Alexandria Journal of Medicine

ISSN: 2090-5068 (Print) 2090-5076 (Online) Journal homepage: https://www.tandfonline.com/loi/tajm20

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**To cite this article:** Alejandra Rojas-Olivos, Rodolfo Solano-Gómez, Alfonso Alexander-Aguilera, Manuel Jiménez-Estrada, Stefan Zilli-Hernández & Luicita Lagunez-Rivera (2017) Effect of *Prosthechea karwinskii* (Orchidaceae) on obesity and dyslipidemia in Wistar rats, Alexandria Journal of Medicine, 53:4, 311-315, DOI: 10.1016/j.ajme.2016.11.004

**To link to this article:** https://doi.org/10.1016/j.ajme.2016.11.004



Published online: 17 May 2019.

Alexandria Journal of Medicine 53 (2017) 311–315

HOSTED BY

Contents lists available at ScienceDirect

Alexandria Journal of Medicine

journal homepage: http://www.elsevier.com/locate/ajme

Short Communication

# Effect of *Prosthechea karwinskii* (Orchidaceae) on obesity and dyslipidemia in Wistar rats

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ARTICLE INFO

*Article history:*
Received 29 July 2016
Revised 24 November 2016
Accepted 24 November 2016
Available online 2 January 2017

*Keywords:*
Antioxidant activity
Cholesterol
Diabetes
Glucose
Medicinal plants
Triglycerides

ABSTRACT

*Background:* *Prosthechea karwinskii* is an endemic Mexican orchid, it's currently used as decorative element and in the traditional medicine to treat diabetes and some problems related to inflammatory processes.
*Aim:* To determine antioxidant activity index (AAI) and to validate by the first time and through an rat model the hydroalcoholic extract obtained from *Prosthechea karwinskii*, a plant used in traditional medicine for treat conditions relate to the metabolic syndrome.
*Methods:* For *in vivo* assays 25 weaned male Wistar rats were divided into a control group (CG; n = 5) and a Metabolic Syndrome group (MS; n = 20). The rats of the latter were induced to MS with 40% sucrose in the drink water during 13 weeks. After MS induction this group was subdivided into 4 groups: MS group (n = 5) received sucrose, and three groups receiving 200 mg/kg of body weight of each extract pseudobulb (P, n = 5), leaf (L, n = 5), and flower (F, n = 5). All treatments were followed for 13 days. Blood was collected at the end of the study to measure glucose, cholesterol and triglycerides. AAI were measured in the extracts by the method of DPPH. The results were analyzed using MINITAB 16.1.0, and the statistical significance was determined by ANOVA and a Tukey's test ($P < 0.05$).
*Results:* Leaves (L) extract had highest values in AAI, followed by flowers (F) and pseudobulb (P) extracts. Leaves extract had highest reducing effect on glucose level, while flower extract had highest reducing effect on the cholesterol and triglycerides levels.
*Conclusions:* The *P. karwinskii* extracts evaluated here reduces the glycemic and lipidemic parameters in Wistar rats with MS induced. These effects may be attributed to the high antioxidant capacity of the extracts.


## 1. Introduction

Medicinal plants play an important role in the introduction of new therapeutic agents as a source of biologically active substances with antihyperlipidemic and antihyperglycemic properties, among other effects.[1] Hyperglycemia results in an increased oxidative stress due to reduced endogenous antioxidants in the body[2] and imbalance in adipose tissue, influencing lipid regulation and triggering cardiovascular disease. Metabolic syndrome (MS) is the term used to designate a set of interrelated conditions that include hyperglycemia, hyperlipidemia, obesity and hypertension.[3–6] In traditional medicine, plants with biological activity affecting metabolic disturbances related to the pathophysiology of MS have been evaluated, including species from Orchidaceae family. Asian orchids such as *Nervilia plicata* (Andrews) Schltr.,[7] *Dendrobium chrysotoxum* Lindl.[8] and *Dendrobium denneanum* Kerr.,[9] have shown to have hypoglycemic effects. In Mexico some orchid species have also been evaluated for conditions related to MS; *Scaphyglottis fasciculate* Hook. for its potential relaxing effect on cardiac contractions,[10] *Laelia autumnalis* (La Llave & Lex.) Lindl. for its antihypertensive effect,[11] *Laelia anceps* Lindl. for its antihypertensive and vasorelaxant effects,[12] and *Prosthechea michuacana* (La Llave & Lex.) W.E. Higgins for its hypoglycemic activity.[13]

