

The Therapeutic Effect of Passion Fruit (*Passiflora edulis*) on Blood Acidity in Rats

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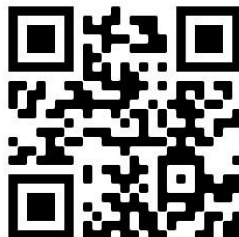
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التأثير العلاجي لفاكهة الباشن على حموضة الدم في الفئران

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ملخص البحث باللغة العربية:

تهدف هذه الدراسة الى تقييم التأثير العلاجي لعصير فاكهة الباشن كمضاد حموضة طبيعي في الفئران. حيث تم تقدير بعض الخواص الطبيعية الكيميائية مثل رقم الـ pH والحموضة الكلية والمواد الصلبة الذائبة الكلية وفيتامين سي والفينولات والفلانويدات الكلية ونشاط مضادات الأكسدة. وقد أجريت الدراسة البيولوجية الحالية على ثلاثون من ذكور الفئران قسمت عشوائيا الى خمس مجموعات، المجموعة الأولى كمجموعة ضابطة سالبة تغذت على الوجبة القياسية فقط، بينما المجموعات الخمس الأخرى فتم تغذيتها على الوجبة القياسية المضاف لها 15% من مسحوق الخبز الابيض لمدة ٤ أسابيع لإصابتها بالحموضة ، وتركت مجموعة واحدة كمجموعة ضابطة موجبة غير معالجة ، بينما المجموعات الأخرى عولجت بمستويات مختلفة من عصير الباشن ٢ % ، ٤ % ، ٦ % لمدة ٨ أسابيع. وقد اظهرت النتائج الى ارتفاع حموضة فاكهة الباشن (انخفاض رقم الـ pH ٣,٦٥) وارتفاع محتوى فاكهة الباشن من فيتامين سي ٣٣,٧٠ ملجم / 100جم كذلك محتوى مرتفع من مضادات الاكسدة. و أشارت النتائج أن كل من المجموعات المعالجة أظهرت ارتفاع معنوي في زيادة وزن الجسم و نسبة الاستفاداة من الطعام بالإضافة إلى ارتفاع معنوي ملحوظ في pH الدم و البيكربونات و الكالسيوم و الفوسفور، المحتوى الكلي لمضادات الاكسدة مقارنة مع المجموعة الضابطة الموجبة بينما سجلت النتائج انخفاض معنوي ملحوظ في pH المعدة، TNF- α ، المحتوى الكلية للأكسدة مقارنة بالمجموعة الضابطة الموجبة. ومن خلال الدراسة تؤكد النتائج أن عصير الباشن له تأثير جيد على حموضة الدم في الفئران.

الكلمات الافتتاحية: فاكهة الباشن - الفئران - حموضة المعدة - مضادان الأكسدة.

The Therapeutic Effect of Passion Fruit (*Passiflora edulis*) on Blood Acidity in Rats

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ABSTRACT:

The purpose of this study was to assess the therapeutic effect on rats of passion fruit juice (*Passiflora edulis*) as a natural anti-acidity agent. Some physicochemical such as pH, titratable acidity, total soluble solid, vitamin C, total phenols total flavonoids, antioxidant activity were determined. Thirty male Sprague-Dawley rats were split into five groups at random (6 rats each). The first group was the negative control fed on basal diet, the other groups were fed to induce acidity for four weeks on high white bread powder (15 % of diet), then divided into four groups, positive control group and three curative groups orally given passion fruit juice 2 %, 4 % and 6 % juice/Kg/bwt/rats during the experimental period, respectively (8 weeks). The findings showed that the passion juice is a rich source acidity being 3.65, vitamin C 33.70 mg/100g and high amounts of natural antioxidants. The therapeutic groups also showed a substantial increase in body weight gain, feed efficiency ratio, blood pH, bicarbonate, calcium, phosphorus and total antioxidant capacity, while showing a significant decrease in feed consumption, stomach pH, TNF-a and total oxidant capacity compared to the untreated community (+ve). This research concluded that it has a positive effect on acidity in rats using passion fruit juice.

Key words: Passion fruit, acidity, stomach pH, antioxidant, bicarbonate.

