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EFFECT OF CHIA SEEDS ON HYPERGLYCEMIA IN THE BLOOD OF ALBINO RATS

Habib, M. A.; Ashoush, Y. A.; El-Sayed, S. M. and Hegazy, M. M.
Biochemistry Department, Faculty of Agriculture, Menoufia University, Egypt.

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ABSTRACT: The present study aims to delve into the comprehensive chemical composition of chia seeds, with a specific emphasis on elucidating the detailed profile of amino acids. Furthermore, the study sought to evaluate the potential therapeutic efficacy of chia seed protein in modulating blood glucose levels in male diabetic rats. The chemical composition of seeds was as follows: moisture (5.55 %), crude fiber (18.99 %), ash (1.92%), crude protein (18.16 %), total lipids (33.59 %), and total carbohydrate (21.79%). The amino acids composition was as mentioned next: glutamic acid 20.6% followed by arginine 12.47%, meanwhile the aspartic and leucine recorded 9.57 and 7.76% respectively. After 30 days of treatment with chia seeds protein, hyperglycemic rats experienced a significant decrease in glucose levels from 244.17 mg/dl to 119.24 mg/dl. Additionally, triglycerides decreased from 119.74 mg/dl to 81.34 mg/dl, and the total cholesterol level decreased from 126.41 mg/dl to 98.13 mg/dl. Furthermore, the hyperglycemic group's LDL-C and HDL-C were compared. Consequently, the protein found in chia seeds may be advantageous for individuals with diabetes.

Keywords: Chia seeds, hyperglycemia, diabetes mellitus, amino acids, glucose level, liver function.

INTRODUCTION

Diabetes mellitus is a widespread metabolic disorder that continues to impact more people globally. The current count of individuals with diabetes stands at 336 million, but it's anticipated to surge to 552 million by 2030. (Esubalew *et al.*, 2024).

Diabetes damages the cardiovascular system by causing narrowing and reduced flexibility of blood vessels, impeding blood flow and oxygen delivery, which can lead to high blood pressure and damage to blood vessels. High blood sugar levels in diabetes increase the risk of macrovascular diseases such as cardiac arrest, stroke, and peripheral artery disease, impacting overall cardiovascular health. The relationship between diabetes and cardiovascular issues is significant, as all components of the cardiovascular system are exposed to harm from high blood sugar levels. Complications of diabetes can result in microvascular issues affecting the cardiovascular system, leading to problems with the eyes, kidneys, and nervous system. (Bulut Gokten, *et al.*, 2024).

Diabetes can lead to serious health issues, and most of these problems are caused by elevated blood sugar. This includes heart disease and other problems with blood vessels. (Soman *et al.*, 2013).

Dietary proteins and amino acids exert a significant role in glucose metabolism and insulin sensitivity. While elevated protein intake demonstrates beneficial effects on energy homeostasis through satiety induction and potential augmentation of energy expenditure, it concomitantly exhibits detrimental consequences on glucose homeostasis by promoting insulin resistance and increasing gluconeogenesis. Investigations into the modulation of insulin resistance induced by Western diets through varying protein quality rather than quantity have revealed that proteins derived from fish may exhibit the most favorable effects on insulin sensitivity. (Tremblay, *et al.*, 2007).

Many studies have indicated the vital role that plants and their effective components play in improving public health in general, especially improving blood lipid profile indicators (Abozid *et al.*, 2018; Farid *et al.*, 2012; Sakr *et al.*, 2019).

*Corresponding author: salah.mansour32@agr.menofia.edu.eg

The chia plant, (*Salvia hispanica L.*), is an herbaceous annual species within the *Lamiaceae* family, characterized by its oval-shaped seeds measuring 1 to 2 mm in size, which can range in color from black, grey, black spotted to white (Ferdous, *et al*., 2020). Chia seeds, a dietary staple since 1500 BCE, offer a plethora of health advantages. Their nutritional profile, rich in omega-3 fatty acids, antioxidants, dietary fiber, unsaturated fatty acids, gluten-free protein, vitamins, minerals, and phenolic compounds, makes them a valuable dietary addition for promoting overall well-being. (Rahman, *et al*., 2017).

The abundant dietary fibers in chia seeds contribute to effective glycemic management, making them a potential aid in controlling diabetes mellitus. Beyond their numerous health advantages, chia seeds offer significant potential for application in the food industry. This includes their utilization in developing various baked goods, producing biodegradable edible films, and their potential as emulsifiers and stabilizers, among other applications. (Ikumi, *et al*., 2019).

