INTRODUCTION

Plants are extremely an important source of medicinal agents. Since ancient times, plants have been used due to their impactful pharmacological properties. WHO estimates that 80% of the population living in rural areas is dependent on herbal medicine for their health needs. Crude plant extracts in the form of infusion, decoction, tincture or herbal extracts have been traditionally used by the population for the treatment of various diseases. Plants used in treatment of diseases is as old as civilization and traditional medicines are still a major part of habitual treatments of different maladies. Diabetes mellitus is the commonest endocrine disorder that affects more than 171 million people worldwide. It is a condition in which a person has a high blood sugar level. The reason may be either the body doesn’t produce enough insulin, or because body cells don’t properly respond to the insulin that is produced. When the blood glucose increases for example, after eating food material, insulin is released from the pancreas to normalize the glucose level. If the body cells do not absorb glucose, the glucose accumulates in the blood, leading to vascular, neuron, and other complications. Our body is exposed to a variety of oxidizing agents. Oxidation is a chemical reaction that produces free radicals, leading to chain reactions that may damage cells. Body is equally inbuilt with antioxidants to cater for the free radicals generated from the oxidants, so a balance between the production of free radicals and neutralization by antioxidants is maintained. Antioxidants are substances known to protect the body from damage caused by reactive oxygen species induced oxidative stress. Oxidative stress results because of imbalance between formation and neutralization of free radicals by antioxidants, it results to oxidative stress. Oxidative stress has been implicated in the etiology of diseases such as...
The acclimatized animals were fasted for 24 h with water ad libitum\textsuperscript{13}. Hyperglycemia was induced using a single intraperitoneal injection of alloxan monohydrate (160 mg/kg). All animals were returned to their cages and given free access to food and water. Blood glucose levels were monitored by using a Glucometer after 72 h of injection and recorded as 1\textsuperscript{st} day. Rats with fasting blood glucose >7.8 mmol/l or 140 mg/dl were considered hyperglycemic and were selected for the study. Diabetic rats were randomly assigned to 5 groups, each group contains six animals. The animals were grouped as follow

- Group I: Normal control
- Group II: Diabetic control
- Group III: Diabetic rats treated with \textit{G. latifolium} extract (250 mg/kg)
- Group IV: Diabetic rats treated with \textit{G. latifolium} extract (500 mg/kg)
- Group V: Diabetic rats treated with glibenclamide (0.25 mg/kg).

Blood samples were obtained from the cut tail tip of the conscious rat and glucose test strip soaked with blood and then inserted to be read by the glucometer. Blood glucose levels were examined after 2, 12, 24, 72 hrs of orally administration of test drugs\textsuperscript{14}.

### Anti oxidant study

#### DPPH antioxidant assay

The free radical scavenging activity of \textit{G. latifolium} extract was analyzed by, 1, 1-diphenyl-2-hydrayl (DPPH) photometric assay. 2 ml of test extract at concentrations ranging from 10 to 400 μg/ml was each mixed with 1 ml of 0.5mM DPPH in methanol\textsuperscript{14}. Absorbance at 520 nm was taken after 30 min incubation in the dark at room temperature using a spectrophotometer\textsuperscript{15}. The experiment was done in triplicates and the percentage antioxidant activity was calculated as follows:

\[
\text{% of DPPH free radical scavenging} = \frac{\text{AbsC} - \text{AbsB}}{\text{AbsC}} \times 100
\]

Where; AbsC=Absorbance of control, AbsB=Absorbance of blank.

One ml methanol and 2ml extract were used as blank, while 1 ml 0.5 mM DPPH solution and 2 ml methanol was used as control. Ascorbic acid was used as reference standard.

| Treatment                  | Blood glucose level (mg/dl) |
|----------------------------|-----------------------------|
|                            | 0\textsuperscript{th} Day   | 7\textsuperscript{th} Day  | 14\textsuperscript{th} Day | 21\textsuperscript{st} Day |
| Normal control             | 90.4±0.25                   | 93.21±0.26                  | 92.47±0.22                  | 91.65±0.15                  |
| Diabetic control           | 247.58±0.43                 | 265.28±0.31                 | 280.37±0.41                 | 300.48±0.42                 |
| \textit{G. latifolium} (250 mg/kg) | 245.72±0.51                 | 200.82±0.42                 | 150.47±0.53                 | 122.37±0.37                 |
| \textit{G. latifolium} (500 mg/kg) | 247.38±0.09                 | 195.28±0.51                 | 142.37±0.08                 | 120.25±0.22                 |
| Glibenclamide (0.25 mg/kg) | 248.46±0.13                 | 179.23±0.38                 | 125.38±0.06                 | 112.37±0.07                 |

\(N=6, p < 0.05\)