INTRODUCTION:

Blood acidity is an imbalance in the chemical composition of the blood that affects the body's health, making it a fertile area for diseases. Number 7, which is the neutral ratio between acid and alkali symbolizes the standard pH (**Yee and Rabinstein, 2010**), Acidic blood, followed by many signs, including joint pain, idle and chronic tiredness, low blood pressure, fatigue and stress, migraine, hair loss, skin problems, gum bleeding, nervousness and tension, digestive problems and weakened (**Pizzorno, 2014**). A variety of causes of acidity in the blood, including an excessive intake of acidic foods and an excessive intake of poultry, meat, fish, yellow cheese, whole milk, butter, cream, and white sugar, while vegetables and fruits, various forms and almonds are found in foods that make blood alkali (**Liamis, et al., 2019**).

The passion fruit, which is widely cultivated in subtropical and tropical regions of Asia, America, Australia, and Africa, is one of the common species in the Passifloraceae family. It is used in folk medicine for the treatment of anxiety and insomnia (**Rotta et al., 2019**). The Passion fruit is commonly referred to as the purple passion fruit and is rich in aromatic compounds in its juice. The fruit is ellipse-round, with a diameter ranging from 8 to 10 cm, and at maturity the purple epicarp where the anthocyanin pigments are located. Most of these fruits are used as co-products for the production of juices and produce lots of outside peels. Passion fruit epicarp accounts for 20% of fruit (in mass) and is rich in anthocyanins that concentrate primarily on antioxidants, anti-inflammatory, anti-cardiovascular diseases, etc. (**Zeraik et al., 2011**). A variety of pharmaceutical products based on ingredients have also been developed and used in folk medicine, apart from being a food item. Polyphenols, triterpenes and glycosides, carotenoids, cyanogenic glycosides, polysaccharides, amino acids, essential oils, microelements, and so on are the key components of passion fruit (**Hu et al., 2018**). Among these compounds, derivatives of luteolin, apigenin, and quercetin are the most published. Passion fruit contains, most notably, nutritionally valuable compounds such as vitamin C, dietary fiber, B vitamins, niacin, iron, phosphorus, etc. Various promising bioactivities of passion fruit have been revealed in a broad

range of in vitro and in vivo pharmacological studies, including antioxidant, antimicrobial, anti-inflammatory, anti-hypertensive, hepatoprotective and lung defensive activities, anti-diabetic, sedative, antidepressant function, and anxiolytic-like behavior (**Panelli et al., 2018**). Like hemolysin, passion fruit has been shown to have hemolytic activity. When male Wistar rats were treated with passion fruit, biochemical parameters and antioxidant defenses (superoxide dismutase [SOD] and glutathione peroxidase [GSH-Px] activities and total glutathione [GSH-t] and thiobarbituric acid-reactive substances [TBARS] concentrations) were affected (**Chau and Huang, 2005**).

Gastrointestinal disorders, including constipation, diarrhea and indigestion, abdominal and intestinal gas, can be treated by Passion fruit. It contains a high soluble dietary fiber content, which lowers LDL cholesterol and prevents cancer of the colon and stomach (**Doyama et al., 2005 and Ramos et al., 2007**). Strengthen the body's immune system by containing antioxidants that combat free radicals and are a leading cause of cancer and viral and bacterial infections, and by restricting the development of malignant tumors in healthy (**Akali and Maiti, 2006**).

Maintain a safe heart, because of its flavonoids and its phenolic substances. The use of vegetables and fruits to maintain gastric acidity has been verified by previous studies. The present study was therefore intended to investigate the possible effects on rats of passion fruit juice as a natural anti-acidity agent.

MATERIALS AND METHODS:

Materials:

Source of passion fruit: Passion fruit (*Passiflora edulis*) were obtained from local market in Cairo City, Cairo Governorate, Egypt.

White bread: The white bread was obtained from a local market and roasted at 100°C for 10 min. and grinded to obtain powder form.

Rats: Thirty male rats weighing between (140-150±10g) were randomly distributed into five groups. All the experimental rats were not suffering from any pain or diseases of any kind during the experiment. Rats were obtained from the Agricultural Reached Center, Giza, Egypt. Rats were fed on basal diet and used distilled water as a drink.

Methods:

Preparation of passion juice: The passion fruit pulp was extracted using a blender and the process of filtration. The filtrate obtained was collected in a beaker and about 650ml of juice was obtained in the end according to the method described by (Akpan and Kovo, 2005). The rat was treated orally by stomach tube with various doses of juice such as 2 %, 4 % and 6 % /kg rat body weight.