Peer review under responsibility of Alexandria University Faculty of Medicine.
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http://dx.doi.org/10.1016/j.ajme.2016.11.004
2090-5068/

An endemic orchid to the mountains of southern Mexico that is used in the traditional medicine is *Prosthechea karwinskii* (Mart.) J. M.H. Shaw. Different parts of this orchid are used as remedy to treat hyperglycemia (pseudobulb and leaves), cough (pseudobulb and flowers), burns (pseudobulb), and to prevent miscarriages (flowers).[14] The flowering of this species coincides with Easter celebrations and therefore this orchid is also used in religious decorations at homes and churches.[14–16] A previous phytochemical study[17] of our research group has identified the presence of phenolic constituents in this orchid, like tyrosol, apigenin-7-glucoside, caffeic acid, vainillin, *p*-cumaric acid and ferulic acid. These compounds are known by their cardioprotective effects[18,19] due to its ability to inhibit cholesterol oxidation. Furthermore, phenolic compounds can inhibit adipose tissue growth due to their antiangiogenic activity and their ability to regulate adipocyte metabolism.[20]

The goal of this research was to determine the antioxidant activity and to validate, by the first time, el traditional use of *Prosthechea karwinskii* in the treatment of some conditions relate to the metabolic syndrome, for which some parts of the plant are employed (pseudobulb, leaf, or flower). For this, the plant extracts were obtained with ethanol-deionized water and then were evaluated in a rat model.

## 2. Materials and methods

### 2.1. Plant material and extracts

Plant material was collected in 2012 and 2013 from specimens that were used as Easter decorations in Villa de Zaachila, Oaxaca. Additional material was collected in San Pedro and San Pablo Teposcolula with the permission of the local authority. Taxonomic determination of the plant was done and pressed specimens were prepared and deposited in OAX Herbarium of the Instituto Politécnico Nacional (Solano 4037). Since in traditional medicine is known that each part of this orchid is used separately for particular condition[14], the plant material was separated into pseudobulbs, leaves and flowers; each portion was dried, pulverized, and stored at room temperature (R.T.) until use. Ten grams of each dried part were placed in 400 ml of ethanol-deionized water solution (1:1, w/v) for 7 days at R.T., frequently stirred, then filtered and concentrated by solvent evaporation at R.T. The yield of the extraction for each plant part was: 18.8% for pseudobulbs, 16.8% for leaves, and 33.6% for flower. The sticky extracts obtained were placed at R.T. until it use.

### 2.2. Analysis of antioxidant activity index

Antioxidant capacity of the extracts was determined by the 2,2-diphenyl-1-picrylhydrazyl radical (DPPH·) method; antioxidant activity index (AAI) and $IC_{50}$ values were determined according to Sherer and Godoy.[21]

### 2.3. Sucrose-induced metabolic syndrome model

A total of 25 weaned male Wistar rats of 21 days age and 150–200 g body weight (Table 2) were individually housed and maintained in a 12-h light/dark cycle at 25 °C. This study was approved by the Research Committee of the Universidad Cristobal Colón following the guidelines of Mexican legislation, NOM-062-ZOO-1999[22] for the care and use of laboratory animals. Animals were divided into two groups: the control group (CG; n = 5) was given a standard diet (Lab Diet 2004 S, Harlan Teklad Inc.) and water *ad libitum*; the Metabolic Syndrome group (MS; n = 20) which was given the same standard diet plus 40% sucrose in the drinking water *ad libitum* for 13 weeks to induce MS.