### RESULTS AND DISCUSSION

The anti-hyperglycemic effect of methanol extract of \textit{G. latifolium} leaves was evaluated for 21 days. Alloxan-induced hyperglycemia is due to selective toxicity of alloxan on the pancreatic beta cells, generation of superoxide radicals and cytotoxic action mediated by generation of reactive oxygen species. The cytotoxic action of alloxan is mediated by reactive oxygen species, with a simultaneous massive increase in cytosolic calcium concentration, leading to a rapid destruction of beta cells\textsuperscript{16}. The effect of administration of extracts of \textit{G. latifolium} on blood glucose level of
The effect of different treatment on body weight of alloxan induced diabetic rats is shown in Table 1. Treatment of extract of *G. latifolium* showed a significant reduction in the blood glucose level. It also shows that 0.25 mg/kg glibenclamide is lowering glucose level significantly compared to normal control. Leaf extract at a dose of 500 mg/kg was more effective in reducing the blood glucose level than a dose of 250 mg/kg. The variation in blood glucose level in normal and experimental rats on 0, 7, 14 and 21 days of treatment has also been recorded. Treatment with *G. latifolium* extract showed signs of recovery as comparable with the standard drug glibenclamide. There was significant loss in body weight of diabetic rats compared to normal rats. In diabetics, glucose is not available therefore the cells use alternatively proteins for energy; consequently due to excessive breakdown of tissue protein a loss in body weight occurs. Body weight slightly increased in the normal control rats compared to initial body weight, whereas in diabetic control rats there was a significant decrease in body weight. Groups that were treated with glibenclamide and *G. latifolium* extract (250 and 500 mg/kg) showed significant reduction in body weights. The final body weights of treated groups were significantly lower than the final weights of normal control group. Hence, present study showed a good antidiabetic response of leaf extract against the diabetic rats, most likely to be associated with glucose uptake increasing mechanism. Lower doses of the extract should be tried in future study to establish the most appropriate dose for clinical trial. *G. latifolium* leaves has shown to be a potential anti-diabetic and thus it can be a promising source for anti-diabetic agent. In the present study, the observed DPPH scavenging activity of the methanolic extract of *G. latifolium* might be useful for the development of newer and more potent natural antioxidants. So, present concentration was found to be a suitable concentration for the treatment of diabetes.

**Statistical Analysis**

Graph Pad Prism Version 5.0 for Windows was used for all statistical analyses. Data are presented as mean±SEM and analyzed by one-way ANOVA followed by Dunnett’s multiple comparison test.

**CONCLUSION**

At present scenario, many researchers are showing their interest in medicinal plants for routine scientific investigation of numerous plants extract for biological effects and potential therapeutic properties in human. Methanol leaf extract of *G. latifolium* have anti-hyperglycemic and anti-oxidant activities. The methanol extract have antihyperglycemic activity in diabetic rats, most likely to be associated with glucose scavenging activity of the methanolic extract of *G. latifolium* is shown in Table 2. The result showed that *G. latifolium* extract caused a concentration dependent percentage increase of antioxidant activity. DPPH is a stable free radical, it accepts an electron or hydrogen radical to become a stable diamagnetic molecule which is widely used to investigate radical scavenging activity. The degree of discoloration indicates the radical-scavenging potential of the antioxidant. Result shows that DPPH scavenging was increased in a concentration dependent manner compared to ascorbic acid.

**Table 2:** The effect of different treatment on body weight of alloxan induced diabetic rats

| Treatment     | Initial (gm) | 10th Day (gm) | 21th Day (gm) |
|---------------|--------------|---------------|---------------|
| Normal control| 154±0.31     | 174.00±0.8    | 180.50±0.32   |
| Diabetic control | 181±0.28    | 165.00±0.64   | 140.67±0.43   |
| *G. latifolium* (250 mg/kg) | 158±0.17     | 147.67±2.10   | 148.33±0.7    |
| *G. latifolium* (500 mg/kg) | 174±0.36     | 149.00±0.7    | 144.37±1.2    |
| Glibenclamide (0.25 mg/kg) | 176±0.28     | 157.00±0.53   | 167.83±0.9    |

N=6, p < 0.05

**Table 3:** Antioxidant activity of the methanol extract of *G. latifolium*

| Sample                  | Conc (µg/ml) | Absorbance | Absorbance of control | Mean % DPPH scavenging activity |
|-------------------------|--------------|------------|-----------------------|-------------------------------|
| Ascorbic acid           | 10           | 0.069      | 0.069                 | 86.43±0.17                   |
|                         | 100          | 0.060      | 0.060                 | 90.63±0.32                   |
|                         | 200          | 0.049      | 0.049                 | 89.49±0.53                   |
|                         | 400          | 0.038      | 0.038                 | 93.69±0.82                   |
| Methanol extract        | 10           | 0.269      | 0.269                 | 70.33±0.38                   |
|                         | 100          | 0.250      | 0.250                 | 76.09±0.24                   |
| *G. latifolium*         | 200          | 0.215      | 0.215                 | 78.18±0.31                   |
|                         | 400          | 0.190      | 0.190                 | 80.27±0.78                   |

N=6, p<0.05
study concluded that detailed investigation of plants used in local health traditions and pharmacological evaluation of these plants and their taxonomical relatives can lead to the development of invaluable plant drug for many dreaded diseases including diabetes mellitus.

