Bull. Egypt. Soc. Physiol. Sci. Vol. (41), Issue (4), 500-508

Bull. of Egyp. Soc. Physiol. Sci.

(Official Journal of Egyptian Society for Physiological Sciences)

(pISSN: 1110-0842; eISSN: 2356-9514)

p- Coumaric Acid Prevents Fructose-induced Dyslipedemia and Hypertension

Ahmed Abdelhakam [1], Akef Khowailed [2] ,Marwa Abdeltawab Mohammed[1]

[1]Department of Physiology, Faculty of Medicine, Beni-Suef University, Beni-Suef, Egypt
[2]Department of Physiology, Faculty of Medicine, Cairo University, Beni-Suef, Egypt

Submit Date: Jan 12, 2021
Revise Date: August 23, 2021
Accept Date: August 31, 2021

Keywords

- *p*-Coumaric acid
- Fructose
- Atherogenic index
- Hypertension

Abstract

In recent decades, rates of insulin resistance and hypertension increase dramatically due to many factors. One of them is excessive fructose consumption. *p*-coumaric acid has potent antioxidant and anti hyperglycemic properties. Aim of the work: In this study, we examined the ability of *p*-coumaric acid to prevent high fructose induced hypertension and the possible underlying mechanisms. Material and methods: 24 adult male albino rats were divided randomly into 3 groups; control group, fructose group received fructose 60% dissolved in water for 5 weeks and *p*-coumaric acid + fructose group received *p*-coumaric acid dissolved in carboxy methyl cellulose 100ml/kg/day orally for 2 weeks then fructose 60% dissolved in water for 5 weeks. For each rat, blood pressure was measured and a blood sample was taken by retro orbital method under ether anesthesia for measurement of plasma level of glucose,triglycerides (TG) and high density lipoprotein (HDL), then triglycerides glucose (TYG) index and atherogenic index were calculated. Results: Fructose group showed significant increases of blood pressure, plasma glucose, TG, TYG index and atherogenic index while showed a significant decrease of plasma HDL when compared to control group ($p<0.01$). *p*-Coumaric acid reversed these results. Conclusion: *p*-Coumaric acid protects against hypertension induced by high fructose diet.

Corresponding author: Marwa Abdeltawab Mohammed, Department of Physiology, Faculty of Medicine, Beni-Suef University, Beni-Suef, Egypt. Email: marwaseloma@yahoo.com. Telephone :01100616755

INTRODUCTION

During the last decades, food habits have greatly altered beside sedentary lifestyle leading to high rate of development of obesity, fatty liver, hypertension, and type 2 diabetes [1, 2]. Consuming of excess sugar sweetened beverages (SSBs) are considered the main change in the dietary patterns, these sweeteners include sucrose which contains 50% saccharose and 50% fructose, while high fructose syrup contains about 55% fructose [3]. A great evidence estimates high risk factors for development of cardiovascular diseases and insulin resistance as a result to an extra fructose consumption [4]. Also, fatty liver is reported to be associated with high incidence of cardiometabolic problems [5].For all these systemic disturbances, it is necessary to search for ultimate solutions.

The insulin resistance is considered to be secondary to the suppression or inhibition of insulin signaling [6]. The interface between insulin resistance and hypertension can be thought to be two independent processes or as development of hypertension secondary to insulin resistance. In the independent mechanisms, both insulin resistance and hypertension may develop due to the same disorder, in the form of an increase in intracellular free calcium, with a result of vasoconstriction and impaired insulin action[7].

On the other hand, hyperinsulinemia can be considered as a main factor in the origin of hypertension through different mechanisms, including stimulation of the sympathetic system, increased sodium reabsorption in the renal tubules, and increase vascular resistance in smooth muscle cells [7].

Phenolic acids like ferulic, chlorogenic and coumaric acids have been documented to have great antioxidant characters. The antioxidant properties of phenolic acids have been established to provide healthy endothelium and prevent thrombosis or atherosclerosis [8]. The anti hypertensive effect is related to different mechanisms, like vasodilatation secondary to enhancement of NO bioavailability, antioxidant properties and the inhibition of angiotensin converting enzyme activity [9, 10]. Taking these data in consideration, we hypothesized that p-coumaric acid can protect against the development of hypertension secondary to high fructose consumption.

