Melia Azedarach L. Fruit Extract Effect on Plasma Lipid Profile and Cardiac and Hepatic Functions of Diabetic Rats

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Abstract

Objective: There are controversial reports about biological effects of the Melia Azedarach Linn. *(MA)*. In the present study, the effect of MA fruit extract on lipid profile, cardiac and hepatic functions of diabetic rats was investigated.

Materials and Methods: Thirty male Wistar rats were divided into five groups. Group 1 as the control group, Group 2 as the diabetic group, Group 3 and 4 as the diabetic rats treated with *MA* fruit extract (100 and 200 mg/kg for 30 days, orally) and Group 5 as the positive control group treated by Glibenclamide (0.5 mg/kg for 30 days). Diabetes was induced by nicotine amide and streptozotocin. After 30 days, the plasma concentration of glucose, hemoglobin A1C (HbA1c), lipid profile, hepatic enzymes were measured. Also, electrocardiogram and interventricular pressures were recoded.

Results: The data showed that the plasma levels of glucose, HbA1c, aspartate aminotransferase, alanine aminotransferase and alkaline phosphatase were significantly increased in the diabetic group. These parameters were higher in groups treated with *MA* extract, without significant difference. The plasma lipid profile significantly changed in diabetic rats and did not come back to normal level in groups treated with *MA* extract. Compared to the control group, corrected QT interval was increased and the rate of interventricular pressure changes decreased in all groups. *MA* extract was as an antioxidant compound against diphenylpicrylhydrazyl (DPPH) free radicals.

Conclusion: These results demonstrated that although *MA* extract had an antioxidant effect, it did not improve the signs of diabetes in diabetic rats.

Keywords: *Melia Azedarach* L., Diabetes, Heart, Liver, Lipid

Introduction

Historically, the use of herbal medicines dates back to early human civilization (1). The current trend shows that consumption of herbal medicine is increasing worldwide (2,3). According to the World Health Organization (WHO) approximately 80% of people around the world have taken herbal medicine (2, 4). Nowadays, herbal medicine is available and accessible (2). One problem associated with herbal medicines is

that people often assume herbal medicine is safe (2). However, some plants are safe and others are unsafe (3). It follows that more research is needed on herbal medicines on animal models. This study addressed the effect of *Melia Azedarach* L. (*MA*) fruit extract on hepatic and cardiac parameters of type 2 diabetic rats, as well as plasma glucose and lipid profiles.

MA belongs to the meliaceae family and is native of Iran, India and China (5,6). Although the plant has wide distribution globally, it is sometimes confused with Azadirachtaindica (A.), which is a different species of the meliaceae family (7). *MA* has traditionally been used to treat leprosy, inflammation, and cardiovascular diseases (8).

Controversial effects have been reported in some experimental studies on *MA*.(5). Most studies reported that the plant has parasitical, antibacterial, antifungal, larvicideal, anticomplementary, cytotoxic, phytotoxic and antiovipoition effects (1,9-14). However, a case study reported that all parts of *MA* are toxic. This study used the database of the Taiwan National Poison Center at the Taipei Veterans General Hospital in which records showed that during years 1998 and 2005 five patients had been referred to the hospital poisoning from *MA* fruit extract. The most common signs of *MA* poisoning were reported as weakness, ptosis and increased plasma levels of hepatic enzymes (6). Other studies reported poisoning and mortality as a result of *MA* fruit in cattle, dog, rats and mice (15-18). Another report mentioned that the extract had an antioxidant effect (19).Seifu et al recently reported the anti-diabetic effects of *MA* leaf, not its fruit extract on ob/ob mice (20).

In light of the past information, this study aimed to investigate the effect of *MA* fruit ethanolic extract on the plasma levels of fasting blood sugar (FBS), hemoglobin A1C (HbA1C), lipid profiles, hepatic enzymes, cardiac function and electrocardiogram parameters in type 2 diabetes mellitus (T2DM) rats.

