

Effects of *Passiflora edulis* on the Metabolic Profile of Diabetic Wistar Rat Offspring

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ABSTRACT Dry extract of the genus *Passiflora* has been shown to help control glycemia and lipid levels. The objective of this study was to evaluate the effects of passion fruit (*P. edulis*) on the biochemical profile of offspring from diabetic rats. Diabetes was induced by streptozotocin. The diabetes group consisted of 10 rats with glucose levels greater than 200 mg/dL; the nondiabetic (control) group consisted of 10 rats with glucose levels less than 120 mg/dL. After the diagnosis of diabetes, the mating phase was started. By day 21 of pregnancy, the offspring were born; the dams were kept in individual cages with their offspring until the weaning period. The offspring were then divided into 4 groups ($n = 15$ each): G1 were offspring from control dams, G2 were offspring from treated nondiabetic dams, G3 were offspring from diabetic dams, and G4 were offspring from treated diabetic dams. For 30 consecutive days, G1 and G3 offspring were treated with vehicle (oral gavage) and G2 and G4 offspring were treated with passion fruit juice (oral gavage). After 30-day treatment, the animals were anesthetized and killed, and blood was drawn immediately for analysis of the biochemical profile (total cholesterol, high-density lipoprotein cholesterol, low-density lipoprotein cholesterol, triglycerides, and glucose). The G2 and G4 rats showed significantly reduced total cholesterol, triglyceride, and low-density lipoprotein cholesterol levels and an increased high-density lipoprotein cholesterol level. The use of passion fruit juice improved lipid profiles, suggesting that this plant may have beneficial effects in the prevention and treatment of dyslipidemias and hyperglycemia.

KEY WORDS: • diabetes • dyslipidemias • glycemia • *Passiflora edulis* • rats

INTRODUCTION

DIABETES MELLITUS IS AN endocrine disease characterized by hyperglycemia. It results from total or partial deficiency of insulin production or the inability of this hormone to adequately exert its effects on target tissues.¹

A gestation is considered diabetogenic when insulin resistance occurs with increased serum levels of estrogen, prolactin, progesterone, cortisol, and chorionic somatomammotropin in an attempt to maintain a constant supply of glucose for the fetus. Elevation of maternal glucose levels causes fetal hyperglycemia and hyperinsulinemia, leading to increased perinatal morbidity and mortality and affecting the synthesis of lipids and proteins.^{2–6}

Diabetes mellitus causes approximately 10% of fetal malformations and is responsible for 40% of neonatal

deaths.^{7–9} Live newborns may already have metabolic alterations and higher risk for future complications. Epidemiologic studies show that in pregnancy, the lack of strict maternal glycemic control changes fetal metabolism, which leads to higher risk for metabolic alterations in fetuses, both in the gestational period and throughout their lives.^{4,5} Additionally, maternal hyperglycemia stimulates fetal growth (macrosomia) because of the greater availability of glucose in the bloodstream and by the regulation of growth factors.^{5,6,8} Greater weight at birth is related to the risk for developing insulin resistance, obesity, and type 2 diabetes mellitus in the future.¹⁰

Associated with diabetes mellitus are dyslipidemias, which are considered determining factors for the development of cardiovascular diseases. Elevated concentrations of plasma triglycerides, total cholesterol, and low-density lipoprotein (LDL) cholesterol along with decreased high-density lipoprotein (HDL) cholesterol levels lead to a group of risk factors called the metabolic syndrome, which increases the risk for vascular diseases.^{10,11}

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The treatment of diabetes mellitus and its complications is costly, and people often seek less expensive alternatives, such as medicinal plants. Use of these plants is an ancient human practice.