### 2.4. Experimental diet and co-treatment

After MS induction this group (n = 20) was subdivided into 4 groups: MS group (n = 5) received a high caloric sucrose diet, P group (n = 5) received pseudobulb extract, L group (n = 5) received leaf extract, and F group (n = 5) received flower extract; these groups were compared with the control group (CG) as was mentioned above. P, L. and F groups received during 13 days by oral via, through a standard esophageal cannula, 200 mg/kg of body weight of the corresponding evaluated extract dissolved in water. Animals from all groups received the previously mentioned standard diet, during the same period of 13 days; additionally, for the MS group its diet included 40% sucrose in the drinking water *ad libitum*. At the end of the treatment the final weight was registered and a blood sample was taken from each rat with 18-h-fasted using cardiac puncture under anesthetic condition, prior to the killing of the animals. The blood was centrifuged and serum was kept at −20 °C until use.

### 2.5. Adipose tissue and Biochemical parameters

At the end of the treatment, rats were killed with anesthesia (0.1 ml intraperitoneal of 1% sodium barbiturate) to obtain the abdominal adipose tissue, epididymal and pericardial fat, and to determine the serum levels of glucose, cholesterol and triglyceride. These parameters were determined by enzymatic-colorimetric methods according to the manufacturer's instruction using a biochromatic analyzer model Vitalab Selectra E.

### 2.6. Data analysis

Data obtained from the serum parameters, weight gain, organs and adipose tissue are presented as the mean ± SD. Data was evaluated using Minitab 16.1.0 software. Statistical significance was determined with an analysis of variance, a Tukey's multiple range test was performed to test for significant differences between the different treatment groups. For all analyses the level of significance was $P < 0.05$.

## 3. Results

### 3.1. Total flavonoids and antioxidant activity of Prosthechea karwinskii extracts

Table 1 presents the concentration required to inhibit 50% of free radicals ($IC_{50}$), antioxidant activity index (AAI), and total flavonoid content of the extracts administered for biological evaluation.

The highest antioxidant capacity was showed by the extract of leaves (AAI = 5.7), followed by that from flowers (AAI = 1.276), and pseudobulbs (AAI = 0.925).

### 3.2. Caloric intake induced MS model

The daily caloric intake of the MS group compared with that of the control group (CG) during the 13 weeks MS induction period is

**Table 1**
Antioxidant capacity of *Prosthechea karwinskii* extracts.

| Extract | Yield w/w (%) | $IC_{50}$ | AAI |
|---|---|---|---|
| Pseudobulb | 18.8 ± 1.335 | 43.06 ± 7.311 | 0.925 ± 0.162 |
| Leaf | 16.8 ± 2.335 | 6.91 ± 0.187 | 5.7 ± 0.157 |
| Flower | 33.6 ± 2.93 | 30.85 ± 2.51 | 1.276 ± 0.971 |

$IC_{50}$: 50% inhibitory concentration. AAI: antioxidant activity index = [DPPH]/$IC_{50}$.

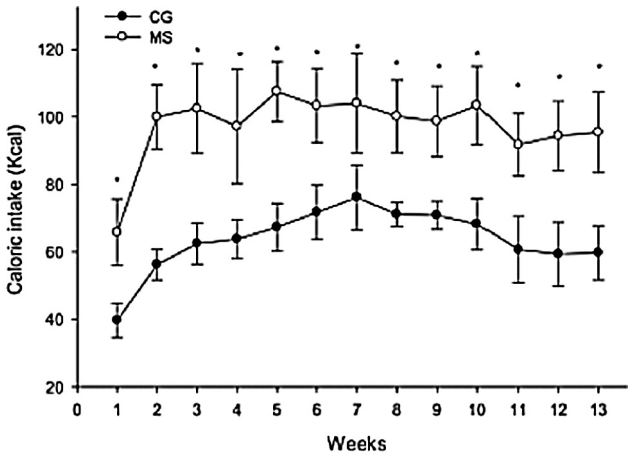


**Fig. 1.** Caloric intake during Metabolic Syndrome development. CG: control group, MS: sucrose diet, kcal: kilocalories. The values represent the mean ± DS $^{*}P < 0.05$.

shown in Fig. 1. The MS group showed significant differences from the CG due to sucrose consumption.