Materials and Methods

To perform this study, 30 male Wistar rats with a weight range of 250-300 g were used. They were housed under standard conditions with 12/12 hours light–dark cycle. All animals had free access to water and a standard diet *ad libitum*. All animal procedures were performed accordance with the Guides for the Care and Use of Laboratory Animals and confirmed by the ethics committee of Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

MA fruit was collected from Yazd, Iran at the summer. It was identified at the Department of Botany, Faculty of Pharmacy, Shahid Sadoughi University of Medical Sciences, Yazd-Iran. A voucher specimen (M1395) is deposited at the Herbarium of the Herbal Medicine Research Center of Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

Briefly, *MA* fruit was dried in shade. Then, 500 grams of the dried fruit was soaked in 70% ethanolic solution for 72 hours. Afterword, the extract was filtered through a filter paper and then dried using a rotary evaporator. Finally, the dried extract was kept in a dark bottle.

The total flavonoids content of the extract was measured using colorimetric assay. One ml of sample was added to a 15 ml falcon tube and completely mixed by 4 ml distilled water. Then, 0.3 ml of NaNO2 5% (w/v), 0.3 ml of AlCl3 10%, and 2 ml of solution containing NaOH (1M) were added at 0, 5 and 6 minutes, respectively. The final volume was immediately reached to 10 ml by adding distilled water. The mixture was strongly shaken and its absorbance was read at the wave length of 520 nm. Flavonoid content is expressed in terms of quercetin equivalent (mg/g of extracted compound). (21).

The total phenolic content of extract was determined using the Folin–Ciocalteu reagent Evropi Botsoglou. Briefly, 0.5 g of sample was mixed with 25 ml methanol 80% in a 50 ml falcon tube and homogenized for 30 sec. Then, the homogenized solution was filtered. 50 µl of filtered solution was added to a 15 ml falcon tube and mixed with 6.45 ml distilled

water and 0.5 ml Folin–Ciocalteu. It was kept at room temperature for 3 min. 3 ml Na_2CO_3 7.5% was added and incubated for 60 min at room temperature. It was centrifuged at 2000 g for 5 min and the absorbance of the supernatant was read at 760 nm. Gallic acid (10–100 µg/ml) was considered as a standard reference substance. Finally, Total phenolic content of extract was determined as the equivalents of gallic acid per gram sample. (21).

The antioxidant capacity of *MA* extract was evaluated using the electroreduction and oxidation of diphenylpicrylhydrazyl (DPPH). In brief, several concentrations of *MA* extract (100-1000 µg) were selected. Each sample (1 ml) was mixed with 3 ml methanolic solution of DPPH (40 µg/ml) in dark environment for 30 min. Then, the absorbance of samples was read at 517 nm (Bio Tek Instrument Model: Box998). Finally, the antioxidant activity of extract was measured as amount of antioxidant required to reduce the initial absorbance of DPPH by 50% (IC50). (21).

To induce T2DM, animals were first kept in a fasting condition overnight. Then, after taking a blood sample they were injected with a single dose of nicotinamide (120 mg/kg, i.p) 30 min before injection of streptozotocin (40 mg/kg, i.p). After 1 week, rats had fasting blood sugar level above 200 mg/dl were considered as diabetic and grouped according to those set out below:

Group 1 as the control group (n=8): rats were not given anything.

Group 2 as the diabetic group (n=8): rats were diabetic with nicotine amide and streptozotocin (STZ).

Group 3 as the treatment 100 group: rats were diabetic and treated with 100 mg/kg *MA* fruit extract (orally) for 30 days.

Group 4 as the treatment 200 group: rats were diabetic and treated with 200 mg/kg *MA* fruit extract (orally) for 30 days.

Group 5 as the positive control group: rats were diabetic and treated with 0.5 mg/kg Glibenclamide (orally) as a conventional drug for 30 days.