Many studies have shown that plants can be used to control diabetes mellitus and hypercholesterolemia. *Passiflora edulis* (tanga passion fruit or maracuja) is a popular remedy for treating anxiety, epilepsy, headache, and abdominal pain. The dry extract of *Passiflora* helps control glycemia and lipid levels. Ramos showed that *P. edulis* can reduce total and LDL cholesterol levels and Doyama *et al.* showed that *P. alata* increases HDL cholesterol levels in rats at a dose of 1000 mg/kg.^{12,13} Given the hypoglycemic effects of passion fruit, the objective of our study was to evaluate the effects of this plant on the biochemical profile of offspring of diabetic rats.

MATERIALS AND METHODS

Parental generation

Wistar rats were kept in the vivarium of Universidade Metodista de Piracicaba (Lins Campus), Brazil, under controlled conditions (12-hour/12-hour light/dark cycle, mean ambient temperature \pm standard deviation of $22^{\circ}\text{C} \pm 2^{\circ}\text{C}$, relative humidity of $60\% \pm 5\%$, and water and chow *ad libitum*). Rats were treated according to the "Guide to the Care and Use of Experimental Animals" of the Canadian Council on Animal Care. The study was initiated after its approval by the ethics committee under registration number 2500000764/2007-47.

Diabetes Induction

Nondiabetic rats ($n=20$) weighing approximately 250 g underwent a 7-day adaptation period in the room where the experiment was designed. After this period, randomly selected rats ($n=10$) underwent intravenous administration (through the caudal vein) of 40 mg of streptozotocin per kg (Sigma Chemical Co.) diluted in citrate buffer (0.1 M; pH, 4.5). Nondiabetic animals ($n=10$) received only the vehicle (citrate buffer) in a volume that was equivalent to that of the diabetogenic drug.¹⁴ Blood glucose levels were measured by using a glucose meter (OneTouch Ultra, Johnson & Johnson) 7 days after the induction of diabetes. As an inclusion criterion, rats were considered to be diabetic if the glucose level was greater than 200 mg/dL and nondiabetic if the glucose level was less than 120 mg/dL.

Mating period

One week after diabetes induction, each of the 4 female rats was mated overnight to nondiabetic male rats. The morning in which sperm was found in the vaginal smear was designated gestational day 0.^{15,16}

Pregnancy period

In the morning on days 0, 7, 14, and 21 of pregnancy, a blood drop was collected from the caudal vein for glycemia

measurement by using a glucose meter. By day 21 of pregnancy, after birth of offspring, the dams remained in individual cages with their offspring until the weaning period (21 days).

Offspring

After weaning, the offspring (60 males) were kept in collective cages until they reached adulthood. The adult animals, weighing approximately 250 g, were divided into 4 experimental groups ($n=15$ animals per group): G1 were offspring from nondiabetic dams treated with vehicle (water), G2 were offspring from nondiabetic dams treated with *P. edulis* (passion fruit) juice, G3 were offspring from diabetic dams treated with vehicle (water), and G4 were offspring from diabetic dams treated with *P. edulis* juice. The juice and the vehicle were administered by oral gavage.

The animals in groups G2 and G4 received passion fruit juice at a dose of 0.58 g/kg once a day (early morning) for 30 consecutive days. The dose administered to the animals was based on 200 g/L, which corresponds to the daily intake of 200 mL of juice by an adult man weighing 70.0 kg (this amount was based on folk medicine consultation about preparing the juice). Animals in the control group received a similar volume of water.

Passion fruit juice preparation

Passion fruits were obtained from Agricultural Cooperative of Lins-SP (COALINS). They were washed and cut, and the pulp was weighed (200g/L) and triturated with water in a blender for 1 minute. The juice was filtered and frozen in amber flasks. The preparation of the juice was based on folk medicine consultation. Each flask was thawed daily at ambient temperature 2 hours before administration.

Blood collection and determination of biochemical profile

At the end of the treatment period (30 days), the animals were anesthetized with sodium pentobarbital (150 mg/kg) and killed. Blood samples were collected from the cava vein to determine the biochemical profile. The tests were performed according to the methods proposed by commercial kits: LABTEST (Lagoa Santa) for glycemia, total cholesterol, HDL cholesterol, and triglycerides and WIENER LAB for LDL cholesterol (values in mg/dL). The results were interpreted according to criteria established by the American Diabetes Association.¹

Statistical analysis

Data were analyzed by using the Student *t*-test, and the level of significance adopted was 5% (BIOESTAT 5.0).