2.Materials and methods:

-Chemicals:

p-Coumaric acid and sodium carboxymethyl cellulose (Na CMC) were purchased from Sigma-Aldrich (St. Louis, MO, USA). Fructose was obtained from El-shark company (Cairo, Egypt). Glucose, triglycerides (TG), high -density lipoprotein (HDL) colorimetric kits were purchased from Bio-Diagnostic company (Cairo, Egypt).

-Animals and experiment:

Twenty four adult male rats with weights rang between 180 - 220 grams were involved in this study. The rats were randomly divided into 3 groups (8/each group) with normal temperature of 24 _ 28°C and 12-h light and dark cycle.

Rats were allowed for one week for acclimatization before beginning of the experiment with a free access for rodent chow and water ad libitum. The study was performed according to the guidelines of institutional animal care and use committee of Beni-Suef University (IACUC).

Experimental design and treatments:

The rats were randomly divided into three groups as follows:

Group1 (Control)8 rats: normal rats were given an equivalent volume of 0.5% Na CMC

Group 2 (Fructose) 8 rats: Fructose 60% were dissolved in the drinking water for 5 weeks for rats in this group [11].

Group 3 (*p*-Coumaric acid + fructose)8 rats: rats were given P-coumaric acid dissolved in NA CMC (100 mg/kg BW/orally) for two weeks followed by fructose 60% dissolved in the drinking water for 5 weeks [12].

After the determined duration of the experiment, the rats fasted for 12 hours at night and in the morning, blood pressure was measurement then blood samples were taken in a Capillary tube by retro-orbital route under ether anesthesia. Each blood sample was immediately collected in two tubes one contained fluoride for measurement of glucose and the other contained EDTA for TG and HDL and then plasma was separated by centrifuge for 20 minutes after that, samples were Store at – 80 ° C.

-Measurement of systolic blood pressure:

Systolic blood pressure was measured by tail-cuff non-invasive method (Power Lab 16/35, AD Instruments Company, Australia)

Diastolic pressure can't be measured by this instrument and we considered the systolic pressure is sufficient for detection of hypertension.

-Measurement of plasma glucose:

Plasma glucose was measured by the colorimetric kits (Bio-Diagnostic, Cairo, Egypt) according to the manufacturer's instructions[13].

-Measurement of plasms triglycerides (TG) & high density lipoprotein(HDL) :

Plasma TG and HDL were measured using the colorimetric kits (Bio-Diagnostic, Cairo, Egypt) according to manufacture's descriptions[14,15].

-Calculation of triglycerides glucose- index (TYG- index): was calculated according to the following formula: Ln

Fasting triglycerides (mg/dl) – Fasting glucose(mg/dl) /2 [16].

-Calculation of atherogenic index of plasma (AIP) was calculated by this equation:

$\log^{10}$(TG/HDL-C).

-Statistical analysis:

The values of this work were represented as mean± SEM . One-way analysis of variance (ANOVA) was performed then Tukey HSD test was done for multiple comparison between different groups. P value < 0.05 was to be significant.

3.Results:

Table (1) demonstrates significant increases of plasma glucose and TG

levels in fructose group when compared to control group (p <0.01), while mean value of HDL showed a significant decrease in fructose group in comparison to control group (p<0.01). On the other hand, significant decreases were observed in plasma glucose and TG levels in P-coumaric acid + fructose group when compared to fructose group (p <0.01) but a significant increase of HDL level in *p*-coumaric acid + fructose group in comparison to fructose group (p <0.01).

As detected in table (2), significant increases of systolic blood pressure, TYG index and AIP were detected in fructose group when compared to control group (p <0.01). On contrary, significant decreases of systolic blood pressure, TYG index and AIP in *p*-coumaric acid + fructose group in comparison to fructose group (p<0.01).

Table(1): Changes in plasma levels of glucose, triglycerides (TG), and high density lipoprotein cholesterol (HDL) in the three study groups:

	Control group /8 rats	Fructose group/8 rats	Fructose +*p*-coumaric acid group /8 rats
Glucose mg/dl	86 ±7.9	350 ±0.6 [a]	108 ±11 [b]
TG mg/dl	181 ±14.4	229 ±0.97 [a]	198 ±29.2 [b]
HDL mg/l	51 ±0.41	38 ±0.22 [a]	65 ±0.65 [b]

Values are presented as Mean ± SEM
a: significance of difference from control group by Tukey's HSD test at $p< 0.05$.
b: significance of difference from fructose group by Tukey's HSD test at $p< 0.05$.