Thus, this study aimed to examine the potential therapeutic role of chia seed protein in mitigating the complications of diabetes, specifically focusing on blood sugar regulation and cardiovascular protection.

MATERIALS AND METHODS

Materials

The chia seeds used in this study were obtained from the National Research Center in Giza, Egypt, specifically from the department specializing in medical and aromatic plants. The seeds were then dehydrated and ground to a fine powder.

Methods

Chemical Composition

The proximate analysis of chia seeds was conducted using standard AOAC (1990) methods. Moisture content was determined by drying samples in an oven, while ash content was measured by incineration. Protein content was assessed using the Kjeldahl method. Crude fiber and fat content were determined through digestion and Soxhlet extraction, respectively. All analyses were performed in triplicate. The carbohydrate content was calculated by subtraction, using the other nutrient values. (Pearson, 1976).

%CHO = 100 – (% moisture + % fat. + % ash + % fiber + % protein)

Identification of amino acids

Amino acids were determined in the seeds using a Microtechna AAA 881 amino acid analyzer according to the method described by (Moor and Sein, (1963). Hydrolysis of the samples was performed in the presence of 6 M HCl at 110 ◦C for 24h under nitrogen gas. Following alkaline hydrolysis, tryptophan content was determined colorimetrically using Miller's (1967) method.

Biological Evaluation

Animals

Adult male Sprague-Dawley albino rats (150-170 grams) were procured from the VACSERA animal facility in Egypt. The rats were housed in wire-bottomed cages and provided with a specialized feeding system to minimize food spillage. Water was supplied via a glass tube attached to an inverted bottle.

Experimental design:

The experimental animals were divided into 3 groups, each of them having 5 rats as follows:

Group 1: Rats allowed to feed without any treatment as control fed on a standard diet, named negative control.

Group 2: Rats are allowed to feed a hyperglycemic diet to induce hyperglycemia by using alloxan through the feeding period, called positive control or hyperglycemic control.

Group 3: Rats were allowed to feed a hyperglycemic diet plus powder of chia seeds protein by 5% of the meal.

Blood sampling and analysis:

Blood samples were obtained from the eye plexuses of animals under anesthesia. These samples were collected in heparinized tubes, centrifuged to separate the plasma, and then stored frozen for later analysis. Blood levels were measured using a standard enzymatic method of Tinder, (1969). The total cholesterol concentration was measured spectrophotometrically using the Allain et al. (1974) method, and triglycerides were analyzed using the Fossati and Prencipe (1982) method. HDL cholesterol was determined using the Lopez et al. (1977) method, while LDL cholesterol was calculated using the Kikuchi (1998) method.

Statistical analysis

Data from animal studies were presented as mean values with standard error and analyzed using one-way ANOVA followed by Duncan's multiple comparison test. Statistical significance was determined at a p-value of less than 0.05. Landue and Everitt (2003).

RESULTS AND DISCUSSION

Chemical composition of chia seeds

Proximate compositions of Chia seeds (*Salvia hispanica*) as shown in Table 1. *Salvia hispanica* seeds where they were as follows: moisture (5.55 %), crude fiber (18.99 %), ash (1.92%), crude protein (18.16 %), crude fat (33.59 %) and total carbohydrate (21.79 %). These results are consistent with the findings of Biswas *et al.* (2023), who determined that the crude protein percentage in Salvia hispanica seeds was 18–24%. Additionally, Weber et al. (1991) reported that the nutritional compositions of chia seeds from different species varied, with protein levels ranging from 19.0% to 26.5%, oil from 15.9% to 34.1%, and fiber content.

Table 1: Major chemical composition of chia seeds (w/w, %).

Chemical composition of plant	Mois ture	Crude protein	Total lipids	Crude fiber	Total ash	Total carbohydrates
Chia seeds	5.55	18.16	33.59	18.99	1.92	21.79

Amino acid composition

Data demonstrated in Table 2, Shows that the amino acids of Salvia hispanica contain 32.72% essential amino acids and 67.28 % nonessential amino acids. The essential amino acid was leucine, which accounted for (7.76 %) followed by phenylalanine (5.13%). Meanwhile, the most abundant nonessential amino acids were glutamic acid. (20.6%) followed by arginine and aspartic acid which accounted (12.46 and 9.57%), respectively.