RESULTS

Rats in group G3 (offspring from diabetic dams that received water) showed significantly higher glucose, cholesterol, and triglyceride levels and lower HDL cholesterol levels than group G1 (offspring from nondiabetic dams that received water) (Table 1). These data indicate that adult

TABLE 1. BIOCHEMICAL PROFILE FROM NONDIABETIC OFFSPRING TREATED WITH VEHICLE (G1) AND DIABETIC OFFSPRING TREATED WITH VEHICLE (G3)

| Variable | Groups | | P value |
|-----------------|---------------------------|---------------------------|---------|
| | G1 | G3 | |
| Glucose | 60.7 ± 11.9 ^a | 88.9 ± 11.6 ^b | .0008 |
| Cholesterol | 76.8 ± 17.35 ^a | 92.7 ± 4.7 ^b | .0469 |
| Triglycerides | 119.9 ± 12.4 ^a | 149.0 ± 12.5 ^b | .04701 |
| HDL cholesterol | 34.4 ± 3.33 ^a | 30.4 ± 4.1 ^b | .0175 |
| LDL cholesterol | 65.0 ± 19.5 ^a | 84.5 ± 17.1 ^b | .0693 |

Unless otherwise noted, values are expressed as mean ± standard deviation.
^{ab}Mean values and standard deviation followed by same superscript letter do not differ between groups G1 and G3.
 HDL, high-density lipoprotein; LDL, low-density lipoprotein.

offspring from diabetic mothers have higher glycemic and lipid profiles than the offspring from nondiabetic mothers.

Table 2 shows that offspring from nondiabetic dams treated with *P. edulis* juice (G2) had significantly decreased cholesterol, triglyceride, and LDL cholesterol levels and significantly increased HDL cholesterol levels compared with the offspring of nondiabetic dams that received water (G1). These results show that the use of *P. edulis* juice may have beneficial effects on the lipid profile of offspring from nondiabetic dams.

Rats in group G4 (offspring from diabetic dams treated with *P. edulis*) showed significantly reduced levels of glucose, cholesterol, triglycerides, and LDL cholesterol and increased HDL cholesterol values compared with rats in group G3 (offspring from diabetic dams that received water) (Table 3). The treatment with *P. edulis* decreased lipid and glucose levels in the offspring of diabetic mothers when compared with the offspring of diabetic mothers that received vehicle.

Table 4 shows that G4 (offspring from diabetic dams treated with *P. edulis*) had lower HDL cholesterol values than group G2 (offspring from nondiabetic dams treated with *P. edulis*). Glucose, cholesterol, triglyceride, and LDL cholesterol levels did not significantly differ between these groups.

TABLE 2. BIOCHEMICAL PROFILE FROM NONDIABETIC OFFSPRING TREATED WITH VEHICLE (G1) AND NONDIABETIC OFFSPRING TREATED WITH PASSION FRUIT JUICE (G2)

| Variable | Groups | | P value |
|-----------------|---------------------------|--------------------------|---------|
| | G1 | G2 | |
| Glucose | 60.7 ± 11.9 ^a | 65.4 ± 8.3 ^a | .3276 |
| Cholesterol | 76.8 ± 17.35 ^a | 55.1 ± 13.8 ^b | .0011 |
| Triglycerides | 119.9 ± 12.4 ^a | 74.1 ± 16.7 ^b | .0326 |
| HDL cholesterol | 34.4 ± 3.33 ^a | 52.4 ± 7.4 ^b | .0000 |
| LDL cholesterol | 65.0 ± 19.5 ^a | 46.5 ± 7.5 ^b | .0327 |

Unless otherwise noted, values are expressed as mean ± standard deviation.
^{ab}Mean values and standard deviation followed by same superscript letter do not differ between groups G1 and G2.