All rats were anesthetized with sodium thiopental (75 mg/kg, i.p). Then, using two electrodes attached to the right hand and the left feet of animals, lead II electrocardiogram were recorded by Power lab data acquisition system (AD instrument, Australia). Finally, ECG parameters, including heart rate, QRS, RR, PR, QT, QTc and JT interval were analyzed by ECG analysis software of Power lab (ADinstrument, Australia).

Following recording of ECG, all animals were heparinized (1000 IU, i.p) and their hearts removed. The isolated hearts were placed under the Langendorff apparatus and perfused retrograde through aorta with a constant pressure of 70-80 mmHg. Then, a water–filled latex balloon was inserted into the left ventricle. The volume of balloon was increase to obtain left ventricular end diastolic pressure (LVEDP) of 4-7 mmHg. Twenty min after the stability, the left ventricular end systolic pressure (LVESP), and left ventricular developed pressure (LVDP), and the minimum and maximum rate of pressure change in the ventricle (max dp/dt and min dp/dt) were recorded .

Two blood samples were taken from each rat: the first sample was taken before heparinization and maintained in tubes containing Ethylenediaminetetraacetic acid to measure the plasma level of HbA1C, and the second sample was collected before excision of the heart to measure fasting blood sugar (FBS), Lipid profile, Creatine phosphokinase (CPK), Lactate dehydrogenase (LDH), aspartate aminotransferase, alanine aminotransferase (ALT), and alkaline phosphatase (ALP).

Statistical analysis

All data are shown as mean ± standard error. One–way analysis of variance (one–way ANOVA) with Tukey' s post hoc test was used to analyze differences among groups. $P<0.05$ was considered statistically significant.

Results

The total flavonoids content of the *MA* extract (as catechin equivalents) was 9±2.1 mg/g dry weight and its total phenolic content was 16±2.4 mg extract. Results showed that it significantly inhibited free radical capacity of DPPH (figure 1) in a dose-dependent manner with a 50% inhibitory concentration (IC_{50}) of 178 µg/ml.

As shown in Table 1, FBS and HbA1c showed a significant increase in diabetic rats (Group 2) that was reduced to approximately normal level in diabetic rats treated by Glibenclamide (Group 5). Treatment of diabetic rats with *MA* fruit extract at both doses (100 and 200 mg/kg) had no significant effect on plasma levels of FBS and HbA1c in diabetic rats and it appears higher dose may increase the plasma levels of FBS and HbA1c (Groups 3 and 4).

Table 1 showed that the plasma levels of total cholesterol, triglyceride and low-density lipoprotein (LDL) were markedly increased in diabetic rats (Group 2) that were reduced near to normal level in the diabetic rats treated with glibenclamide (Group 5). Compared to the diabetic group (Group 2), the lipid profiles of diabetic rats treated with 100 or 200 mg/kg *MA* fruit extract (Groups 3 and 4) had no significant difference. The changes in the plasma level of high-density lipoprotein (HDL) among groups were similar to those of LDL, but in the opposite direction.

Table 2 indicated that, compared to the control group (Group 1), the plasma levels of hepatic enzymes including AST, ALT and ALP significantly increased in diabetic rats (Group 2) and less increase was observed in diabetic rats treated with the conventional clinical drug glibenclamide (Group 5). Compared to group 2, the plasma levels of hepatic enzymes showed a significant increase in groups treated with *MA* fruit extract (Groups 3 and 4).

As shown in table 2 the plasma levels of CPK and LDH showed no significant difference among all experimental groups.

Since, the left ventricular end diastolic pressure (LVEDP) was adjusted within the range of 4–7 mmHg, it showed no significant difference between groups (Table 3), but left ventricular end systolic pressure (LVESP) showed a significant decrease in the diabetic rats (Group 2) compared to control group.