Our findings corroborate the results reported by (Ikumi, *et al*., 2019). Who reported that chia seeds are a rich source of high-quality protein, containing all nine essential amino acids and a variety of essential minerals. Also, (Bushway, *et al*., 1981), reported that Analyses of chia seed amino acid composition revealed the presence of all ten essential amino acids, with arginine, leucine, phenylalanine, valine, and lysine being particularly abundant. Additionally, chia seed proteins were found to be rich in non-essential amino acids, including glutamic acid, aspartic acid, alanine, serine, and glycine.

In vivo study of the effect of Chia seeds on albino rats.

Glucose level and lipids profile

The impact of chia seed supplementation on blood glucose and total cholesterol levels in hyperglycemic rats was assessed over 30 days. The results of this study are presented in Table 3,4 and Figure 1,2 which depict the average values throughout the experimental duration. The glucose level in the negative control group was 123.34 mg/dl at zero time, while in the hyperglycemic group and chia group glucose levels were 127.27 and 123.72 mg/dl respectively. After 30 days, the glucose level in the negative control group was 125.82 mg/dl, while in the hyperglycemic group, it was 244.17 mg/dl and decreased significantly to 119.24 mg/dl in chia group.

The elevation of glucose level in the hyperglycemic group may be due to a decrease in both insulin secretion and insulin action (Altan 2003), because of injection by alloxan, [do1]is because diabetes causes lipid peroxidation that mediates damage in the pancreas (Ahmed *et al.*, 2014).

Table 2: The amino acid composition of Chia seeds.

Amino acids	Relative concentration (%)
Aspartic acid	9.57
Glutamic acid	20.6
Serine	2.82
Histidine	2.73
Glycine	4.43
Threonine	3.44
Arginine	12.47
Alanine	4.46
Tyrosine	3.72
Cystine	1.51
Valine	3.32
Methionine	1.75
Phenylalanine	5.13
Isoleucine	3.96
Leucine	7.76
Lysine	4.63
Hydroxy proline	2.75
Proline	4.95

Table 3: Effect of chia seeds on glucose level of albino rats.

Group	Glucose (mg/dl)		
	0 day	15 day	30 day
Negative control group	123.34±0.65[a]	124.58±0.81[a]	125.82±0.79 [a]
Positive control group	127.27±0.94[a]	217.72±1.05[b]	244.17±1.08[b]
Chia seeds group	123.72±0.78 [a]	125.21±0.85[a]	119.24±0.95 [a]

Table 4: Effect of chia seeds on total cholesterol of albino rats:

Group	Total cholesterol (mg/dl)		
	0 day	15 day	30 day
Negative control group	97.73±1.12 [a]	98.21±0.98 [a]	99.16±1.21 [a]
Positive control group	97.91±1.04 [a]	121.37±1.07[b]	126.41±1.14[b]
Chia seeds group	97.57±1.21[a]	99.46±1.16 [a]	98.13±1.09 [a]

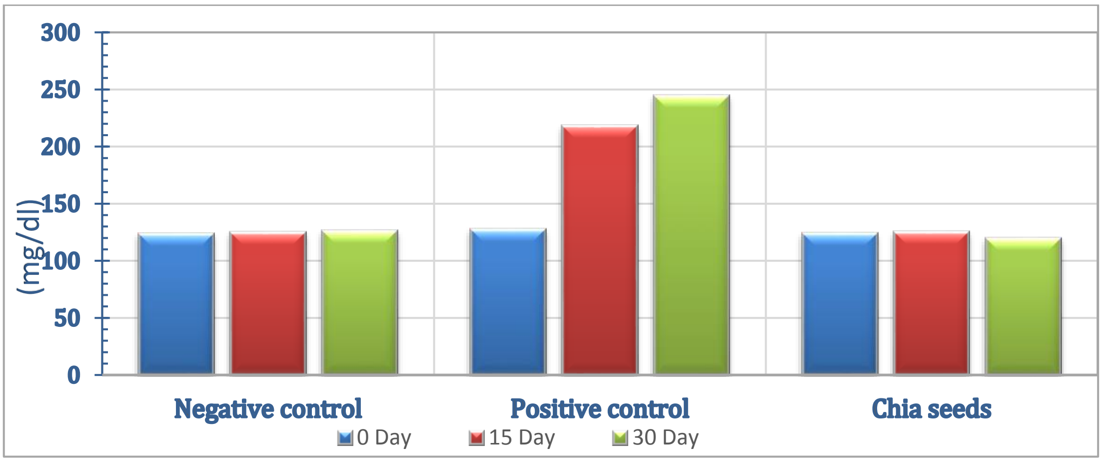

Figure 1: Effect of chia seeds on glucose levels of albino rats (mg/dl).