Table (2): Changes in systolic pressure, TYG index and atherogenic index in the three study groups:

	Control group /8 rats	Fructose group /8 rats	Fructose + *p*-coumaric acid group /8 rats
Systolic pressure mmHg	99 ±7.9	168 ±0.6 [a]	88 ±11.1 [b]
TYG index	4.72±0.037	5.66±0.06 [a]	4.8 ±0.32 [b]
Atherogenic index	0.19 ±0.003	38 ±0.005 [a]	0.13 ±0.03 [b]

Values are presented as Mean ± SEM
a: significance of difference from control group by Tukey's HSD test at $p< 0.05$.
b: significance of difference from fructose group by Tukey's HSD test at $p< 0.05$.

4.Discussion:

No debt that increase the incidence of hypertension, insulin resistance, metabolic syndrome and type 2 diabetes mellitus is greatly linked to alternation of life style and excess use of drinks containing fructose in high concentration.

The present study was designed to clarify the possible protective effect of P-coumaric acid against hyperglycemia,dyslipidemia and hypertension induced by fructose feeding. In this work, rats fed fructose showed significant increases of glucose and TG and a significant decrease of HDL as reported by previous studies [17].

Concerning the mechanism by which hyperglycemia was developed, fructose has been reported to stimulate gluconeogenesis and glycogenolysis through up regulation of enzymes involved and suppress glycogenesis via down regulation of enzymes involved [18].

At the same time, fructose has been approved to induce dyslipidemia through different mechanisms. Fructose enhances lipogenesis as it directly stimulates sterol regulatory element-binding protein 1 (SREBP-1c), a major transcriptional regulator of de novo lipogenesis (DNL) [19]. Moreover,fructose stimulates lipid accumulation in the liver by down regulation of peroxisome proliferator-activated receptor α (PPARα) leading to inhibition of β-oxidation and activation of ER stress and uric acid formation. in addition,fructose increases circulating fibroblast growth factor 21 (FGF21) [20] which in turn activates DNL [20].

On the other hand, rats fed *p*-coumaric acid prior to fructose exhibited a protection and showed significant decreases of glucose and TG levels, while showed a significant increase of HDL level. *p*-Coumaric acid has been established to attenuate T2DM through improvement of fasting blood glucose and HbA1c levels in STZ-induced diabetic rats [12]. Moreover, Yoon et al., confirmed the ability of p-coumaric acid to stimulate glucose

transport via AMP-activated protein kinase (AMPK) activation [21].

p- Coumaric acid can alleviate insulin resistance by different mechanisms including reduction of the intestinal absorption of dietary carbohydrate through inhibition of glucosidase and amylase [22, 23]. Also,p-coumaric acid has been documented to enhance the Phosphorylation of acetyl-CoA carboxylase (ACC) and up regulation of PPARα, leading to promotion of the β-oxidation of fatty acids. In addition, it suppresses oleic acid-induced triglyceride accumulation [24]. Besides these mechanisms,antioxidant and anti- inflammatory properties of *p*-coumaric acid have a vital role in prevention of insulin resistance [25].

In the present study, rats fed fructose exhibited a significant increase of the systolic blood pressure when compared with control group as detected previously [25], while rats fed *p*-coumaric acid before fructose were protected. A strong relation was reported between obesity, insulin resistance and hypertension. Studies established that fructose induces hypertension through various ways, it can enhance sodium and chloride absorption with salt overload and elevation of blood pressure [26].

On the other hand, fructose feeding has been observed to decrease the production of nitric oxide (NO), an effective vasodilator essential for maintaining BP, through inhibition and down regulation of eNOS[27]. Fructose considers the only carbohydrate that can produce uric acid through multiple steps including,phosphorylation by fructokinase enzyme forming fructose 1-phosphate, this reaction needs ATP as a phosphate donor[28] resulting in depletion of intracellular ATP levels [29]. Consequently,intracellular phosphate levels fall with activation of adenosine mono phosphate(AMP) deaminase [28],which converts AMP to inosine monophosphate (IMP), that in turn metabolized to hypoxanthine [30]. Hypoxanthine then is converted to xanthine and then to uric acid under the effect of xanthine oxidase [31].