Table 1. The effect of *MA* fruit extract on plasma glucose and lipid profile of diabetic rats

Groups / Parameters	Control	Diabetic	MA (100 mg/kg)	MA (200 mg/kg)	Glibenclamide (0.5 mg/kg)
FBS (mg/dl)	113 ± 8	414 ± 30 ***	395 ± 62 ***	411 ± 29 ***	258 ± 21 *
HbA1c (% Hb)	6.05 ± 1.6	8.3 ± 1.8 **	8.9 ± 1.5 **	8.3 ± 1.7 **	6.4 ± 1.5
TC (mg/dl)	44 ± 3.5	73 ± 5 ***	61 ± 3.6 **	70 ± 5.1**	50 ± 5.7
TG (mg/dl)	24 ± 4.7	56 ± 11 **	43 ± 9 *	62 ± 6.2 **	32 ± 6.1
LDL (mg/dl)	23 ± 3.1	42 ± 6.1**	36 ± 3.5 *	38 ± 4.2 *	30 ± 3.4
HDL (mg/dl)	17.4 ± 2.3	14.4 ± 3.7 *	13.7 ± 3.8 *	12.6 ± 2.3 *	19 ± 4.3

Data Shown as mean ± standard error of mean (SEM)
FBS: fasting blood sugar, HbA1C: hemoglobin A1C, TC: total cholesterol, TG: triglyceride, LDL: low density lipoprotein, HDL: High density lipoprotein
* $P<0.05$, ** $P<0.01$ and *** $P<0.001$ versus to control group.

Table 2. The effect of *MA* fruit extract on plasma level of hepatic and cardiac enzymes of diabetic rats

Groups / Parameters	Control	Diabetic	MA (100 mg/kg)	MA (200 mg/kg)	Glibenclamide (0.5 mg/kg)
AST (IU/l)	113 ± 21	146 ± 29 *	180 ± 29 **	158 ± 26 **	134 ± 17
ALT (IU/l)	58 ± 6.9	122 ± 21 **	113 ± 18 **	127 ± 12 ***	101 ± 7.5 *
ALP (IU/l)	349 ± 58	1465 ± 173 **	1508 ± 260 **	1473 ± 90 **	811 ± 151 *
CK (IU/l)	717 ± 140	684 ± 79	866 ± 107	727 ± 142	670 ± 65
LDH (IU/l)	894 ± 279	743 ± 98	1035 ± 224	871 ± 219	844 ± 141

Data shown as mean ± SEM
AST: aspartate aminotransferase, ALT: alanine aminotransferase, ALP: alkaline phosphatase, CK: creatine kinase, LDH: lactate dehydrogenase
* $P< 0.05$, ** $P< 0.01$ and *** $P< 0.001$ versus to control group.

Treatment of diabetic rats with *MA* fruit extract at both doses (100 and 200 mg/kg) showed no significant effect on LVESP (Groups 3 and 4). It was significantly lower in diabetic rats treated with glibenclamide (Group 5) compared to the control group (Group 1).

Heart rate did have no significant difference in isolated hearts among all groups (Table 3). Compared to the control group, max dp/dt showed a significant decrease in the diabetic rats (Group 2), (Table 3). It was more reduced in diabetic rats treated with *MA* fruit extract at both doses of 100 and 200 mg/kg. Level of max dp/dt was lower in diabetic rats treated with glibenclamide and results showed no significance.

Table 3 shows that like max dp/dt, min dp/dt was much reduced in diabetic rats treated with both doses of *MA* 100 and 200 mg/kg. mindp/dt was lower in diabetic rats treated with glibenclamide, results showed no significant difference.

Table 4 showed that the heart rate decreased significantly in diabetic rats. It was more reduced in diabetic rats treated with both doses of *MA* as well as in those treated with glibenclamide.

RR, PR, QT and QTc intervals were increased in all diabetic rats. These showed more increase in rats treated with *MA* and glibenclamide. P and QRS duration showed no significant difference among all groups (Table 4).