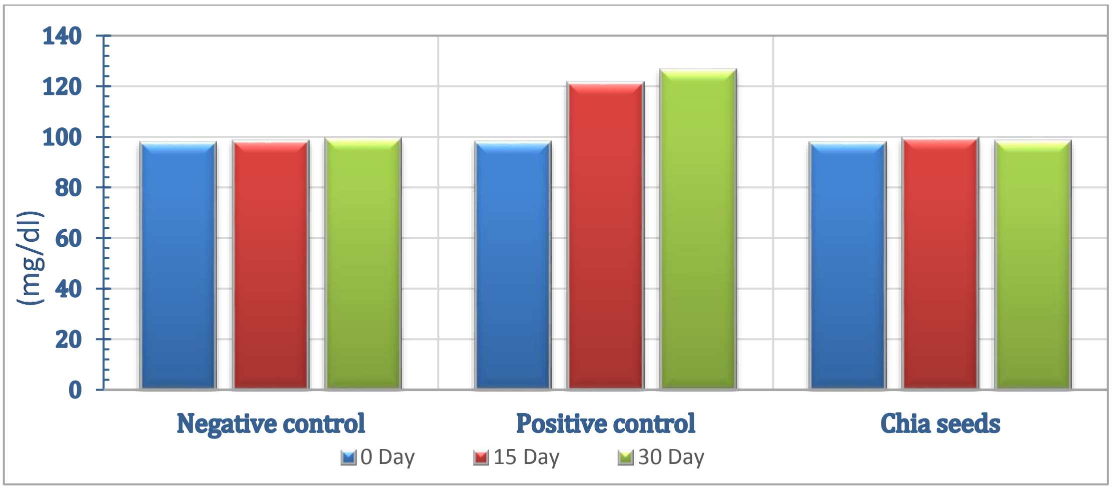

Figure 2: Effect of chia seeds on total cholesterol levels of albino rats (mg/dl).

The plasma total cholesterol level in the negative control group was 97.73 mg/dl at zero time, while in the hyperglycemic group and chia group total cholesterol levels were 97.91 and 97.57 mg/dl respectively. After 30 days of treatment, the total cholesterol level in the negative control group rats was 99.16 mg/dl, while in the hyperglycemic group, the total cholesterol level was 126.41 and decreased significantly to 98.13 mg/dl in the chia group.

Plasma triglycerides levels as indicated in Table 5 and Figure 3 showed that the level of TG in the negative control group was 79.94 mg/dl at zero time, while in the hyperglycemic group and chia group, triglycerides levels were 80.13 and 80.38 mg/dl respectively. After 30 days, the triglycerides level in the negative control group was 81.65 mg/dl, while in the hyperglycemic group, the triglycerides level was 119.74 mg/dl and decreased significantly to 81.34 mg/dl in the chia group.

Table 5: Effect of chia seeds on triglycerides of albino rats.

Group	Triglycerides (mg/dl)		
	0 day	15 day	30 day
Negative control group	79.94±1.09 ^{a}	80.21±0.94 ^{a}	81.65±1.17 ^{a}
Positive control group	80.13±1.07 ^{a}	115.41±0.89^{b}	119.74±1.04^{b}
Chia seeds group	80.38±1.21 ^{a}	83.13±1.43 ^{a}	81.34±1.19 ^{a}

