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Contemplation Impact of Pulp Seeds *Cucurbita Pepo L.* and its Paste on Oxidative Stress in Rats

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Abstract

Cucurbita Pepo L. seeds and seeds paste which are rich sources of phytochemicals and act as a rich source of antioxidants. The most important phytochemicals present in the cucurbits are cucurbitacin's, saponins, carotenoids, phytosterols, and polyphenols. These bioactive phyto-constituents are responsible for the pharmacological effects including antioxidant effect. Aim of this study was to investigate the effect of Cucurbita Pepo L. seeds and seeds paste on rats suffering from oxidative stress. Thirty-six male albino rats were used in the experiment (Sprague-Dawley strain). The animals randomly divided 6 rats each group according to the following the first Group: Rats were fed basal diet and set as negative control. The other rats (n = 30) were fed on basal diet containing monosodium glutamate (120 mg/kg) for induce stress condition. After that, rats further divided into 5 groups (n = 6) each for six weeks as follows: -2nd Group: Rats were fed on basal diet containing monosodium glutamate and set as positive control. 3rd Group: Rats were fed on diet containing monosodium glutamate with addition of Cucurbita Pepo L. seeds 5%. 4th Group: Rats were fed on diet containing monosodium glutamate with addition of Cucurbita Pepo L. seeds 10%. 5th Group: Rats were fed on diet containing monosodium glutamate with addition of Cucurbita Pepo L. seeds paste 5%. 6th Group: Rats were fed on diet containing monosodium glutamate with addition of Cucurbita Pepo L. seeds paste 10%. The experimental period was six weeks; Blood samples were collected. At the end of the experiment, the results showed that using seeds 5% & 10% and seeds paste 5% & 10% in feeding the stressed rats increased (BWG%, FI, FER, Superoxide dismutase (SOD), catalase and glutathione peroxidase (GPx)) with highest results in group fed on 10% seeds paste and decreased (Urea, Creatinine, ALT, AST, IL-6 and INF-gamma) with lowest results in group fed on 10% Cucurbita Pepo L. seeds paste.

Keywords: *Oxidative Stress, Cucurbita Pepo L. Seeds, Pumpkin Seeds, Natural anti-oxidants, Seeds Paste.*

1. Introduction

Excessive burnout symptoms can affect everyone; however, job burnout and work stress affect certain groups disproportionately. It is also significant for students at various educational levels during test seasons with situations of psychological stress. They also experience certain shifts and mood swings as a result of physiological changes during the various phases of development, particularly during the adolescent years. As a result of these pressures, some of them may have headaches, forgetfulness, lack of attention, and lethargy, or/and ignore sufficient and nutritious diet in favor of harmful habits, resulting in the formation of various symptoms of disease states **(Zhan *et al.*, 2019).** Every day, one million employees are away from work owing to workplace stress. It is critical to keep an eye out for signs of extreme exhaustion, especially if you work in a demanding environment. Neglected feelings

of exhaustion, rage, anxiety, and stress might erupt in the workplace and elsewhere **(Zhang *et al.*, 2022).**

Furthermore, as a bodily reaction to the disruption of an equilibrium state, stress has its own negative impacts on health. It contributes to a general decrease in quality of life as well as a variety of illnesses such as hypertension, cardiovascular disease, inflammatory bowel syndrome, and diabetes mellitus. Stress, in particular, can cause extended cortisol release and subsequent activation of immune cells, particularly neutrophils, followed by the formation of free radicals. Nutritional imbalances, inadequate vitamin intake, and high saturated fat consumption can all lead to stress hormone dysregulation and inflammation **(Penttinen *et al.*, 2021).**

Specific nutrients, such as omega-3 polyunsaturated fatty acids (PUFAs), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), and alpha-tocopherol, were found to improve mental health and well-being. DHA levels in the brain decline with age, age-related cognitive alterations can develop from mild cognitive impairment to dementia, and cognition is affected as a result of brain neurodegeneration **(Hu *et al.*, 2017).**

