

Effect of Ginger Drink on the Effectiveness on Glimepiride Drug Used to Treat Diabetes in Rats

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Abstract Diabetes mellitus (DM) is a metabolic disease, involving inappropriately elevated blood glucose levels. The combined use of herbs and antidiabetic drugs increases the like hood of pharmacokinetic and pharmacodynamic interactions. Therefore this study aims to investigate the effect of ginger drink on the efficiency glimepiride drug used to treat diabetes in rats. Thirty six adult albino rats Sprague Dawley strain, weighing (150+5g) were randomly divided into two main groups. Group I: negative control group (6 rats), fed standard diet only. Group II (diabetic rats), (n = 30) . Divided into 5 subgroups, (6 rats each) according the following: Subgroup 1: positive control fed standard diet only. Subgroup 2: fed standard diet plus glimepiride drug as (4 mg/kg orally once daily) a treatment of diabetic rats. Subgroup 4, 5 and 6 were fed standard diets and received a daily oral dose of glimepiride drug plus a daily oral dose of ginger drink 5, 7.5 and 10 µl/g of body weight respectively. At the end of the trial (30 days) rats were fasting for 12h and blood samples were collected and centrifuged to obtain serum and kept frozen until analysis. Giving rats with ginger drink at different concentration with glimepiride drug enhanced the levels of glucose, insulin, lipids profile, malondialdehyde and catalase and compared to positive control group. Conclusion: The use of ginger drink in different concentrations has a strong effect in increase the efficacy of glimepiride drug.

Keywords: Ginger, diabetes, blood sugar, insulin, glimepiride

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1. Introduction

Diabetes mellitus is a chronic condition caused by either insufficient insulin production by the pancreas or inefficient insulin utilization by the body that controls blood sugar levels [1]. It is one of the most common non-communicable diseases of the current world, affecting 422 million has increased exponentially around the globe in the last 3 decades. Similarly, 108 million people had had diabetes mellitus in 1985, the number increased to 422 million by 2016 which is predicted to be doubled by 2030 [2,3]. The International Diabetes Federation (IDF) recorded Egypt among the world's top 10 countries in the number of patients with diabetes. Clinical symptoms of diabetes include- frequent urination, frequent thirst, increased hunger, weight loss and fatigue. In addition to high blood glucose levels, there are usually high levels of glucose in the urine. The United States Food and Drug Administration (FDA) approved glimepiride for the treatment of T2DM as monotherapy as well as in combination with metformin or insulin. [4]. Treatment with glimepiride as therapy results in a 1.5-2.0% reduction in HbA1c [5].

Most patients often make use of herbs and prescribed medications without the knowledge of their health care

provider and the fact that these health care providers are less informed of herb-drug interactions and resultant adverse effects is a serious cause for concern [6]. The Evidence from clinical studies, the combined use of herbs and antidiabetic drugs can increase or decrease the efficacy and toxicity of the drugs [7]. Ginger, has been consumed worldwide in cookeries, as a spice and as a flavoring agent. It has been cultivated for thousands of years for medicinal purposes and used extensively in traditional medicine [8]. [9] found that interaction of ginger with glibenclamide was found to be promising in reducing blood glucose level in streptozotocin- (STZ-) induced diabetes. Therefore, this study aims to evaluate effect of ginger drink on the effectiveness of glimepiride drug used to treat diabetes in rats.

2. Materials

Ginger was purchased from Harraz company Main Store at -Down Town1 Ahmed Maher St. Bab Alkhalq, Cairo, Alloxan and Glimepiride (1 tablet contain 6 mg Glimepiride) (an oral antidiabetic drug for non-insulin-dependent diabetes) obtained from Sigma (29 Mawardi Street - Qasr al-Aini. Cairo – Egypt). Casein, vitamins, cellulose, choline chloride, methionine and salt mixture

were obtained from Techno gene Pharmaceutical Company, Cairo, Egypt. The kits were purchased from Bio Diagnostics Company, Cairo, Egypt.

Thirty adult albino rats weighting (150 ± 5 g) were obtained from Giza, Cairo.

3. Methods

Preparation of ginger drink

Put four grams of dried ginger powder in a cup, pour 100 ml of boiling distilled water over it, leave for 10 minutes, then filter and place in a sterilized bottle in the refrigerator until use according to [10].