Physicochemical analysis:

PH measurement: The pH value was measured using a pH meter of a glass electrode. The pH meter could stabilize for one minute and then the pH of the samples was directly reported according to the official method of analysis (AOAC, 1984).

Total Acidity: Titratable acidity was determined according to the official method (AOAC, 1984).

Total Soluble Solids (TSS): The total soluble solids (TSS) were determined at room temperature using hand refractometer with degree °Brix scale 0- 50 according to (AOAC, 1984) standard methods.

Ascorbic acid (Vitamin C): Vitamin C was determined by using 2, 6-dichloro-phenol-indophenol dye according to the official method (AOAC, 1984).

Determination of total phenolic (TPC), total flavonoid and antioxidant activity (DPPH) contents of passion fruit:

The passion fruit content of total phenolic (TPC), total flavonoid (TFC) and antioxidant activity (DPPH) was calculated according to the methods defined by Antolak *et al.*, (2017); Lin & Tang, (2007) and Mensor *et al.*, (2001), respectively.

HPLC analysis of polyphenols: HPLC analysis was performed using a HPLC system consisting of a dual λ detector and a Waters 1525

binary pump and equipped with a Waters Symmetry[®] C18 column (5 mm, 4.6 × 50 mm) with Waters Sentry universalguard column (5 mm, 4.6 × 20 mm) (Waters Corporation, Milford, MA, USA). Phenolic compounds of passion juice were studied using the reference HPLC method by comparing experimental retention times with reported reference values (**Sakakibara et al., 2003**).

Experimental design: Composition of standard basal diet casein (82 % protein), corn oil, fiber (cellulose), mineral mixture, vitamin mixture, methionine, choline chloride and corn starch according to (**Reeves et al., 1993**). Rats were divided into five groups each group has six rats as follow: group one was fed on basal diet only and kept as control negative group, while groups (2, 3, 4 & 5) of rats (n=24) were fed on high white bread powder (15% of diet) for four weeks to induced acidity according to (**Robertson et al., 2007**). These rats were then divided into four groups, positive control group and three curative groups given passion juice orally 2 ml, 4 ml and 6 ml juice/Kg/bwt/ rats, respectively. The experiment lasted for 8 weeks, at the end of experiment, the rats were slaughtered and drawing samples from hepatic partial vein in clean centrifuge tubes. Blood serum was separated by centrifugation at 4000r.p.m. for 15 minutes according to (**Schermer ,1967**).

Biochemical analysis: The pH value of the blood and stomach were determined by direct immersion of pH electrode in Blood and gastric fluid at the room temperature (25 °C) using the digital pH metermodel 3020 Dunmou (Jenway, Essex, UK). Calcium content determination using the atomic absorption method (Pye Unicom model 3300), the calcium of sample was measured at 422 nm according to (**Pearson ,1970**). Phosphorus content was determined according to the colorimetric methods as described by (**Page ,1982**). Bicarbonate (HCO₃⁻) was determined according to (**Tietz ,1999**). Serum total antioxidant, antioxidant capacities and TNF-α were measured using standard spectrophotometric methods according to (**Cao et al., 1993; Flohe & Gunzler, 1984 and Thorell, 1973**), respectively.

Statistical analysis: The data obtained were subjected to Analysis of Variance (ANOVA) appropriate for a completely randomized design, using the statistical program SPSS.20[®] (IBM Cooperation, USA) to assess the significant differences with Duncan's multiple range test.

RESULTS AND DISCUSSION:

Physicochemical properties of passion juice: Data in Table (1) showed the physicochemical properties of passion fruit juice. It is clear to mention that the values of pH, titratable acidity (T.A %), total soluble solids (T.S.S.%) ascorbic acid, total phenols, total flavonoids and antioxidant activity of passion fruit juice were 3.65, 1.80 %, 17.25 %, 33.70 mg/100g, 347.20 mg GAE/100 g, 211.60 mg GAE/100 g and 18.24 %, respectively. These findings are in accordance with **Contreras- Calderon *et al.*, 2011**), who reported that the TPC in the lyophilized passion fruit powder ranged from 45.4 to 833 mg GAE/100 g. While the TFC ranged from RUE/100 g 84 to 1163 mg. They suggested that passion fruits, including antioxidant phenolic compounds, essential amino acids, unsaturated fatty acids, and minerals, could be considered a new natural source of health-promoting compounds.