Pumpkin *(Cucurbita Pepo L.)* seeds have health advantages because of its macro and micronutrient content, phytosterols, and antioxidants such as tocopherols and carotenoids **(Benalia *et al.*, 2015).** Furthermore, it is regarded as a great natural source of unsaturated fatty acids like oleic and linoleic. These chemicals have a significant impact on brain health and cognitive performance. Furthermore, they have antioxidant and anti-inflammatory properties that effectively prevent lipid peroxidation **(Lemus-Mondaca *et al.*, 2019).**

This study aimed to illustrate the anti-oxidant, anti-obesity, hepatoprotective, positive effect on invitro antioxidants, anti-inflammatory and nephroprotective effects of *Cucurbita Pepo L.* seeds and its paste on stressed rats.

2. Materials and Methods

Materials:

1. Ingredients for preparing product purchased from the Agriculture Research Center.
2. Diet ingredients, chemicals and kits obtained from different companies such as Alkan, El–Gomhoriya and Biomed Company.
3. Animals; Rats obtained from National Research Center, Dokki, Egypt. The proposal approved by Scientific Committee at National Research Centre (NRC), Egypt. Animal experiments conducted according to the guidelines of Animal Care and Ethics Committee of the NRC.
4. *Cucurbita Pepo L.* seeds obtained from Agriculture Research Center, Giza, Egypt.

Preparation of Product

Cucurbita Pepo L. pulp paste was made by traditional method as described by **(Shibli *et al.*, 2019)** with some modification.

Experimental Design

The experimental animal was done using (36) male rats (Sprague Dawley strain), with body weight 100 ±10 g. The rats were housed in cages under hygienic conditions, at temperature-controlled room 25°C. The food and water were allowed ad-libitum. Basal diet was semi-synthetic and nutritionally adequate (AIN-93 G), vitamins mixture and minerals mixture were prepared as described by **(Reeves *et al.*, 2009).**

The animals randomly divided 6 rats each group according to the following:

Group 1: Rats were fed basal diet and set as negative control.

The other rats (n = 30) were fed on basal diet containing monosodium glutamate (120 mg/kg) for induce stress condition. After that, rats further divided into 5 groups (n = 6) each for six weeks as follows:

Group 2: Rats were fed on basal diet containing monosodium glutamate and set as positive control.

Group 3: Rats were fed on diet containing monosodium glutamate with addition of *Cucurbita Pepo L.* seeds 5%.

Group 4: Rats were fed on diet containing monosodium glutamate with addition of *Cucurbita Pepo L.* seeds 10%.

Group 5: Rats were fed on diet containing monosodium glutamate with addition of *Cucurbita Pepo L.* seeds paste 5%.
Group 6: Rats were fed on diet containing monosodium glutamate with addition of *Cucurbita Pepo L.* seeds paste 10%.

Through the experiment growth rate was followed. Food intake and weight gain were recorded during the experiment. The experiment continued for 6 wks. At the end of the experimental period, rats were fasted overnight, and then the blood was collected under slight ether anesthesia. Blood samples were collected on EDTA. One portion of the blood samples centrifuged at 4000 rpm for 15 min to separate plasma which was stored in the deep freeze at -70°C till analysis of the studied parameters. The rest of the sample collected and allowed to clot then; serum was separated by centrifugation at 4000 rpm for 15 min. The obtained serum was used immediately for routine laboratory investigation. The rest of the serum divided into aliquots and stored at -80^{O}C for further assessment. The organs weight was measured after the end of the experiment.