Table 2 shows the caloric intake of CG, MS, P, L and F groups during 13 days of treatment consisting in the administration of the extracts and sucrose water. The initial weight for each evaluated group was nonuniform because evaluation of extracts began after induction of the animal model metabolic syndrome with 40% sucrose in drinking water; also, it must be considered the variation presented in the caloric intake of the animals during the development of MS, as is shown in Fig. 1.

Body weight of the CG increased by 1.1% while the body weight of the MS group increased by 5.6%. After the treatment, the P group lost the most weight, 7.8% of the total weight. Body weight of the L group decreased by 5.2% whereas that of the F group decreased by 5%, these results were respect to MS group.

### 3.3. Effect of Prosthechea karwinskii extracts in adipose tissue and serum parameters

Table 3 shows the effect of *P. karwinskii* extracts on abdominal, epididymal and pericardial adipose tissue weight over the 13 days experimental treatment.

In comparison with the MS group, the other groups experienced significant decreases in total adipose tissue: 49% in the P group, 36% in the L group, and 33% in the F group. According to the serum parameters of the CG and the MS group, glucose, cholesterol and triglycerides levels decreased in three groups: P, L and F (Table 4).

The leaf extract (L) had greatest effect in decrease the fasting glucose levels (64.12 mg/dl) in comparison with the MS group (135.26 mg/dl). The other extracts (F and P) also decreased glucose levels to 73.72 and 92.20 mg/dl, respectively. The flower extract (F) had the greatest effect on cholesterol levels (53.89 mg/dl) compared to the MS group (87.52 mg/dl); cholesterol levels in the CG were 69.43 mg/dl. P and L extracts also decreased this parameter but in a lesser extent: 75.55 mg/dl and 76.04 mg/dl, respectively. The extracts also had a reductive effect on triglyceride levels compared to the CG and MS groups, 214.28 and 259.28 mg/dl, respectively. The F group had the lowest triglycerides level (74.58 mg/dl), followed by L (108.78 mg/dl) and P (127.75 mg/dl) groups (Table 4).

## 4. Discussion

This work reveals by the first time that the pseudobulb, leaf and flower hydroalcoholic extracts from *P. karwinskii* ameliorated key parameters of the MS in a rat model and their relation with antioxidant capacity of the extracts. At the end of the treatment, the group given pseudobulb extract lost the greatest percentage of body weight, followed by the groups that received leaf and flower extracts. The results indicate that the evaluated extracts, administered in a dose of 200 mg/kg/day, significantly decreased adipose tissue mass and serum parameters levels that are associated with MS. The extracts evaluated here, not only reduced the adipose tissue mass but glucose levels as well; in contrast the results reported for aqueous extract from *Ilex* paraguariensis A. St.-Hil.[23] reduced abdominal and epididymal adipose tissue too, but raised glucose levels during 30 days of treatment. As the adiposity index data reveals, *Prosthechea karwinskii* extracts also had a greater effect on reducing adipose tissue than the previously reported for *Salacia reticulate* Wight[24] and *Citrus grandis* (L.) Osbeck.[25]

The extracts also had a lowering effect on cholesterol and triglycerides levels; in this case, the flower extract had a greater effect than the reported for *Camellia sinensis* (L.) Kuntze.[26] Evaluation of *Citrus grandis* hydroalcoholic extract[25] indicates that after two weeks of treatment with a higher administered dose, cholesterol levels remained higher than our results. The dosage of leaf extract evaluated here had a greater glucose lowering effect than that reported for the polysaccharide obtained from *Dendrobium chrysotoxum* pseudobulbs[8] using dosage of 200 mg/kg/day and 500 mg/kg/day. The leaf extract evaluated here also has a better dampening effect than the reported for *Dendrobium denneanum* polysaccharide[9] using a dose of 300 mg/kg/day.