Hyperuricemia contributes in endothelial dysfunction through induction of oxidative stress and reduction of eNOS and nitric oxide levels [30]. Furthermore, uric acid upregulates C-reactive protein (CRP) in the endothelium which can inhibit nitric oxide release[32].On the other hand, fructose has been observed to activate the sympathetic nervous system, with subsequent development of hypertension[33].

The anti hypertensive effect of *p*-coumaric acid is highly linked to various mechanisms. The antioxidant properties of phenolic acids have been distinguished providing healthy endothelium with subsequent prevention of thrombosis and atherosclerosis [8]. oxygen free radicles has been found to have a role in the endothelial dysfunction and hypertension development through reduction of the NO bioavailability [34]. Moreover, phenolic acids can inhibit angiotensin-converting enzyme, a great incidence confirms the contribution of renin angiotensin system in the development of hypertension[35].

In the present work, fructose group showed significant increases of Tyg index and AIP in comparison to control group, on the other hand, *p*-coumaric acid administration avoid these results. TYG is a marker with a high specificity and sensitivity to distinguish insulin resistance [36], diabetes mellitus and metabolic syndrome [37, 38]. TyG index was found to be associated with increase of cardiovascular risk factors (39).While AIP is

considered as an optimal sign of dyslipidemia [40] and acts as an indicator of plasma atherosclerosis [41].

-Conflict of interest :

No conflict of interest

5. References:

[1] Blüher M. Obesity: global epidemiology and pathogenesis. Nature Reviews Endocrinology. 2019 Feb 27:1.

[2] Miller A, Adeli K. Dietary fructose and the metabolic syndrome. Current opinion in gastroenterology. 2008 Mar 1;24(2):204-9.

[3] Vos, M.B., Abrams, S.H., Barlow, S.E., Caprio, S., Daniels, S.R., Kohli, R., Mouzaki, M., Sathya, P., Schwimmer, J.B., Sundaram, S.S. and Xanthakos, S.A., 2017. NASPGHAN clinical practice guideline for the diagnosis and treatment of nonalcoholic fatty liver disease in children: recommendations from the Expert Committee on NAFLD (ECON) and the North American Society of Pediatric Gastroenterology, Hepatology and Nutrition (NASPGHAN). Journal of pediatric gastroenterology and nutrition,(2) 64 p.319.

[4] Lim S, Taskinen MR, Borén J. Crosstalk between nonalcoholic fatty liver disease and cardiometabolic syndrome. Obesity Reviews. 2019

Apr;20(4):599-611.

[5] Schulze, M., 2018. Metabolic health in normal-weight and obese individuals. *Diabetologia*, 62(4), pp.558-566.

[6] Nakamura M, Yamazaki O, Shirai A, Horita S, Satoh N, Suzuki M, Hamasaki Y, Noiri E, Kume H, Enomoto Y, Homma Y. Preserved Na/HCO3 cotransporter sensitivity to insulin may promote hypertension in metabolic syndrome. Kidney International. 2015 Mar 1;87(3):535-42.

[7] Landsberg L, Aronne LJ, Beilin LJ, Burke V, Igel LI, Lloyd-Jones D, Sowers J. Obesity-related hypertension: pathogenesis, cardiovascular risk,and treatment: a position paper of the Obesity Society and the American Society of Hypertension. The Journal of Clinical Hypertension. 2013 Jan;15(1):14-33.

[8] Barbour JA, Howe PR, Buckley JD, Bryan J, Coates AM. Nut consumption for vascular health and cognitive function. Nutrition research reviews. 2014 Jun;27(1):131-58.

[9] Ochiai R, Sugiura Y, Otsuka K, Katsuragi Y, Hashiguchi T. Coffee bean polyphenols ameliorate postprandial endothelial dysfunction in healthy male adults. International journal of food sciences and nutrition. 2015 Apr.