Discussion

The main findings of this study showed that although *MA* fruit extract has antioxidant activity but did not improve the signs of diabetes and the diabetic-induced cardiotoxicity and hepatotoxicity in rats.

MA plant is often grown for shade in open situations such as parks and roadsides. Traditionally, the plant was used as an anthelmintic, tonic, antipyretic and to treat leprosy, eczema and asthma, however early reports mention that almost all parts of *MA* are toxic, especially ingestion of its fruits (5,6). Some studies reported cases of human death

Table 3. Cardiac hemodynamic parameters of isolated hearts of diabetic rats treated with *MA* fruit extract for 30 days.

Parameters \ Groups	Control	Diabetic	MA (100 mg/kg)	MA (200 mg/kg)	Glibenclamide (0.5 mg/kg)
Heart Rate (beats/min)	274 ± 19	256 ± 10	268 ± 22	246 ± 17	243 ± 17
Systolic duration (mmHg)	0.095 ± 0.010	0.110 ± 0.007 *	0.122 ± 0.013 **	0.125 ± 0.011 **	0.128 ± 0.015 **
Diastolic duration	0.128 ± 0.031	0.127 ± 0.029	0.114 ± 0.033	0.128 ± 0.029	0.132 ± 0.39
LVEDP (mmHg)	5.6 ± 1.42	5.00 ± 0.86	4.9 ± 2.15	5.1 ± 1.17	5.3 ± 1.01
LVDP (mmHg)	118 ± 11	87 ± 9.4 *	71 ± 10.6 **	85 ± 9.5 *	93 ± 7.4 *
Max dp/dt (mm Hg)	3458 ± 222	2503 ± 211 *	1938 ± 294 **	2155 ± 228 **	2998 ± 446
Min dp/dt (mmHg)	-2511 ± 181	-1759 ± 149 *	-1333 ± 227 *	-1601 ± 139 **	-1914 ± 197 *

Data shown as mean ± SEM

LVEDP: left ventricular end-diastolic pressure, LVDP: left ventricular developed pressure, Max: maximal pressure generation, Min: minimal pressure generation.

* $P< 0.05$, and ** $P< 0.01$ versus to control group.

Table 4. Cardiac electrical parameters of diabetic rats treated with *MA* fruit extract for 30 days.

Parameters \ Groups	Control	Diabetic	MA (100 mg/kg)	MA (200 mg/kg)	Glibenclamide (0.5 mg/kg)
Heart Rate (beats/min)	338 ± 21	271 ± 20 *	274 ± 16 *	244 ± 19 **	235 ± 17 **
RR interval (sec)	0.18 ± 0.03	0.23 ± 0.04 *	0.22 ± 0.03 *	0.25 ± 0.05 **	0.26 ± 0.05 **
PR interval (sec)	0.046 ± 00.3	0.049 ± 0.005	0.049 ± 0.006	0.050 ± 0.002 *	0.053 ± 0.003 *
P duration (sec)	0.017 ± 0.005	0.016 ± 0.003	0.018 ± 0.002	0.016 ± 0.002	0.018 0.002
QRS interval (sec)	0.018 ± 0.007	0.018 ±0.003	0.0016 ± 0.001	0.017 ± 0.005	0.017 ±0.003
QT interval (sec)	0.053 ± 0.007	0.070 ± 0.019 *	0.076 ± 0.006 *	0.075 ± 0.015 *	0.085 ± 0.011 **
QTc interval (sec)	0.127 ± 0.01	0.146 ± 0.04 *	0.161 ± 0.02 **	O.162 ± 0.02 **	0.173 ± 0.03 **
JT interval (sec)	0.027 ± 0.006	0.051 ± 0.02 *	0.060 ± 0.006 **	0.054 ± 0.02 *	0.067 ± 0.01 **

Data shown as mean ± SEM

* $P< 0.05$, ** $P< 0.01$ and *** $P< 0.001$ versus to control group.