The antihyperglycemic effect of leaf extract in this work could be attributed to its greater antioxidant capacity compared with that for pseudobulb and flower extracts. Since hyperglycemia is associated with diminished endogenous antioxidants and increased oxidative stress, antioxidants have been shown to reduce the risk of hyperglycemia improving glucose disposal in the body.[5,27,28] According to the methodology described by Sherer and Godoy,[21] these results show that *P. karwinskii* extracts have a higher AAI (Table 1) than other species reported in the following studies: ethanol-water leaves extract from *Dendrobium speciosum* Sm.[29] with IAA of $3.46 * 10^{-5}$, the hydromethanolic and chloroform pseudobulb extracts from *Prosthechea michuacana* with an AAI of 0.0901 and 0.1268, respectively,[13] the hydrometanolic rhizomes extract from *Curculigo orchioides* Gaertn.[30] with an AAI = 0.3720, the hydrometanolic leaf extract of *Juglans regia* L.[31] with an AAI = $1.98 * 10^{-9}$, and *Jasminum humile* L.[32] with an AAI = 0.5598.

**Table 2**
Caloric intake and body weight of rats under the effect of *P. karwinskii* extracts after 13 days of experimental treatment.

| Parameter | CG | MS | P | L | F |
|---|---|---|---|---|---|
| kcal/day (B) | 65.10 ± 19.53 | 125.68 ± 22.62[a] | 87.43 ± 2.38 | 91.27 ± 10.38 | 81.30 ± 9.26 |
| Initial weight (g) | 372.0 ± 70.14 | 440.0 ± 83.19 | 321.67 ± 96.3 | 385.0 ± 69.28 | 431.67 ± 23.63 |
| kcal/day (E) | 79.05 ± 6.48 | 139.42 ± 6.77[a] | 128.35 ± 6.47[a] | 147.93 ± 31.13[a] | 147.78 ± 13.96[a] |
| Final weight (g) | 376.0 ± 77.73 | 464.50 ± 44.04 | 296.67 ± 88.8 | 365.0 ± 73.65 | 410.0 ± 20.00 |

CG: control group, MS: sucrose diet, P: sucrose diet and pseudobulb extract, L: sucrose diet and leaf extract, F: sucrose diet and flower extract, B: Beginning of the treatment, E: End of the treatment. The values represent the mean ± SD, superscript values show statistically significant difference as revealed by the Tukey's test.
[a] $P < 0.05$.

**Table 3**
Abdominal, epididymal and pericardial adipose tissue weight and adiposity index after co-treatment with *P. karwinskii* extracts.

| Parameter (g) | GC | MS | P | L | F |
|---|---|---|---|---|---|
| AT abdominal | 12.20 ± 1.92 | 16.25 ± 5.91 | 9.33 ± 3.78[a] | 11.66 ± 2.30 | 11.0 ± 1.73[a] |
| AT epididymal | 10.0 ± 2.55 | 15.83 ± 6.67 | 6.66 ± 2.08[a] | 8.33 ± 2.88[a] | 10.33 ± 0.57[a] |
| AT pericardial | 0.62 ± 0.40 | 0.48 ± 0.18 | 0.70 ± 0.26 | 0.56 ± 0.30 | 0.40 ± 0.10 |
| Total fat | 22.82 ± 4.87 | 32.56 ± 12.76 | 16.69 ± 6.12[a] | 20.55 ± 5.48[a] | 21.73 ± 2.4[a] |

AT: adipose tissue, GC: control group, MS: sucrose diet, P: sucrose diet and pseudobulb extract, L: sucrose diet and leaf extract, F: sucrose diet and flower extract. The values represent the mean ± SD, superscript values show statistically significant difference as revealed by the Tukey's test.
[a] $P < 0.05$.

