 4.06	89.8 ± 5.03	88.8 ± 3.76
II	**DC**	266.2 ± 8.58	276.4 ± 9.47	291.6 ± 7.76	305.6 ± 7.86	324.2 ± 10.28	346.2 ± 8.20	365.8 ± 15.54	396.0 ± 15.33
III	**with Metformin**	272.4 ± 12.04	137.8 ± 11.21	113.8 ± 4.66	104.2 ± 5.77	92.8 ± 6.33	92.2 ± 3.37	90.4 ± 3.00	86.6 ± 4.27
IV	**MK**	271.6 ± 7.28	158.6 ± 8.59	152.0 ± 9.59	142.4 ± 11.94	134.4 ± 8.30	132.0 ± 3.74	118.6 ± 10..55	109.2 ± 6.14
V	**AV**	268.2 ± 11.28	170.2 ± 8.70	152.8 ± 9.82	143.4 ± 9.97	141.2 ± 10.79	135.2 ± 8.32	121.6 ± 6.74	112.2 ± 7.83
VI	**MK+AV**	289.8 ± 18.11	169.8 ± 8.63	150.6 ± 7.41	130.2 ± 7.75	125.0 ± 4.81	122.2 ± 7.13	112.4 ± 4.84	101.8 ± 3.86

n= 5, values express as Mean ± SD, significance P<0.05



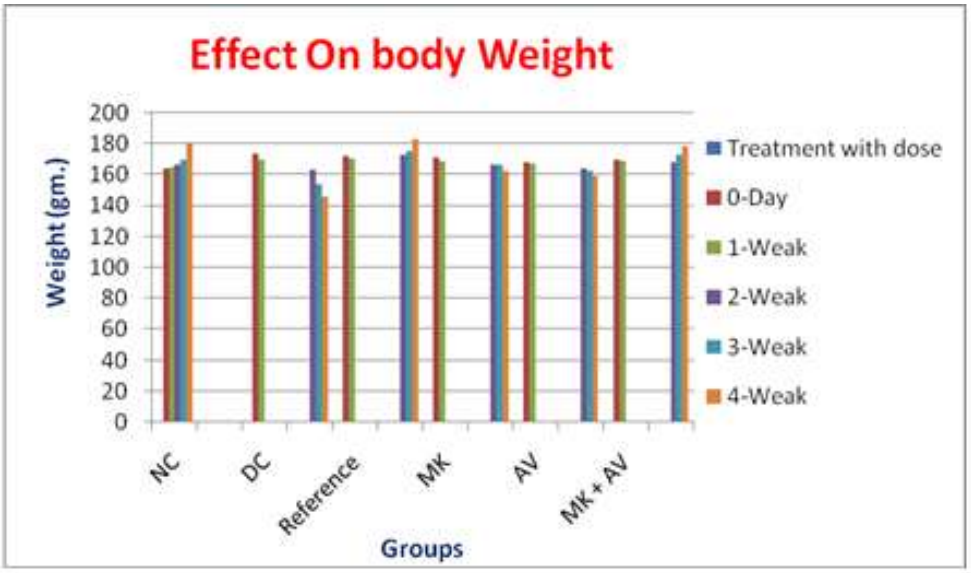
Effect on body weight

Body weight of all the rats in all groups were carried out before the treatment (0 day) and post the treatment on 7th, 14th, 21st and 28th day of study with the help of electronic balance. The results are shown in table 2.