[10] Godos J, Sinatra D, Blanco I, Mulè S, La Verde M, Marranzano M. Association between dietary phenolic acids and hypertension in a mediterranean cohort. Nutrients. 2017;9(10):1069 .

[11] Abdelrahman AM, Al Suleimani YM, Ashique M, Manoj P, Ali BH. Effect of infliximab and tocilizumab on fructose-induced hyperinsulinemia and hypertension in rats. Biomedicine & Pharmacotherapy. 2018 Sep

[12] Amalan V, Vijayakumar N, Ramakrishnan A. p-Coumaric acid regulates blood glucose and antioxidant levels in streptozotocin induced diabetic rats.J Chem Pharm Res, 2015,7:831-839

[13]Trinder P. Evaluation of Trinder glucose oxidase method. Clin 1975, 21, 1754–1760.

[14] Buckley G C, Cutler J M, Little J A. Serum triglyceride: method of estimation and levels in normal humans. Can Med Asso J,1966,94(17): 886-888.

[15]Bachorik P S, Walker R, Brownell K D, Stunkard A J, Kwiterovich, P. O. Determination of high density lipoprotein-cholesterol in stored human plasma. J of Lipid Res 1980, 21(5), 608–616.

[16] Sánchez-Íñigo, L., Navarro-González, D., Fernández-Montero, A., Pastrana-Delgado, J., & Martínez, J. A. The TyG index may predict the development of cardiovascular events. European journal of clinical investigation, 2016,46(2), 189-197.

[17] Liu IM, Tzeng TF, Liou SS. A Chinese herbal decoction, Dang Gui Buy Xue Tang, prepared from Radix Astragali and Radix Angelicae sinensis,ameliorates insulin resistance induced by a high-fructose diet in rats.Evidence-Based Complementary and Alternative Medicine. 2011.

[18] Geidl-Flueck B, Gerber PA. Insights into the hexose liver metabolism Glucose versus fructose. Nutrients. 2017 Sep;9(9):1026.

[19] Rutledge AC, Adeli K. Fructose and the metabolic syndrome: pathophysiology and molecular mechanisms. Nutrition reviews. 2007 Jun:suppl_1):S13-23.

[20] Stanhope KL, Havel PJ. Fructose consumption: recent results and their potential implications. Annals of the New York Academy of Sciences. 2010 Mar;1190:15.

[**21] Yoon SA, Kang SI, Shin HS, Kang SW, Kim JH, Ko HC, Kim SJ.** p- Coumaric acid modulates glucose and lipid metabolism via AMP-activated protein kinase in L6 skeletal muscle cells. Biochemical and biophysical research Biochemical and biophysical research communications, 2013, 432.4: 553-557.

[22] Adisakwattana S, Sookkongwaree K, Roengsumran S, Petsom A,Ngamrojnavanich N, Chavasiri W, Deesamer S, Yibchok-anun S. Structure activity relationships of trans-cinnamic acid derivatives on α-glucosidase inhibition. Bioorganic & medicinal chemistry letters. 2004 Jun 7;14(11):2893-

[23] Pei, K., Ou, J., Huang, J., & Ou, S. (2016). p-Coumaric acid and its conjugates: dietary sources, pharmacokinetic properties and biological activities. *Journal of the Science of Food and Agriculture*,2016, *96*(9), 2952-2962.

[24] Kim EO, Min KJ, Kwon TK, Um BH, Moreau RA, Choi SW. Anti-inflammatory activity of hydroxycinnamic acid derivatives isolated from corn bran in lipopolysaccharide-stimulated Raw 264.7 macrophages. Food and Chemical Toxicology. 2012 May 1;50(5):1309-16.

[25] Song D, Hutchings S, Pang CC. Chronic N-acetylcysteine prevents fructose-induced insulin resistance and hypertension in rats. European Journal of pharmacology. 2005 Jan 31;508(1-3):205-10.

[26] Cabral PD, Hong NJ, Hye Khan MA, Ortiz PA, Beierwaltes WH, Imig JD,Garvin JL. Fructose stimulates Na/H exchange activity

and sensitizes the proximal tubule to angiotensin II. Hypertension. 2014 Mar;63(3):e68-73.