**Table 4**
Variation of serum parameters in Wistar rats after 13 days of treatment with *P. karwinskii* extracts.

| Parameter (mg/dl) | CG | MS | P | L | F |
|---|---|---|---|---|---|
| Glucose | 96.80 ± 10.64[a] | 135.26 ± 30.70 | 92.20 ± 7.95[a] | 64.12 ± 11.39[a] | 73.72 ± 25.76[a] |
| Cholesterol | 69.43 ± 10.39[a] | 87.52 ± 7.44 | 75.55 ± 0.32[a] | 76.04 ± 9.59[a] | 53.89 ± 10.54[a] |
| Triglycerides | 214.28 ± 18.07[a] | 259.28 ± 23.89 | 127.75 ± 46.56[a] | 108.78 ± 11.30[a] | 74.58 ± 22.35[a] |

CG: control group, MS: sucrose diet, P: sucrose diet and pseudobulb extract, L: sucrose diet and leaf extract, F: sucrose diet and flower extract. The values represent the mean ± SD, superscript values show statistically significant difference as revealed by the Tukey's test.
[a] $P < 0.05$.

Furthermore, this study confirms the potential antioxidant activity of compounds previously identified in *P. karwinskii* by Mijangos-Ricardez and López-Luna.[17]

Polyphenolic compounds also have been considered as a potential alternative for the treatment of MS given their effect in the absorption and metabolism of simple carbohydrates,[18,33] mainly reflected in the hypoglycemic and hypolipidemic effect of the extracts evaluated in this work. Recent studies have established that phenolic and flavonoid compounds are both capable of inhibiting lipid accumulation and apoptosis induction,[34] although they regulate the adipocyte physiology in a different way. An hypercaloric diet and genetic predisposition are the immediate causes of developing MS risk factors, such as obesity, insulin resistance, hyperlipidemia and hypertension, as well as of other metabolic diseases such as type II diabetes and cardiovascular disease.[35] In this work the hypercaloric diet (40% sucrose solution) administered at the beginning of the experiments was the principal factor who caused MS in the rats, thereby this work demonstrates that the extracts herein evaluated decreased the adiposity index and thus its relationship with associated metabolic disorders (hyperglycemia, hypercholesterolemia and hypertriglyceridemia).

*Prosthechea karwisnkii* extracts, by reducing adipose tissue, may also decrease characteristics MS serum parameters. This is because adipose tissue regulates the activation of macrophages, which favor the secretion of adipokines that regulates insulin resistance and the accumulation of triglycerides and cholesterol caused by increase of adiponectin and leptin secretions, in order to maintain equilibrium with the excess nutrients consumed by the organism.[34,35] For this reason, is require carry out studies that allow to evaluate the effect of the extracts on adipose tissue and dyslipidemia in a MS rat model, this research are actually in progress. In conclusion, the hydroalcoholic extracts studied hereof represent an alternative for the treatment of MS in the traditional medicine. Their reductive effect was showed even with the continued administration of a hyper-caloric diet to the groups treated with the extracts.

## Conflict of interest statement

The authors declare that this article content has no conflicts of interest.

## Acknowledgements

At the Consejo Nacional de Ciencia y Tecnología (CONACyT-Mexico), at Programa Nacional de Movilidad Estudiantil (ECOES) Santander for the scholarships awarded and Comisión de Operación y Fomento de Actividades Académicas (COFAA-IPN) for the support received. At the University Cristobal Colon for the facilities provided to use the vivarium, particular to M.V.Z. Christian Bautista Piña. Finally to the organizers of the Easter festivities in the Villa de Zaachila, Oaxaca and to the authorities of San Pedro and San Pablo Teposcolula, Oaxaca for the collection of plant material and the facilities provided.