Table 2: Effect of Metformin, MK aq. Extract, AV Aq. Extract and Combination of MK + AV on body weight (gm) in rats (n=5)

Groups	Treatment with dose	0-Day	1-Weak	2-Weak	3-Weak	4-Weak
I	NC	164.12 ± 6.14	165.42 ± 5.96	166.76 ± 5.97	169.50 ± 5.73	180.26 ± 6.51
II	DC	173.88 ± 7.04	169.60 ± 7.21	163.78 ± 7.60	154.30 ± 7.79	145.66 ± 9.43
III	with Metformin	172.54 ± 8.89	170.96 ± 8.53	173.04 ± 8.05	175.06 ± 7.60	183.24 ± 8.01
VI	MK	171.21 ± 6.90	168.78 ± 7.85	166.96 ± 7.25	167.04 ± 9.26	162.66 ± 9.53
V	AV	168.64 ± 8.93	167.12 ± 9.48	164.18 ± 9.53	162.38 ± 8.34	159.26 ± 8.58
VI	MK + AV	169.74 ± 8.37	168.82 ± 8.30	168.58 ± 7.67	172.96 ± 7.17	178.58 ± 7.15

n= 5, values express as Mean ± SD, significance P<0.05



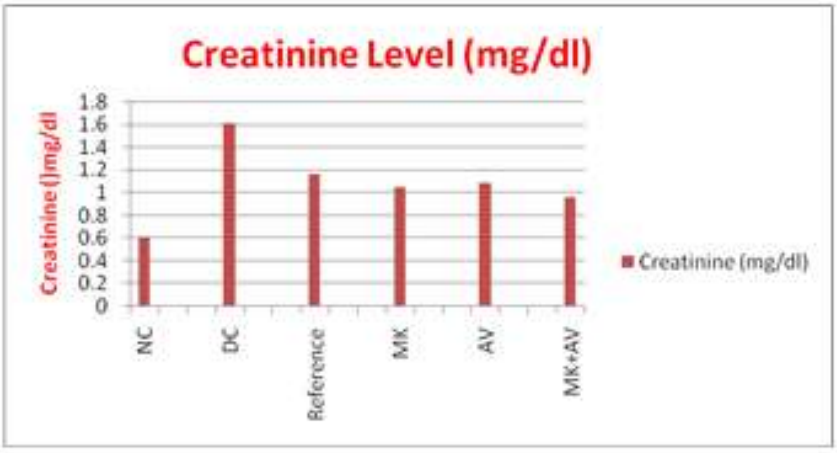
Effect on Biochemical parameters in Alloxan induced diabetic rats

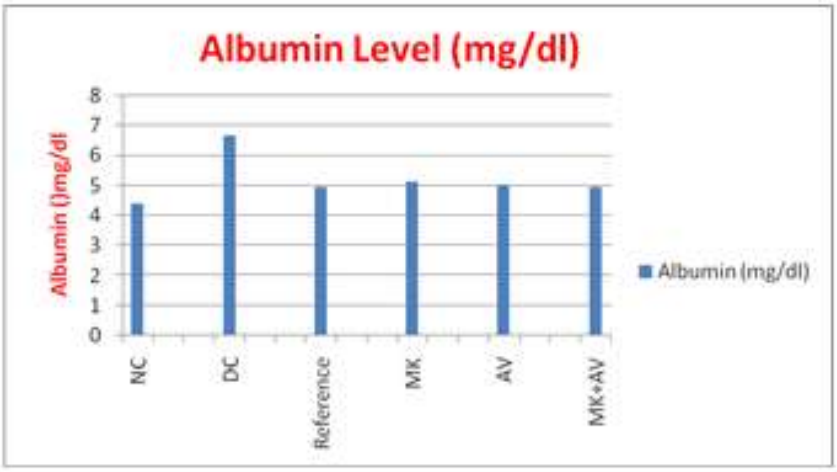
At the end of the experimental study estimation of serum biochemical parameters a varying effect were observed. In this study serum creatinine level was greatly reduced by the combination of MK and AV. The most effective extract to reduce serum creatinine was found to be in order of MK+AV > MK > AV. Serum albumin level was most significantly reduced and the order of effectiveness was found to be MK+AV > AV > MK. The order of extracts to reduce serum urea was MK + AV > MK >AV. Combination of MK and AV significantly reduced serum SGOT and the order was found to be MK+AV > AV > MK. The most significant extract to reduce serum SGPT, order was found MK + AV > MK > AV. Combination of MK and AV extract maintain near about all the biochemical parameters similar to standard drug Metformin. The results are shown in table 3 & 4.