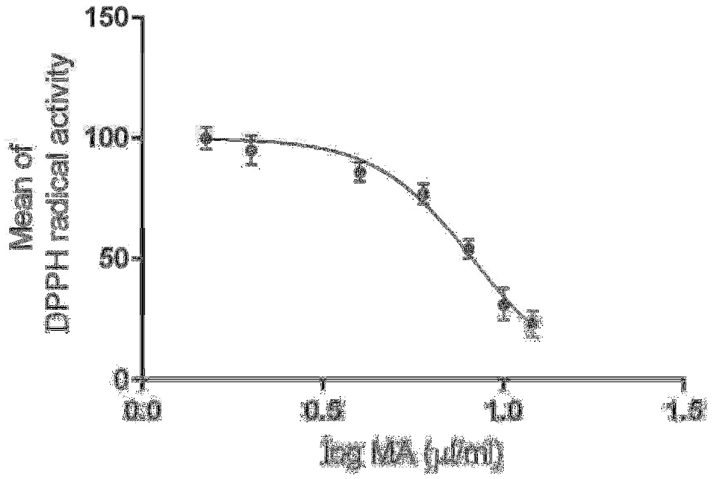

Figure 1. DPPH radical scavenging effect of *MA* fruit ethanolic extract with different concentrations

following ingestion of six to eight ripe fruits (5,8). *MA* poisoning was also reported in livestock for example pigs, cattle, sheep, goats and poultry (15-18). On the other hand, some experiments report that eating a large amount of *MA* fruit has no toxic effect. It appears these different toxic and none-toxic effects of *MA* may be related to the location of the Plant harvesting and/or a particular stage of plant growth (5,8). Recently, Saifue and co-workers reported that intera peritoneal administration of *MA* leaf extract has anti diabetic activity in ob/ob mice (20). In the present work, the effect of *MA* fruit extract was investigated in type 2 diabetic rats.

Present data shown that treatment of diabetic rats (induced by nicotinamide and STZ) with *MA* fruit extract at doses of 100 or 200 mg/kg for 30 days did not affect the signs of diabetes. However, glibenclamide as a conventional clinical drug significantly improved the signs of diabetes in diabetic rats. It has been reported that dried *MA* fruit has traditionally been used as a remedy to treat diabetes by some native inhabitants of Pakistan (22). In 2014, Khan and co–workers using C_2C_{12} myoblasts cells reported that all eleven pure compounds isolated from *MA* inhibited the protein-tyrosine phosphatase 1B (negative inhibitor of insulin signaling pathway). However, only two of eleven compounds stimulated the glucose uptake in C_2C_{12} myoblasts cells (7). These results are partly contrary to those in the present study. Also, the data of present work are also conflict with the results of Seifu and colleagues that reported the anti-diabetic effect of *MA* leaf extract on diabetic ob/ob mice (20). These differences might be attributed to the part of plant, the animal model and extraction method used.

Hepatic cell injury causes cellular enzymes such as AST, ALT, ALP and bilirubin are released into the interstitium and then into the blood (8). This effect may not be reversed to normal levels for days or weeks (23). The rate of plasma level of these enzymes correlated to the magnitude of hepatic injury (23). In the present study, induction of diabetes led to hepatic injury, evident with increased plasma level of hepatic enzymes. On the other hand, treatment of diabetic rats with 100 and 200 mg/kg *MA* fruit extract for 30 days showed no effect on plasma lipid profile and hepatic enzyme. Phua et al, in 2008, reported that five patients poisoned with *MA* fruit extract were referred to Taipei Veteran's General Hospital during 1998 and 2007. The most common signs of poisoning were weakness, ptosis and increased plasma level of ALT, AST and ALP

that recovered following treatment (6). A study that involved traditional Chinese medicine also reports that consumption of 6–9 of fruit, 30–40 of seeds or 400 gram of its bark leads to poisoning (6). Several studies in veterinary literature described toxicity of *MA* (15-18). However, two experimental studies report hepato-protective properties of *MA* leaf extract against simvastatin (8) and CCL4 (24) induced hepatotoxicity that was evident by restoration of the plasma level of hepatic enzyme to normal. It appears that these differences are related to the part of the plant and animal model used (8,24). In 2011, Fatah Al–Harmni and co–workers reported that treatment of male BALB/C infected by leishmanialdonovani with *MA* ripe fruit for 10–20 days had hepato-protective effect (23). So, their results are in opposition to those of this study that may be related to the animal species and model used.