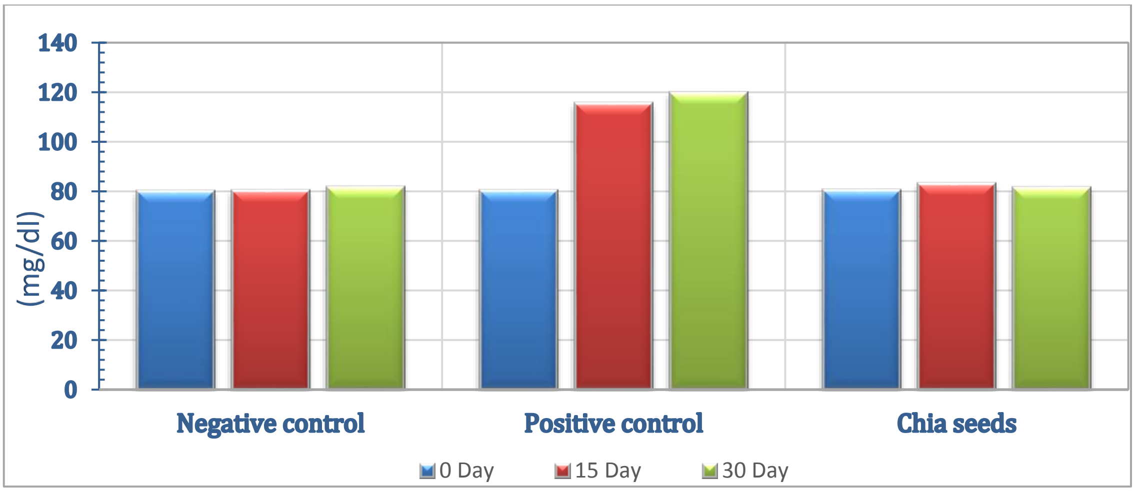

Figure 3: Effect of chia seeds on triglyceride lvels of albino rats (mg/dl).

As shown in Table 6 and Figure 4, the negative control group had a plasma HDL-c level of 45.17 mg/dl at zero time, whereas the hyperglycemic and chia groups had levels of 45.31 and 45.26 mg/dl, respectively. After 30 days, the HDL-c level in the negative control group was 45.73 mg/dl, whereas, in the hyperglycemic group, HDL-c levels were reduced to 40.12 mg/dl and significantly increased to 48.35 mg/dl in the Chia group.

Table 6: Effect of chia seeds on HDL-c of albino rats.

Group	HDL-c (mg/dl)		
	0 day	15 day	30 day
Negative control group	45.17±1.43 [a]	44.97±1.05 [a]	45.73±1.57 [b]
Positive control group	45.31±1.64 [a]	43.84±1.42 [a]	40.12±1.37 [a]
Chia seeds group	45.26±1.71 [a]	47.26±1.22 [b]	48.35±1.67 [c]

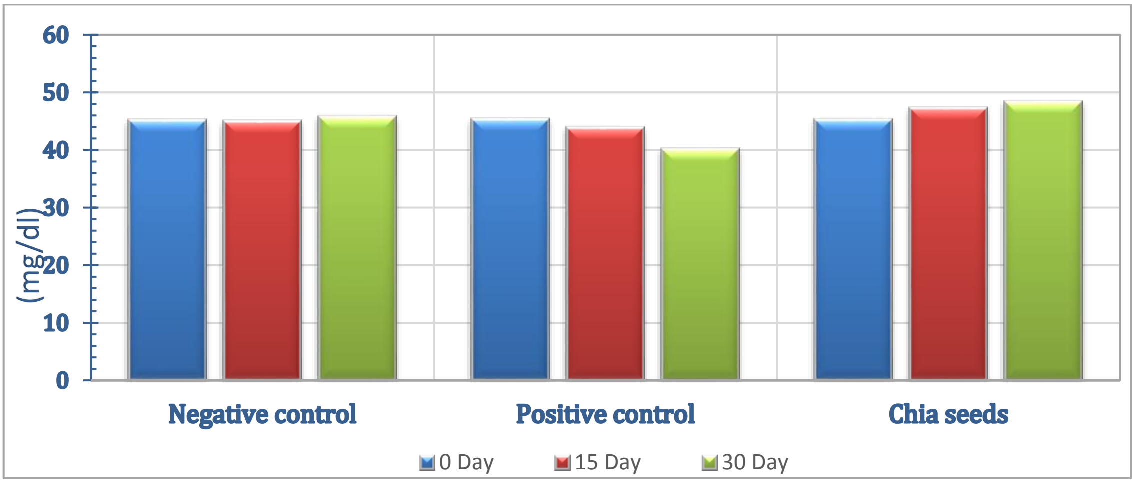

Figure 4: Effect of chia seeds on HDL-c of albino rats (mg/dl).

Plasma LDL-c levels as presented in Table 7 and Figure 5 confirm that in the negative control group was 36.74 mg/dl at zero time, while in the hyperglycemic group and chia group LDL-c levels were 35.94 and 35.92 mg/dl respectively. After 30 days, the LDL-c level in the negative control group was 37.69 mg/dl, while in the hyperglycemic group, the LDL-c level was 67.47 mg/dl and decreased significantly to 36.18 mg/dl in the chia group.

Our findings validate the results presented by Dickens *et al.* (2023), which indicated that the group consuming chia seeds exhibited a 5.75 mg/dl increase in HDL levels and proposed that both chia seeds and oatmeal may be advantageous for the natural enhancement of HDL cholesterol levels.