Data tabulated in Table (2) show the identification of phenolic compounds of passion fruit. The obtained results indicated that the highest phenolic compounds of passion fruit recorded for chlorogenic acid, hydroxybenzoic acid and cyanidin-3- O- glucoside. The values were 672.15, 321.17 and 300.25 mg/100g, respectively. On the other hand, the lowest phenolic compounds of passion fruit recorded for caffeic acid, rosmarinic acid and catechin. The values were 1.41, 4.71 and 9.75 mg/100g, respectively. While quercitrin and vanillic acid did not detect at these conditions. These results are in harmony with **(Giambanelli *et al.*, 2020)** they stated that the total phenolic acids (hydroxybenzoic plus hydroxycinnamic acid derivatives) were the highest concentrated phenolic category in both free and bound phenolic fractions. In short, the high phenolic content and the antioxidant and antihyperglycemic capacity contained in this fruit have verified its functional fruit potential. These findings have also been confirmed by **(Hanaa and Gehan 2016 and Asare *et al.*, 2018)**, who discovered that the passion fruit has a wide variety of plant phytochemical components such as phenols, flavonoids, and tannins.

The nutritional status indicators Table (3) revealed that the differences in feed intake, body weight gain and FER between rats belonging to different study groups were insignificant except for

therapeutic groups 3, 4 and 5 which showed significant reduction in feed intake in addition to significant increase in body weight gain as well as the final when compared to the untreated group (+ve). From such that it could be noticed that the untreated group (+ve) feed showed significant decrease in body weight gain (BWG) and FRE when compared to group (-ve). On the other hand, weight gain was reported as a rare side effect for passion juice consumption. These findings are in the same line as (Maricelma *et al.*, 2012), who claimed that passion juice is a rich source of vitamins, minerals and antioxidants that help protect the cells of the body from harm caused by free radicals known as unstable molecules.

The mean values of stomach pH, blood pH and bicarbonate, of the experimental groups scheduled in Table (4). Therapeutic groups 3, 4 and 5 which showed significant decrease in stomach pH in addition to significant increase in blood pH and bicarbonate when compared to the untreated group (+ve). From such that it could be noticed that the untreated group (+ve) showed significant decrease in Blood pH and bicarbonate when compared to group (-ve). Passion fruit was used to promote digestion, improve appetite and support respiratory health. Passion fruit juice has efficient pathways to achieve acid and alkali balance and enhance the body to ensure proper functioning of this balance to minimize the acid content (acidity, g citric acid/100 ml 3.2 and pH 4.06) (Pragati *et al.*, 2015) and the intake of alkaline food helps to balance the pH of the body (Duc Chinh, *et al.*, 2019).

Table (5) show the values of calcium and phosphorous of rats. Therapeutic groups 3, 4 and 5 showed significant increase in calcium and phosphorous compared to the untreated group (+ve). From such that it could be noticed that the untreated group (+ve) showed significant decrease in calcium and phosphorous compared to group (-ve). Another study found that passion juice may have a positive effect on bone minerals, including calcium and phosphorus (12 mg-1.2 mg) reported (Duc Chinh, *et al.*, 2019).

Data tabulated in Table (6) show the values of TNF- α , total antioxidant capacity and total oxidant capacity in different study groups. Therapeutic groups 3, 4 and 5 which showed significant decrease in TNF- α and total oxidant capacity while showed significant increase in total antioxidant capacity compared to the untreated group

(+ve). Free radicals have variety of adverse effects on cells, resulting in many disorders. In plants, phenolic components may serve as free radical scavengers, resulting in the delay or prevention of free radical oxidative stress. Recently, due to their protective function and improving well-being and wellbeing, plant materials that have proven to be rich in phenolic components are commonly used as food therapies. Oxidative pathways, high cytotoxicity and inhibitory behavior may result from the development and increase of TNF-alpha levels (**Chaturvedi et al., 2013**). Natural antioxidants play a significant role in preventing the response of free radicals and the oxidative sequence to the tissues. Passion fruit's positive effects may be attributed to the compounds in its pulp (**Duc Chinh, et al., 2019**). Species of passion are rich in pectin, minerals, vitamin C, carotenoids and flavonoids (**Sandra et al., 2011**).

Table (1): Physicochemical properties of passion juice.