## References

1. Tang L-Q, Wei W, Chen L-M, Sheng L. Effects of berberine on diabetes induced by alloxan and a high-fat/high-cholesterol diet in rats. *J Ethnopharmacol*. 2006;108:109–115.
2. McCune LM, Johns T. Antioxidant activity relates to plant part, life form and growing condition in some diabetes remedies. *J Ethnopharmacol*. 2007;112:461–469.
3. Alexander-Aguilera A, Hernández-Diaz G, Lara-Barcelata M, Angulo-Guerrero O, Oliart-Ros RM. Effects of fish oil on hypertension, plasma lipids, and tumor necrosis factor-α in rats with sucrose-induced metabolic syndrome. *J Nutr Biochem*. 2004;15:350–357.
4. Aguilar-Salinas CA, Rojas R, Gómez-Pérez FJ, et al.. The metabolic syndrome: a concept hard to define. *Arch Med Res*. 2005;36:223–231.
5. Gurrola-Díaz CM, García-López PM, Sánchez-Enríquez S, Troyo-Sanromán R, Andrade-González I, Gómez-Leyva JF. Effects of *Hibiscus sabdariffa* extract powder and preventive treatment (diet) on the lipid profiles of patients with metabolic syndrome (MeSy). *Phytomed*. 2010;17:500–505.
6. Rogulj D, Konjevoda P, Milic M, Mladinic M, Domijan A-M. Fatty liver index as an indicator of metabolic syndrome. *Clin Biochem*. 2012;45:68–71.
7. Kumar EKD, Janardhana GR. Antidiabetic activity of alcoholic stem extract of *Nervilia Plicata* in streptozotocin-nicotinamida induced type 2 diabetic rats. *J Ethnopharmacol*. 2011;133:480–483.
8. Zhao Y, Son Y-O, Kim S-S, Jang Y-S, Lee J-C. Antioxidant and anti-hyperglycemic activity of polysaccharide isolated from *Dendrobium chrysotoxum* Lindl. *J Biochem Mol Biol*. 2007;40:670–677.
9. Luo A, Chung Z, Ge S, et al.. Effect of *Dendrobium denneanum* polysaccharide reducing blood glucose *in vivo*. *Chin J Appl Environ Biol*. 2006;12:334–337.
10. Estrada S, López-Guerrero JJ, Villalobos-Molina R, Mata R. Endothelium-independent relaxation of aorta rings by two stilbenoids from the orchids *Scaphyglottis livida*. *Fitoterapia*. 2006;77:236–239.
11. Vergara-Galicia J, Ortiz-Andrade R, Castillo-España P, et al.. Antihypertensive and vasorelaxant activities of Laelia autumnalis are mainly trough calcium channel blockade. *Vasc Pharmacol*. 2008;49:26–31.
12. Vergara-Galicia J, Ortiz-Andrade R, Rivera-Leyva J, et al.. Vasorelaxant and antihypertensive effects of methanolic extract from roots of *Laelia anceps* are mediated by calcium-channel antagonism. *Fitoterapia*. 2010;81:350–357.