Analytical Methods

Determination of Total phenolics, total flavonoids contents and DPPH of ginger

Total phenols, total flavonoids contents and DPPH were determined in ginger drinks according to [11,12,13] respectively.

Induction of diabetic mellitus:

Diabetes will be induced in normal healthy male albino rats (45 rats) by subcutaneous injection of alloxan (150 mg /kg body weight) according to the method described by [14]. One week after the injection of alloxan, fasting blood samples will be obtained for estimating fasting serum glucose. Rats having fasting serum glucose >200 mg/dl will be considered diabetics.

Experimental design

The research protocol (MUFHE/S/NFS/9/23) has been approved by the Science Research Ethics Committee of The Institutional Animal Care and Use Committee (IACUC) at Menoufia University. Thirty six adult albino rats Sprague Dawley strain, weighing (150 ± 5 g) were obtained from Research Institute of Ophthalmology, Giza, Egypt. The work was carried out at the Faculty of Home Economic, Menofia University, Egypt. Rats were housed separately in cage and fed standard diet according to AIN-93 guidelines [15] for 7 days for adaptation, then rats were randomly divided into two main groups. Group I: negative control group (6 rats), fed standard diet only. Group II (diabetic rat groups), ($n = 30$) divided into 5 subgroups, (6 rats each) according the following: the first subgroup served as positive control and fed standard diet only. The second subgroup: diabetic rats fed standard diet and received 4 mg/kg orally once daily glimepiride (as suspension in freshly prepared 0.5% w/v carboxy methyl cellulose sodium salt) a treatment of diabetic according to [16]. The third, fourth and fifth subgroup fed standard diet and received 4 mg/kg orally once daily glimepiride plus a

daily oral dose of ginger drink 5, 7.5 and 10 μ l/g of body weight respectively once a day for 30 days, as described by [16].

Blood sampling:

At the end of experimental period (30 days), rats were anesthetized after fasting for 12h and blood samples were collected by using the retro-orbital method by means of a micro capillary glass tube. Blood was collected into a dry clean centrifugal tube and left to clot in a water bath (37°C) at room temperature for half an hour. The blood was centrifuged for 10 minutes at 3000 rpm to separate the serum. Serum was kept in clear quit fit plastic tubes and stored at -20°C until analysis according to the method described by [17].

Blood parameters

Serum glucose, insulin was estimated according to [18] and [19], Catalase, and malonaldehyde (MDA) were determined according to the methods described by [20] and [21] respectively. Total cholesterol (TC), Triglycerides (T.G) and High density lipoprotein (HDL) lipid were determined according to [22,23,24] respectively, Low density lipoprotein (LDL) and Very low density lipoprotein (VLDL) were calculated according to the methods of [25] as follows:

$$\text{VLDL} = \text{TG}/5.$$

$$\text{LDL} = \text{Total cholesterol} - (\text{HDL} + \text{VLDL}).$$

The Atherogenic ratios were calculated according to [26] as follows: Atherogenic Index (AI) = $\log \text{TG}/\text{HDLc}$, Atherogenic Coefficient (AC) = $(\text{TC} - \text{HDLc})/\text{HDLc}$.

Statistical Analysis

Results were expressed as the mean $\pm$ SD. Data for multiple variable comparisons were analyzed by one-way analysis of variance (ANOVA). For the comparison of significance between groups, Duncan's test was used as a post hoc test according to the statistical package program [27].

4. Results and Discussion

Table 1 showed that the total phenols, total flavonoids contents and DPPH of ginger. The results indicated that ginger contained 840 mg GAE/ 100 g DM, 2.98 mg CAT/100 g DM and 73.529% for total phenols, total flavonoids and DPPH respectively. These results are in agreement with [28] who showed that ginger is known for its high total phenolic content, which contributes to its antioxidant properties. [29] reported that ginger exhibits potent DPPH radical scavenging activity, indicating strong antioxidant potential.