TABLE 3. BIOCHEMICAL PROFILE FROM DIABETIC OFFSPRING TREATED WITH VEHICLE (G3) AND DIABETIC OFFSPRING TREATED WITH PASSION FRUIT JUICE (G4)

| Variable | Groups | | P value |
|-----------------|---------------------------|--------------------------|---------|
| | G3 | G4 | |
| Glucose | 88.9 ± 11.6 ^a | 66.9 ± 8.8 ^b | .0000 |
| Cholesterol | 92.7 ± 4.7 ^a | 54.1 ± 24.8 ^b | .0332 |
| Triglycerides | 149.0 ± 12.5 ^a | 72.9 ± 29.3 ^b | .0225 |
| HDL cholesterol | 30.4 ± 4.1 ^a | 34.7 ± 3.3 ^b | .0244 |
| LDL cholesterol | 84.5 ± 17.1 ^a | 54.2 ± 14.1 ^b | .0133 |

Unless otherwise noted, values are expressed as mean ± standard deviation.
^{ab}Mean values and standard deviation followed by same superscript letter do not differ between groups G3 and G4.

DISCUSSION

In this study, the adult offspring from diabetic dams had a higher glycemic profile than the offspring from normoglycemic dams. This finding indicates that maternal hyperglycemia is related to hyperglycemia or insulin resistance in the offspring when they are adults. Maayan-Metzger *et al.*³ showed that the main risk factors for developing hypoglycemia in the first day of life were large size for gestational age and maternal type 1 diabetes mellitus. The young offspring who were born to mothers with diabetes mellitus were heavier, large for their gestational age, and had more severe hypoglycemia in the first day of life compared with infants born to mothers with nonsevere diabetes mellitus.

The lipid profile of offspring from diabetic dams also showed higher mean values than that of the offspring from nondiabetic dams. This may indicate a risk for future alterations. Deviations in glycemia and dyslipidemias are risk factors for metabolic syndrome and vascular disorders.¹⁷

Our results suggest that passion juice can be helpful in the treatment or prevention of diabetes mellitus and its complications in the offspring of mothers with diabetes: The animals that received the juice had reductions in glycemia and lipid profile. Ramos *et al.*¹² studied a flour made with passion fruit and also found beneficial effects on the lipid profile.

TABLE 4. BIOCHEMICAL PROFILE FROM NONDIABETIC OFFSPRING TREATED WITH PASSION FRUIT JUICE (G2) AND DIABETIC OFFSPRING TREATED WITH PASSION FRUIT JUICE (G4)

| Variable | Groups | | P value |
|-----------------|--------------------------|--------------------------|---------|
| | G2 | G4 | |
| Glucose | 65.4 ± 8.3 ^a | 66.9 ± 8.8 ^a | .3606 |
| Cholesterol | 55.1 ± 13.8 ^a | 54.1 ± 24.8 ^a | .0540 |
| Triglycerides | 74.1 ± 16.7 ^a | 72.9 ± 29.3 ^a | .4576 |
| HDL cholesterol | 52.4 ± 7.4 ^a | 34.7 ± 3.3 ^b | .0000 |
| LDL cholesterol | 46.5 ± 7.5 ^a | 54.2 ± 14.1 ^a | .1578 |

Unless otherwise noted, values are expressed as mean ± standard deviation.
^{ab}Mean values and standard deviation followed by same superscript letter do not differ between groups G2 and G4.