**AUTHOR’S CONTRIBUTION**
The manuscript was carried out, written, and approved in collaboration with all authors.

**CONFLICT OF INTEREST**
No conflict of interest is related to this paper.

**REFERENCES**
1. Bernhoft A. A brief review on bioactive compounds in plants, In: Bioactive compounds in plants – benefits and risks for man and animals, Oslo: The Norwegian Academy of Science and Letters 2010; 11-17.
2. Firn R. Nature’s Chemicals. Oxford University Press, Oxford. 2010; 74-75.
3. Piero MN, Nzaro GM, Njagi JM. Diabetes mellitus: a devastating metabolic disorder. Asian J Biomed Pharm Sci 2014; 4(4):1-7. https://doi.org/10.15272/aajbps.v4i40.645
4. Chamnan P, Simmons RK, Forouhi NG, Luben RN, Khaw KT, Wareham NJ, Griffin SJ. Incidence of type 2 diabetes using proposed HbA1c diagnostic criteria in the European prospective investigation of cancer–Norfolk Cohort implications for preventive strategies. Diabetes Care 2011; 34(4): 950-956. https://doi.org/10.2337/dc10-0326
5. Del CM, Sacchetti G, Di Mattia C, Compagnone D, Mastrocola D, Liberatore L, Cichelli A. Contribution of the phenolic fraction to the antioxidant activity and oxidative stability of olive oil. J Agric Food Chem 2004; 52(13): 4072-9. https://doi.org/10.1021/jf049806c
6. Cook NR, Albert CM, Gaziano JM. A randomized factorial trial of vitamins C and E and beta carotene in the secondary prevention of cardiovascular events in women: results from the women's antioxidant cardiovascular study. Arch Intern Med 2007; 167(15): 1610–8. https://doi.org/10.1001/archinte.167.15.1610
7. Jain S, Jain N. Antioxidant: literature review. World J Pharm Pharm Sci 2016; 5(10): 365-377.
8. Edet EE, Akpanabiata MI, Eno AE, Umoh IB, Itam EH. Effects of Gongronema latifolium crude leaf extract on some cardiac enzymes of alloxan-induced diabetic rats. Afr J Biochem Res 2009; 3(11):366-369.
9. Balogun ME, Besong EE, Otimma JN, Mbamalu OS, Djobissie SFA. Gongronema latifolium: A phytochemical, nutritional and pharmacological review. J Phys Pharm Adv 2016; 6(1):811-824. https://doi.org/10.5455/jppa.1969123104000
10. Morebise O. A review on Gongronema latifolium, an extremely useful plant with great prospects. Eur J Med Pl 2015; 10(1):1-9. https://doi.org/10.9734/EJMP/2015/19713
11. Morebise O, Fafunso MA, Makinde JM. Membrane stabilizing activity: Possible mechanism of action for the anti-inflammatory property of Gongronema latifolium leaves. Int J Biom Heal Sci 2005; 1(1):15-19.
12. Kuhara T. Consensus recommendations on effective institutional animal care and use committees. Information on overseas technologies in laboratory animal science. 1988; 7: 14-17.
13. Naowaboot J, Pannangpetch, P, Kukongviriyapan V, Kongyingyoes B, Kongyingyoes K. Antihyperglycemic, antioxidant and antiglycation activities of mulberry leaf extract in streptozotocin-induced chronic diabetic rats. Plant Foods Human Nutrition 2009; 64: 116-121. https://doi.org/10.1007/s11130-009-0112-5
14. Batra S, Batra N, Nagori BP. Preliminary phytochemical studies and evaluation of antidiabetic activity of roots of Cayratia trifolia (L.) Domin in alloxan induced diabetic albino rats. J Appl Pharm Sci 2013; 3:97-100. https://doi.org/10.7324/JAPS.2013.30319
15. Chioma DE. Anti-inflammatory and anti-oxidant activities of methanol extract of Baphia nitida. Universal J Pharm Res. 2016; 12(2): 42-47. https://doi.org/10.22270/ujpr.v12i2.R5
16. Jorns A, Munday R, Tiedge M, Lenzen S. Comparative toxicity of alloxan, N-alkyl-alloxsans and ninhydrin to isolated pancreatic islets in vitro. J Endocrinol. 1997; 155: 283-293