Table 1. Total phenols, total flavonoids, and DPPH contents of ginger

Active compounds	Ginger
Total phenols (mg GAE/ 100 g DM)	840
Total flavonoids (mg CAT/100 g DM)	2.98
DPPH (%)	73.529

CAT: Catechin, GAE: Gallic acid equilibrium, DM: Dry matter, DPPH: 2-diphenyl-1-picrylhydrazyl

Table 2. Effect of ginger on serum insulin and glucose in negative and diabetic groups

Variables	Negative control	Diabetic groups				
		Positive	Drug	Drug + Ginger (5 μ L/g/ BW)	Drug + Ginger (7.5 μ L/g/BW)	Drug + Ginger (10 μ L/g of BW)
Glucose(mg/dl)	92.44 ^a ±3.65	318 ^b ±5.45	147.66 ^b ±3.05	122.78 ^c ±3.65	116.43 ^d ±1.00	112.28 ^d ±1.46
Insulin (mg/dl)	5.51 ^a ±0.30	1.03 ^f ±0.03	1.98 ^e ± 0.08	2.36 ^d ± 0.20	3.77 ^c ± 0.11	4.52 ^b ± 0.25

Values are expressed as means $\pm$ SD. means in the same raw with different letter are significantly different ($P \leq 0.05$).

Table 3. Effect of ginger on serum lipids profile in negative and diabetic groups

Variables	Negative control	Diabetic groups				
		Positive	Drug	Drug + Ginger (5 μ L/g/ BW)	Drug + Ginger (7.5 μ L/g/BW)	Drug + Ginger (10 μ L/g of BW)
TC (mg/dl)	116.5 ^f ±0.82	245.6 ^a ±8.5	169.4 ^c ±2.19	186.8 ^b ±2.68	138.5 ^d ±3.39	124.5 ^e ±3.62
TG (mg/dl)	97.39 ^a ±1.71	178.1 ^a ±2.8	141.8 ^b ±4.85	139.5 ^b ±3.40	114.05 ^c ±1.65	96.64 ^d ±0.91
VLDL(mg/dl)	19.48 ^a ±0.34	35.62 ^a ±0.56	28.36 ^b ±0.96	27.91 ^b ±0.68	22.81 ^c ±0.33	19.47 ^d ±0.34
HDL(mg/dl)	50.18 ^a ±1.41	47.35 ^a ±2.27	49.09 ^a ±2.19	47.38 ^a ±1.94	49.35 ^a ±1.02	49.20 ^a ±1.91
LDL(mg/dl)	46.83 ^a ±1.25	162.7 ^a ±6.55	91.96 ^a ±1.29	111.49 ^b ±4.56	66.28 ^c ±4.19	55.86 ^d ±5.34

Values are expressed as means $\pm$ SD. means in the same raw with different letter are significantly different ($P \leq 0.05$). TC: Total cholesterol, TG: Triglyceride, HD: High density lipoprotein, LDL: Low density lipoprotein cholesterol, VLDL: Very low density lipoprotein.

Table 4. Effect of ginger on antioxidant status in negative and diabetic groups

Variables	Negative control	Diabetic groups				
		Positive	Drug	Drug + Ginger (5 μ L/g/ BW)	Drug + Ginger (7.5 μ L/g/BW)	Drug + Ginger (10 μ L/g of BW)
MDA (mg/dl)	0.54 ^d ±0.01	5.51 ^a ±0.50	3.70 ^b ±0.23	3.48 ^b ±0.20	1.27 ^c ±0.10	1.05 ^c ±0.09
CAT (mg/dl)	13.42 ^a ±1.15	1.04 ^d ±0.07	0.94 ^d ±0.04	2.05 ^c ±0.19	6.33 ^b ±0.56	13.83 ^b ±0.63

Values are expressed as means $\pm$ SD. means in the same raw with different letter are significantly different ($P \leq 0.05$). MDA: Malondialdehyde, CAT: Catalase.

Effect of ginger on serum insulin and glucose in negative and diabetic groups are presented in Table 2. Positive group had significantly ($p \leq 0.05$) higher glucose than negative and other diabetic groups while, serum insulin had opposite trend. Administration rats with 5, 7.5 and 10 μ L of ginger drink plus drug decreased the blood glucose and increased the insulin levels by 61.4, 63.4, 64.7 % and 1.3, 2.7, 3.4 % respectively as compared with positive control group. Also, the results indicated that giving the rats ginger drink led to improve drug efficacy as the improvement in the serum glucose and insulin levels in the diabetic groups that were given 5, 7.5 and 10 μ L/g of body weight of ginger drink with the drug was better than the diabetic group that was given the drug only. The highest improvement of blood glucose and insulin levels was observed in rats administrated with 10 μ L of ginger drink. The obtained results agree with [30] highlights ginger's potential to lower blood glucose and improve insulin sensitivity in diabetic models. Furthermore, [31] found that ginger can significantly reduce blood glucose levels and increase insulin secretion in diabetic subjects.