13. Pérez-Gutierrez RM, Hoyo-Vadillo C. Antidiabetic activity of an hexane extract of *Prosthechea michuacana* in streptozotocin-induced diabetic rats. *Bol Latin Car Plant Med Arom*. 2011;10:570–580.
14. Cruz-García G, Solano-Gómez R, Lagunez-Rivera L. Documentation of the medicinal knowledge of *Prosthechea karwinskii* (Orchidaceae) in a Mixtec community in Mexico. *Rev Bras Farmacog*. 2014;24:731–736.
15. Solano-Gómez R, Cruz-Lustre G, Martínez-Feria A, Lagunez-Rivera L. Plantas utilizadas en la celebración de Semana Santa en Zaachila, Oaxaca, México. *Polibotánica*. 2010;29:263–279.
16. García-Peña MR, Peña M. Uso de las orquídeas de México desde la Época Prehispánica hasta nuestros días. *Orquídea*. 1981;8:59–76.
17. Mijangos-Ricardez OF, López-Luna J. Static-dynamic superheatd liquid extraction of phenols from *Prosthechea varicose* and *Prosthechea karwinskii* (orchids) prior to determination by LC-DAD. *J Nat Prod*. 2013;2013:199–203.
18. Heim K, Tagliaferro A, Bobilya D. Flavonoid antioxidants: chemistry, metabolism and structure activity relationships. *J Nutr Biochem*. 2002;13:572–584.
19. Walle T. Absorption and metabolism of flavonoids. *Free Radical Biol Med*. 2004;7:829–837.
20. González-Castejón M, Rodríguez-Casado A. Dietary phytochemicals and their potential effects on obesity: a review. *Pharmacol Res*. 2011;64:438–455.
21. Scherer R, Godoy HT. Antioxidant activity index (AAI) by the 2,2-diphenyl-1-picrylhydrazyl method. *Food Chem*. 2009;112:654–658.
22. SAGARPA. Norma Oficial Mexicana NOM-062-ZOO-1999, Especificaciones técnicas para la producción, cuidado y uso de los animales de laboratorio*Diario Oficial de la Federación. Fecha de publicación 22 de agosto de 2001*.
23. Silva RD'A, Scopel Bueno AL, Weich Gallon C, et al.. The effect of aqueous extract of gross and commercial yerba mate (Ilex paraguariensis) on intra-abdominal and epididymal fat and glucose levels in male Wistar rats. *Fitoterapia*. 2011;82:818–826.
24. Kishino E, Ito T, Fujita K, Kiuchi Y. A mixture of *Salacia reticulata* (Kotala himbutu) aqueous extract and cyclodextrin reduces body weight gain, visceral fat accumulation, and total cholesterol and insulin increases in male Wistar fatty rats. *Nutr Res*. 2009;29:55–63.
25. Raasmaja A, Lecklin A, Ming Li X, et al.. A water-alcohol extract of *Citrus grandis* whole fruits has beneficial metabolic effects in the obese Zucker rats fed with high fat/high cholesterol diet. *Food Chem*. 2013;138:1392–1399.
26. Chen N, Bezzina R, Hinch E, et al.. Green tea, black tea, and epigallocatechin modify body composition improve glucose tolerance, and differentially alter metabolic gene expression in rats fed a high-fat diet. *Nutr Res*. 2009;29:784–793.
27. Montonen J, Knekt P, Jarvinen R, Reunanen A. Dietary antioxidant intake and risk of type 2 diabetes. *Diab Care*. 2004;27:362–366.
28. Ylonen K, Alfthan G, Groop L, Saloranta C, Aro A, Virtanen SM. Dietary intakes and plasma concentrations of carotenoids and tocopherols in relation to glucose metabolism in subjects at high risk of type 2 diabetes: the Botnia Dietary Study. *Am J Clin Nutr*. 2003;77:1434–1441.
29. Moretti M, Cossignani L, Messina F, et al.. Antigenotoxic effect, composition and antioxidant activity of *Dendrobium speciosum*. *Food Chem*. 2013;4:660–665.
30. Bafna AR, Mishra SH. In vitro antioxidant activity of methanol extract of rhizomes of *Curculigo orchioides* Gaertn. *Ars Pharmaceutica*. 2005;46:125–138.
31. Carvalho M, Ferreira PJ, Mendes VS, et al.. Human cancer cell antiproliferative and antioxidant activities of *Juglans regia* L. *Food Chem Toxicol*. 2010;48:441–447.
32. Nain P, Kumar A, Sharma S, Nain J. In vitro evaluation of antimicrobial and antioxidant activities of methanolic extract of *Jasminum humile* leaves. *Asian Pac J Trop Med*. 2011;804–807.
33. Cherniack EP. Review, poliphenols: planting the seeds of treatment for the metabolic syndrome. *Nutrition*. 2011;27:617–623.
34. Eckel RH, Grundy SM, Zimmet PZ. The metabolic syndrome. *Lancet*. 2005;365:1415–1428.
35. Fantuzzi G, Mazzone T. *Adiposse Tissue and Adipokines in Health and Desease*. Human Press; 2007.