The other findings of the present study indicate that treatment of diabetic rats with *MA* fruit extract worsened their cardiac functions. Similar results for electrocardiogram parameters were obtained that might make their hearts more susceptible to the incidence of arrhythmia. To our knowledge, there was no other animal or human study about heart and *MA*.

Previous studies have shown that the *MA* leaf extract had antioxidant effects against hydroxyl radicals, superoxide anions, nitric oxide radicals and DPPH radicals (19) that is agreement with our results as to *MA* fruit extract. Nevertheless, this extract partly exacerbated cardiac functions of diabetic rats as well as their cardiac electrical activity. This might have increased their susceptibility to arrhythmia that more research in the future.

Heart failure, the end stage of heart disease, is more prevalent in diabetic patients than in none diabetic patients (25). The incidence risk of cardiomyopathy is also more in such cases (25). Several pathophysiological changes at the microscopic levels including impaired myocardial relaxation, increased left ventricular stiffness, vascular endothelial dysfunction and impaired coronary flow reserve has been observed in diabetic patients(25). Although, epidemiology and mechanisms associated with heart failure induced by diabetic mellitus are well known, there is no known effective strategy for its early diagnosis and treatment. Findings of the present study show that treatment of diabetic rats with *MA* fruit extract might worsen the signs and symptoms of diabetes. It appears from results of the present study that none of the cardiac cells undertaken death, since the plasma level of CK showed any significant difference among all experimental groups. There is need future research on heart and *MA* in relation to various models. In this study, individual constituents of the extract were not determined, but it appears that their toxic effects may be attributed to meliatoxins A1, A2, B1 and B2 (5). Also Yuan and colleague in 2013 reported that limonoids in ethanolic extract of *MA* have cytotoxic effect (26).

One of the most obvious changes observed in this study was QTc prolongation in ECG of diabetic rats especially in groups treated with *MA* or glibenclamide. QT interval is from initiation of ventricular depolarization to the end of ventricular repolarization (27). It has been documented that QTc prolongation increases the risk of life–threatening arrhythmia and mortality in patients with diabetes and/or ischemic heart diseases (27,28). Thus, it appears *MA* fruit extract must be used with caution in diabetic patients.

Finally, these results showed that ethanolic extract of *MA* fruit had antioxidant activity against DPPH free radicals are consistent with those of previous research (19). It appears that the observed antioxidant activity might be related to high phenolic and flavonoids contents in the extract. One notable point is that the antioxidant activity of the extract could not prevent diabetic-induced tissues injury and even in some cases it resulted to more tissue injury like hepatic and myocardial injury. It seems that meliatoxins and limonoids present in the extract (5) led to more tissue injury that requires more research in the

future. Also, there is need for more research about the effects of location, climate and time of harvesting of herbs on its biological effects. Another explanation for the negative results of the present study could be attributed to the doses used in the present study. It seems that although the extract had antioxidant effect, the content of its toxic substances such as meliatoxins and limonoids were high and this might have exacerbated diabetic complications in rats.

Conclusions

The results of the present study showed that *MA* fruit did not have beneficial effects on the signs of diabetic and diabetic-induced cardiotoxicity and hepatotoxicity in rats. Then, more research need to be done about the effect of *MA* fruit extract on cardiac and hepatic functions in different models and now it should be used with caution in diabetic patients.

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