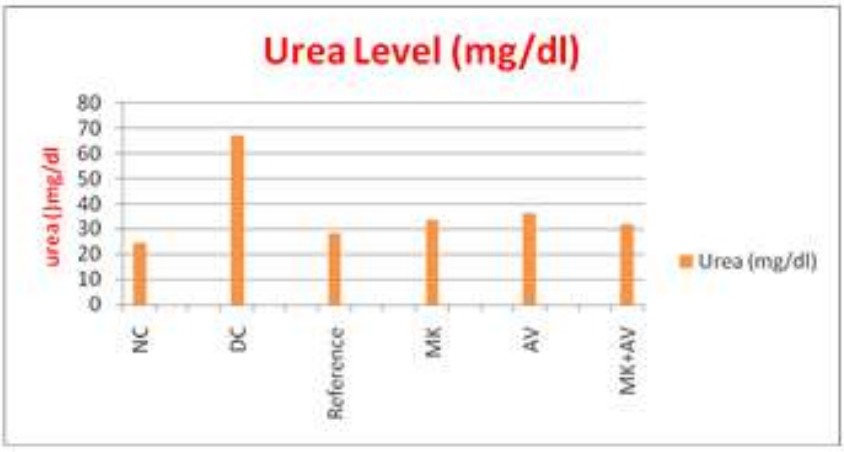
Effects of Plant extracts on Renal functioning

Table 3. Effect of aq. Extracts of MK, AV and MK+AV on serum creatinine, albumin and urea (Kidney function tests)

Groups	Treatment with dose	Creatinine (mg/dl)	Albumin (mg/dl)	Urea (mg/dl)
I	**NC**	0.612 ± 0.020	4.408 ± 0.183	24.670 ± 1.010
II	**DC**	1.620 ± 0.043	6.688 ± 0.155	67.172 ± 4.414
III	**Reference**	1.172 ± 0.106	4.956 ± 0.242	28.528 ± 3.716
IV	**MK**	1.052 ± 0.128	5.144 ± 0.369	33.522 ± 4.056
V	**AV**	1.094 ± 0.152	5.040 ± 0.435	36.260 ± 3.159
VI	**MK+AV**	0.96 ± 0.083	4.962 ± 0.168	31.80 ± 1.050


Creatinine Level (mg/dl)
Creatinine ()mg/dl
1.8
1.6
1.4
1.2
1
0.8
0.6
0.4
0.2
0
NC
DC
Reference
MK
AV
MK+AV
Creatinine (mg/dl)