Passion juice	Value
pH	3.65
Titrateable acidity (T.A %)	1.80
Total soluble solids (T.S.S. %)	17.25
Vitamin C as (ascorbic acid) (mg/100g)	33.70
Total phenols (mgGAE /100g)	347.20
Total flavonoids(mgCE /100g)	211.60
Antioxidant activity (DPPH%)	18.24

GAE= Expressed as gallic acid equivalents,

CE=Expressed as catechin equivalents,

DPPH = 1,1-diphenyl-2-picrylhydrazyl.

Table (2): Identification of phenolic compounds of passion fruit.

Phenolic compounds	Concentrations mg/100g
Gallic acid	100.10
Chlorogenic acid	672.15
Cyanidin-3- O- glucoside	300.25
Ferulic acid	100.83
Iso-querctin	120.42
Quercitrin	ND
Hydroxybenzoic acid	321.17
Syringic acid	59.80
Caffeic acid	1.41
Catechin	9.75
Rosmarinic acid	4.71
Vanillic acid	ND

ND= Not detectable

Table (3): Effect of passion juice on body weight gain, feed intake and feed efficiency ratio in rats.

Variables	Groups Group -ve	Group +ve	Therapeutic group with passion juice		
			2%	4 %	6 %
Feed intake (g/d)	17.33 ^a ±1.21	16.55 ^a ±1.18	16.10 ^b ±1.61	15.65 ^b ±1.27	14.95 ^b ±1.41
Body weight (g)	128.87 ^a ±8.62	87.62 ^c ±5.11	105.44 ^b ±6.61	118.41 ^b ±7.31	121.44 ^b ±8.14
FER	0.089 ^a ±0.008	0.099 ^b ±0.006	0.085 ^b ±0.004	0.083 ^c ±0.002	0.061 ^d ±0.003

Means with the different letters in the same raw meaning significant difference at (P≤0.05).

Table (4): Effect of passion juice on stomach pH, blood pH and bicarbonate in rats.

Variables	Groups Group -ve	Group +ve	Therapeutic group with passion juice		
			2%	4 %	6 %
Stomach pH	4.20 ^c ±0.24	7.80 ^a ±0.62	4.67 ^b ±0.61	5.84 ^b ±0.27	5.01 ^b ±0.41
Blood pH	7.45 ^a ±0.42	5.62 ^c ±0.11	6.44 ^b ±0.61	7.31 ^a ±0.25	7.59 ^a ±0.14
Bicarbonate	26.20 ^a ±3.08	23.20 ^b ±1.06	26.50 ^a ±4.04	27.48 ^a ±4.02	26.33 ^a ±3.03

Means with the different letters in the same raw meaning significant difference at (P≤0.05).

Table (5): Effect of passion juice on calcium and phosphorous in rats.

Variables	Groups	Group -ve	Group +ve	Therapeutic group with passion juice		
				2%	4 %	6 %
Calcium (g/dl)		11.33 ^a ±1.24	8.80 ^c ±0.14	9.67 ^b ±0.91	10.14 ^{ab} ±1.27	11.01 ^a ±1.41
Phosphorous (g/dl)		8.49 ^a ±0.72	6.62 ^c ±0.21	7.02 ^b ±0.41	7.81 ^{ab} ±7.31	8.11 ^a ±8.14

Means with the different letters in the same raw meaning significant difference at (P≤0.05).

Table (6): Effect of passion juice on TNF- α , total antioxidant capacity and total oxidant capacity in rats.

Variables	Groups	Group -ve	Group +ve	Therapeutic group with passion juice		
				2%	4 %	6 %
TNF- α (pg/ml)		3.18 ^d ±0.11	8.73 ^a ±0.17	6.71 ^b ±0.126	4.36 ^{bc} ±0.05	3.91 ^c ±0.05
Total antioxidant capacity (mmol/L)		4.76 ^a ±0.04	1.76 ^d ±0.04	2.94 ^c ± 0.013	3.16 ^b ±0.013	4.04 ^a ±0.013
Total oxidant capacity (mmol/L)		0.235 ^d ±0.01	3.26 ^a ±0.012	1.96 ^b ±0.014	1.57 ^b ±0.01	0.89 ^c ±0.01

Means with the different letters in the same raw meaning significant difference at (P≤0.05).

CONCLUSION:

The potential benefit of passion juice as a good source of natural antioxidants was shown by this study, had an unusual nutritional structure that could draw more attention from nutritionists, health professionals who have a therapeutic action in rats against acid blood.

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