Other authors have observed biochemical benefits with use of plants. Bera *et al.*¹⁸ found that a polyherbal formulation composed of 8 medicinal plants that was used for the management of streptozotocin-induced diabetes in rats had antidiabetic effects. glibenclamide. Babiloni *et al.*¹⁹ showed antihyperglycemic activity of stem-bark extract of *Vatairea macrocarpa* in the treatment of diabetic rats. Umar *et al.*²⁰ observed that *Tetracera scandens* had antidiabetic efficacy in diabetic rats. Tao *et al.*²¹ showed significant reduction in glucose, cholesterol, and triglyceride levels by using a traditional antidiabetic formula prepared with many plants. Salahuddin and Jalalpure²² observed a reduction in blood glucose levels and a reduction in lipid profile after using extract of *Curcumis trigonus* in diabetic rats. Adeneye and Adeyemi²³ and Daisy *et al.*²⁴ found decreases in lipids and glycemia with use of *Hunteria umbellata* and isolated components of *Gymnema sylvestre*, respectively.

Many other studies showed benefits of plants in the biochemical profile of rats with streptozotocin- or aloxan-induced diabetes, such as *Polymnia sonchifolia*,²⁵ conophylline-containing plant extract,²⁶ fruits of *Musa* (Chenkadali),²⁷ *Artemisia sphaerocephala*,²⁸ *Lagenaria siceraria*,²⁹ and *Allium sativum*.³⁰ Helmstädter and Schuster³¹ observed antidiabetic effects of *Vaccinium myrtillus*. Other studies also showed that certain plants have hypolipidemic effects (*Coriandrum sativum*, *Mentha piperita*, asparagus roots, *Allanblackia floribunda*, *Ficus religiosa*, and *Amorphophalus konjac*) and can be used to reduce biomarkers for heart disease (*Origanum onites* and berries).^{32–36}

The use of passion fruit did not lead to a significant difference in the glycemic profile of the offspring of nondiabetic rats. This is a positive result because in the absence of glycidic disorders, there is no need for the reduction of glucose levels. In contrast, the offspring of diabetic mothers had a significantly reduced glycemic profile when treated with this plant. Ramos *et al.*¹² also observed a reduction in glycemia with the use of *P. edulis* in individuals with altered glucose metabolism. These findings demonstrate that this plant can be used in the control of glycemia in patients at risk for developing diabetes mellitus and its complications (including vascular damage).

The positive effects of passion fruit may be related to the compounds in its pulp. *Passiflora* species are rich in pectin, minerals, carotenoids, vitamin C, and flavonoids. Studies have shown that pectins can reduce total cholesterol, triglyceride, and LDL cholesterol levels and increase HDL cholesterol levels.³⁸ Fibers might reduce plasma lipid levels by increasing the excretion of cholesterol and biliary acids in feces. This effect on lipids reduces vascular damage and heart disease.^{39–41} Yapo *et al.*⁴² investigated the fiber content in *P. edulis* juice and found that the total dietary fiber from yellow passion fruit rind was greater than 73% dry matter, of which insoluble dietary fiber accounted for greater than 60% (w/w). The method they used also revealed that nonstarch polysaccharides were the predominant components (approximately 70%, w/w).

The antioxidant properties of vitamins and flavonoids are described as having positive effects in the treatment and

prevention of dyslipidemias and hyperglycemia.⁴³ *P. incarnata* reportedly contains glycosyl flavonoids,^{44,45} and *P. edulis* has antioxidant^{46,47} and anti-inflammatory properties.^{48,49} These properties may explain its effects on glycemia and the lipid profile in the animals we studied. The antioxidant compounds of *P. edulis* are 16 apigenin or luteolin derivatives, which include 4 mono-C-glycosyl, 8 O-glycosyl-C-glycosyl, and 4 O-glycosyl derivatives. Moreover, the uncommon C-deoxyhexosyl derivatives of luteolin and apigenin have been identified.⁵⁰

CONCLUSION

The use of *P. edulis* improved the lipid and glycemic profile of offspring from diabetic and nondiabetic mothers of Wistar rats. This result suggests that this plant has potential as an adjuvant in the prevention and treatment of dyslipidemia and hyperglycemia. However, the effects in humans must be studied. Passion fruit, which is widely consumed, is easy to find at accessible prices, making it potentially interesting for medicinal purposes.

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AUTHOR DISCLOSURE STATEMENT

No competing financial interests exist.

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