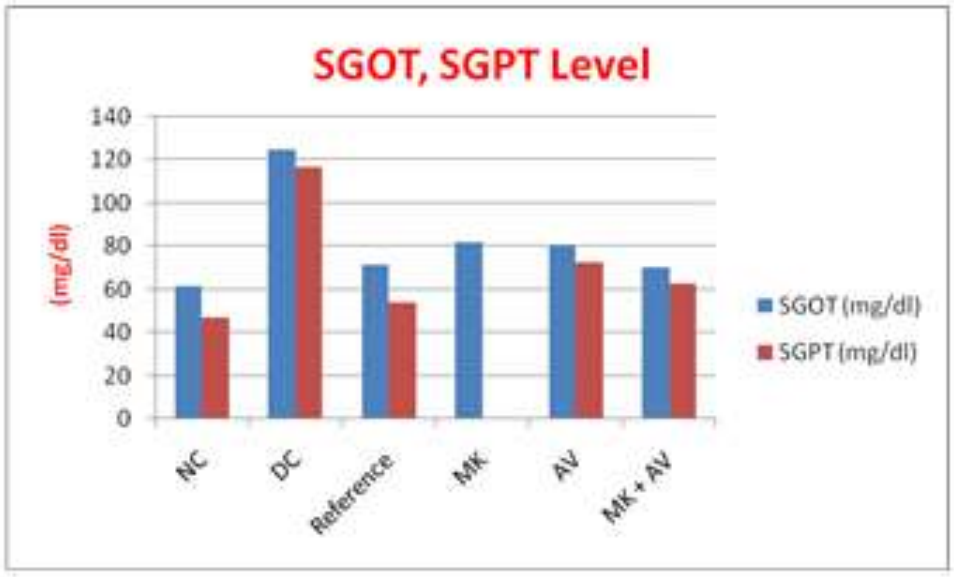

Albumin Level (mg/dl)
Albumin ()mg/dl
8
7
6
5
4
3
2
1
0
NC
DC
Reference
MK
AV
MK+AV
Albumin (mg/dl)


Urea Level (mg/dl)
urea ()mg/dl
80
70
60
50
40
30
20
10
0
NC
DC
Reference
MK
AV
MK+AV
Urea (mg/dl)

Effects of Plant extracts on Liver functioning

Table 4. Effect of of aq. extracts of MK, AV and MK+AV on serum glutamate oxaloacetate and pyruvate transaminase (SGOT and SGPT) (Liver function tests)

Groups	Treatment with dose	SGOT (mg/dl)	SGPT (mg/dl)
I	NC	61.788 ± 1.780	47.240 ± 5.032
II	DC	124.948 ± 5.961	116.604 ± 8.436
III	with Metformin	71.544 ± 7.474	53.844 ± 6.782
VI	MK	82.104 ± 6.896	69.036. ± 9.314
V	AV	80.824 ± 6.810	72.554 ± 7.608
VI	MK + AV	70.312 ± 1.229	62.740 ± 1.902



DISCUSSION

Folk medicinal plants have long been a crucial source of medicine, with many current drugs derived directly or indirectly from them. This study aimed to evaluate and compare the effects of aqueous extracts of Murraya koenigii (curry leaves), Aloe vera, and a combination of these two on blood glucose levels in alloxan-induced diabetic male albino rats. Additionally, the study assessed the impact on other related markers, including liver function tests (SGOT and SGPT), creatinine, serum albumin, and urea as indicators of renal function, as well as changes in body weight. A comparison with Metformin, a commonly used synthetic antidiabetic drug, was also conducted. Effective blood glucose control is essential for preventing or reversing diabetic complications and improving the quality of life for individuals with diabetes. The plant extracts demonstrated significant antidiabetic activity, effectively lowering blood glucose levels compared to the control group. Alloxan-induced diabetic hyperglycemia elevates plasma levels of urea, albumin, and creatinine, which are significant markers of renal dysfunction, as well as SGOT and SGPT levels, which indicate liver dysfunction. The results, as shown in Table 3, revealed a significant decrease in plasma levels of urea, albumin, and creatinine, while Table 4 indicated a significant reduction in serum levels of SGOT and SGPT in diabetic rats treated with aqueous extracts and their combination, compared to the mean values of the diabetic group.

CONCLUSION

In diabetes mellitus, achieving good care and optimal glycemic control is very challenging. The

present study evaluated the effects of aqueous extracts of Murraya koenigii (curry leaves), Aloe vera, and their combination on blood glucose levels in experimental rats. During the treatment, the weight of the group treated with the combined extracts and the group treated with Metformin increased, whereas the weight of the other groups decreased slightly compared to the diabetic control group. The plant extracts also reduced plasma creatinine, albumin, urea, SGOT, and SGPT levels in alloxan-induced diabetic rats, thereby mitigating kidney and liver dysfunction compared to the diabetic control group. Our study demonstrates that the aqueous extracts of Murraya koenigii, Aloe vera, and their combination have a beneficial modulatory effect in the treatment of diabetes mellitus and may be utilized as part of diabetes management. Since these folk medicines have been traditionally used as stable foods in daily life, they may be safe for use as dietary supplements and medications. However, long-term research is necessary, as herbal plant compounds often act more slowly than synthetic medications, and higher dosages might lead to a plateau effect.

CONFLICT OF INTEREST:

The authors declare that they don't have any

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