The effect of ginger on lipids profile in negative control and diabetic rats are presented in table (3). The result showed that positive control rats had higher ($p \leq 0.05$) total cholesterol (TC), triglyceride (TG), low-density lipoprotein (LDL) and very low-density lipoprotein (VLDL) than negative and ginger drink groups. Administrated rats with 5, 7.5 and 10 μ L/g BW of ginger drink with the drug significantly ($p \leq 0.05$) notably reduce TC, TG, VLDL, and LDL levels. However, there were no

significant difference on HDL levels among negative and diabetic groups. Also, the results showed that the best treatment for improving lipid levels was the group given 10 μ L/g BW of ginger drink with the drug, bringing them closer to the negative control values the values were the same as the negative control. Overall, ginger appears to enhance the efficacy of the drug treatment in managing lipid profiles. Additionally, [32] showed that ginger effectively lowers lipid levels in diabetic models. In the same table giving rats with drug or 5 μ L/g BW on ginger drink with the drug did not differ ($p > 0.05$) in their effect of TG and VLDL.

The Table 4 presents the effect of ginger on antioxidant status in negative and diabetic groups. MDA, a marker of oxidative stress, is significantly elevated in the positive control group (5.51 mg/dl) compared to the negative control (0.54 mg/dl). Administrated rats with 5, 7.5 and 10 μ L/g BW of ginger drink with the drug significantly ($p \leq 0.05$) improvement the MDA and CAT levels. The highest improvement was observed in rats that giving 10 μ L/g BW of ginger drink with the drug. This improvement may be due to high total phenolic content and strong DPPH free radical scavenging activity in ginger (as shown in Table 1), suggesting strong antioxidant potential. Overall, ginger enhances the drug's effect by reducing oxidative stress and increasing antioxidant enzyme activity. This is in agreement with [33] found that ginger significantly decreases MDA levels and increases CAT activity in various oxidative stress models. Also, the results indicated that there was no significant ($p > 0.05$) in MDA between rats giving drug and 5 μ L/g BW of ginger drink with the drug.

Table 5. Effect of ginger on atherogenic index in negative control and diabetic groups

Variables	Negative control	Diabetic groups				
		Positive	Drug	Drug + Ginger (5 μ L/g BW)	Drug + Ginger (7.5 μ L/g BW)	Drug + Ginger (10 μ L/g BW)
AI (mg/dl)	0.28 ^a ±0.01	0.57 ^a ±0.02	0.46 ^b ±0.02	0.46 ^b ±0.01	0.36 ^c ±0.01	0.29 ^d ±0.02
AC (mg/dl)	1.32 ^e ±0.05	4.19 ^a ±0.17	2.45 ^c ±0.11	2.94 ^b ±0.20	1.80 ^d ±0.13	1.53 ^e ±0.17

Values are expressed as means $\pm$ SD. means in the same raw with different letter are significantly different ($P \leq 0.05$). AI: atherogenic index, AC: Atherogenic Coefficient. Values are expressed as means $\pm$ SD. means in the same raw with different letter are significantly different ($P \leq 0.05$).

The table displays the impact of ginger on the atherogenic index (AI) and atherogenic coefficient (AC) in both control and diabetic groups. The diabetic group showed the highest AI and AC values, indicating an elevated risk of atherogenesis. However, these values significantly dropped with drug treatment alone, achieving an AI of 0.46 and an AC of 2.45. With progressively higher doses of ginger, further reductions in AI and AC were noted, reaching the lowest levels at a 10 μ L/g BW dose of ginger. At this concentration, the AI decreased to 0.29, and the AC dropped to 1.53, approaching the values of the negative control group. This data suggests that ginger, particularly at higher doses, significantly lowers both AI and AC in diabetic subjects when combined with drug therapy, highlighting its potential role in reducing atherogenic risks linked to diabetes. This aligns with previous research that has demonstrated the beneficial effects of ginger on lipid profiles and cardiovascular health. [34] found that ginger extract significantly reduced atherogenic indices in diabetic rats. Similarly, [35] reported that ginger supplementation reduced lipid peroxidation and improved antioxidant status, contributing to a reduction in atherogenic indices.

Conclusion

From the above results, it could be concluded that ginger drink improved the effectiveness of glimepiride, a drug used to treat diabetes in rats. We recommended conducting clinical trials on diabetic patients by taking glimepiride drug them with ginger drink.

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