[27] Okamura T, Tawa M, Geddawy A, Shimosato T, Iwasaki H, Shintaku H,Yoshida Y, Masada M, Shinozaki K, Imamura T. Effects of atorvastatin, amlodipine, and their combination on vascular dysfunction in insulin-resistant Journal of pharmacological sciences, 2014, 124.1: 76-85.

[28] Van Den Berghe G, Bronfman M, Vanneste R, Hers HG. The mechanism of adenosine triphosphate depletion in the liver after a load of fructose. A kinetic study of liver adenylate deaminase. Biochemical Journal. 1977, 162. 3:601-609.

[29] Abdelmalek MF, Lazo M, Horska A, Bonekamp S, Lipkin EW, Balasubramanyam A, Bantle JP, Johnson RJ, Diehl AM, Clark JM, Fatty Liver Subgroup of the Look AHEAD Research Group. Higher dietary fructose is associated with impaired hepatic adenosine triphosphate homeostasis in obesity individuals with type 2 diabetes. Hepatology. 2012 Sep;56(3):952-60.

[30] Jia G, Aroor AR, Whaley-Connell AT, Sowers JR. Fructose and uric acid :is there a role in endothelial function?. Current hypertension reports. 2014, 434:(6) 16;1

[36] Du T, Yuan G, Zhang M, Zhou X, Sun X, Yu X. Clinical usefulness of lipid ratios, visceral adiposity indicators, and the triglycerides and glucose index as risk markers of insulin resistance. Cardiovascular diabetology. 2014 Dec;13(1):146.

[31] Cao H, Pauff JM, Hille R. Substrate orientation and catalytic specificity in the action of xanthine oxidase the sequential hydroxylation of hypoxanthine to uric acid. Journal of Biological Chemistry. 2010 Sep 3;285(36):28044-53.

[32] Kang DH, Park SK, Lee IK, Johnson RJ. Uric acid–induced C-reactive protein expression: implication on cell proliferation and nitric oxide production of human vascular cells. Journal of the American Society of Nephrology. 2005 Dec 1;16(12):3553-62.

[33] 7Tran LT, MacLeod KM, McNeill JH. Selective alpha 1-adrenoceptor blockade prevents fructose-induced hypertension. Molecular and cellular biochemistry. 2014 Jul 1;392(1-2):205-11.

[34] Paravicini TM, Touyz RM. NADPH oxidases, reactive oxygen species, and hypertension: clinical implications and therapeutic possibilities. Diabetes care.2008, 31.Supplement 2: S170-S180.

[35] Ochiai R, Sugiura Y, Otsuka K, Katsuragi Y, Hashiguchi T. Coffee bean polyphenols ameliorate postprandial endothelial dysfunction in healthy male adults. International journal of food sciences and nutrition. 2015 April, (3)66;350-4 .

[37] Angoorani P, Heshmat R, Ejtahed HS, Motlagh ME, Ziaodini H, Taheri M,Aminaee T, Goodarzi A, Qorbani M, Kelishadi R. Validity of triglyceride–glucose index as an indicator for metabolic syndrome in children and adolescents: the

CASPIAN-V study. Eating and Weight Disorders-Studies on Anorexia, Bulimia and Obesity. 2018 Dec 1;23(6):877-83.

[38] Lee ES, Kim JH, Park SE, Park CY, Oh KW, Park SW, Rhee EJ, Lee WY. Predictive value of triglyceride glucose index for the risk of incident diabetes: a Year retrospective longitudinal study. PLoS One. 2016 Sep, 11.9: e0163465.

[39] Simental-Mendía LE, Hernández-Ronquillo G, Gómez-Díaz R, Rodríguez-Morán M, Guerrero-Romero F. The triglycerides and glucose index is associated with cardiovascular risk factors in normal-weight children and adolescents. Pediatric research. 2017 Dec;82(6):920.

[40] Cai G, Shi G, Xue S, Lu W. The atherogenic index of plasma is a strong and independent predictor for coronary artery disease in the Chinese Han population. Medicina . 2017 Sep;96(37).

[41] Dobiásová M, Frohlich J. The plasma parameter log (TG/HDL-C) as an atherogenic index: correlation with lipoprotein particle size and esterification rate inapob-lipoprotein-depleted plasma (FERHDL). Clinical biochemistry. 2001, 1;34(7):583-8.