Table 7: Effect of chia seeds on LDL-c of albino rats.

Group	LDL-c (mg/dl)		
	0 day	15 day	30 day
Negative control group	36.74±0.97 [a]	38.24±1.08 [a]	37.69±0.74 [a]
Positive control group	35.94±1.27 [a]	64.94±1.54 [b]	67.47±0.94 [b]
Chia seeds group	35.92±0.84 [a]	37.87±1.26 [a]	36.18±1.19 [a]

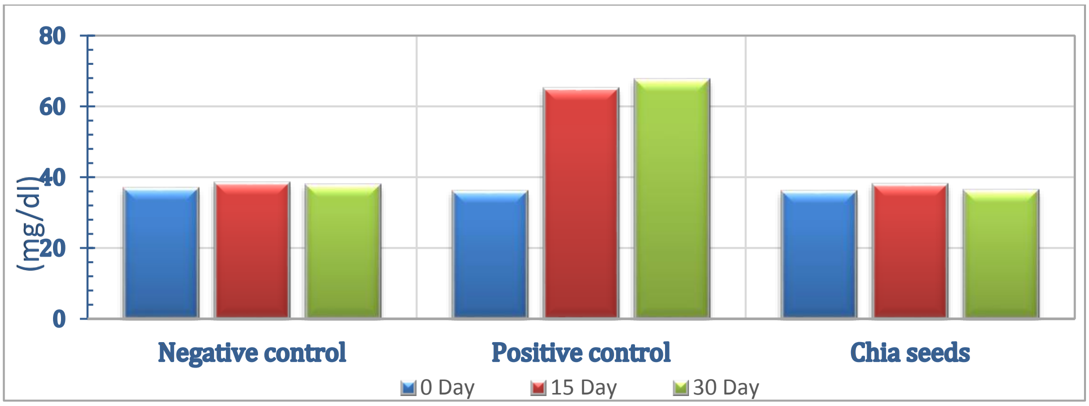

Figure 5: Effect of chia seeds on LDL-c levels of albino rats (mg/dl).

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تأثير بذور الشيا علي ارتفاع سكر الدم في فئران التجارب

محمد عبدالسلام حبيب، يوسف أمين عشوش، صلاح منصور السيد، محمد ممتاز حجازي

قسم الكيمياء الحيوية – كلية الزراعة – جامعة المنوفية – جمهورية مصر العربية

الملخص العربي

تهدف هذة الدراسة إلى دراسة التركيب الكيميائى لبذور الشيا ودراسة مكوناتة من الأحماض الأمينية وتقييم تأثيربروتين بذور الشيا على مستوى الجلوكوز لدى الفئران المصابة بالسكري .كان التركيب الكيميائي للبذور من حيث الرطوبة 5.55٪ والألياف 18.99٪ والرماد 1.92٪ والبروتين الخام 18.16٪ والدهون الكلية 33.59٪ والكربوهيدرات الكلية 21.79٪. كذلك احتوت بذور الشيا علي بعض الأحماض الأمينية ذات الأهمية مثل حمض الجلوتاميك 20.6٪ يليه الأرجينين 12.47٪ ، بينما سجل الأسبارتيك والليوسين 9.57٪ و 7.76٪ على التوالي .أوضحت المعاملة ببروتين بذور الشيا انخفاض مستوى الجلوكوز بشكل ملحوظ بعد ٣٠ يوم من المعاملة من (٢٤٤,١٧ملجم/ديسيلتر) الي(١١٩,٢٤ ملجم/ديسيلتر)، كما انخفض مستوي الجليسريدات الثلاثية من (١١٩,٧٤ ملجم/ديسيلتر) الي (٨١,٣٤ ملجم/ديسيلتر) AST وكذلك الكوليسرول الكلي من (١٢٦,٤١ ملجم/ديسيلتر) الي (٩٨,١٣ ملجم/ديسيلتر) ،وانخفض مستوي HDL-c, LDL-c، مقارنة بالمجموعة المصابة بمرض ارتفاع سكر الدم.

وبناء علي ذلك يمكن اعتبار التغذية علي بروتين بذور الشيا مفيداً لمرضي ارتفاع سكر الدم.

الكلمات المفتاحية: بذور الشيا، ارتفاع سكرالدم، داء السكري، الأحماض الأمينية، مستوى الجلوكوز ، وظائف الكبد.