Chemical Analysis

1. Fatty acids composition of the extracted oil was identified using GC-MS instrument.
2. The phenolic compounds of the extract from oil and seed were identified by HPLC **(Verdi *et al*., 2004).**
3. Chemical composition of the resulted product was analyzed for protein, fat, ash by the method of **(AOAC, 2000).**
4. Organoleptic of the resulted product was evaluated for flavor, body and texture, appearance and overall acceptability according to **(Resurreccion, 2007).**
5. **Nutritional evaluation:** The biological evaluation of the diet was carried by determination of feed intake, body weight gains (BWG %), feed efficiency ratio (FER) and organ weight %, body weight% according to **(Chapman, 1959).**

Biochemical Analysis

1. **Liver function:** Aspartate and alanine aminotransferase (AST) and (ALT) activities were determined calorimetrically according to the method of **(Reitman and Frankel, 1957)** using kits spectrum.
2. **Kidney function:** Plasma urea was estimated as described by **(Fawcett and Soctt 1960)**. Plasma creatinine was determined as shown in **(Bartles *et al*., 1972)**.
3. **Lipid peroxidation:** Lipid peroxides expressed as malondialdehyde (MDA) were estimated as described by **(Ohkawa *et al*., 1979)** using thiobarbituric acid reagent.
4. **The antioxidant enzymes**: Superoxide dismutase (SOD), catalase and glutathione peroxidase (GPx) activity were assaed either in blood or in brain tissue homogenate according to the methods of **(Nishikimi *et al*., 1972; Aebi, 1984; Paglia and Valentine, 1967)**, respectively.
5. Available inflammatory marker such as interleukin 6 and Interferon-gamma were determined using commercially available ELISA assays.

Statistical Analysis

Data was presented as the mean ± S.D. Statistical analysis was performed using SPSS computer program (Graph pad software Inc., San Diego, CA, USA). One-way analysis of variance **(ANOVA)** followed Duncan's multiple tests were done. $P \leq 0.05$ was significant.

3. Results and Discussion

Data presented in **Table (1)** showed the effect of *Cucurbita Pepo L.* seeds and paste with three levels on feed intake (g/day), body weight gain % and feed efficiency ratio in normal rats and stressed rats (Fed on sodium glutamate). The results in this study revealed that, stressed group (Positive control group) showed decreased in feed intake (g/day), body weight gain % and feed efficiency ratio, as compared to rats fed on basal diet (The negative control group), While they were significantly increased in all treated stressed groups as compared with the positive control group.

As illustrated in **Table (2)** it could be observed that, liver enzymes (ALT&AST) were significantly increased in the positive control group as compared with the negative control group (ALT: 90.08±1.46 & 46.28±1.03) & (AST: 104.28±1.35 & 51.64±1.61) respectively. While significant

decrease in liver enzymes in all treating stressed groups as compared with the positive control group with best effect for group treated with 10% seeds paste.

As illustrated in **Table (3)** it could be observed that urea and creatinine concentrations were significantly increased in the control positive group as compared with the negative control group (Urea: 86.02±1.66 & 55.09±1.13) & (Creatinine: 1.09±1.041 & 0.77±1.01) respectively.

While there was a significant decrease in urea and creatinine concentrations in all treated stressed groups as compared with the positive control group with best results for the group treated with 10% seeds paste.

As illustrated in **Table (4)** it could be observed that malondialdehyde (MDA) was significantly increased in the positive control group as compared with the negative control group (1.90±0.02 & 0.83±0.02) respectively. While it was significantly decreased in the treated stressed groups as compared with the positive control group and non-significantly increased as compared with the negative control group.

As illustrated in **Table (5)** it could be observed that antioxidant enzymes activity of Superoxide dismutase (SOD), glutathione peroxidase (GPx) and catalase in the positive control group (729.26±4.12, 752.92±3.65 & 865.89±4.25) respectively was significantly decreased in the blood as compared with the negative control group (1033.86±3.42, 1083.35±5.76 & 1149.14±4.35) respectively. While it was significantly increased in all stressed treated groups as compared with the positive control group with best results for the group fed on 10% seeds paste.

As illustrated in **Table (6)** it could be observed that antioxidant enzymes activity of Superoxide dismutase (SOD), glutathione peroxidase (GPx) and catalase in the positive control group (535.15±11.13, 356.03±4.93 & 345.28±4.89) respectively was significantly decreased in the brain tissues as compared with the negative control group (891.15±8.28, 812.19±4.35a & 839.09±3.46) respectively. While it was significantly increased in all stressed treated groups as compared with the positive control group with best results for the group fed on 10% seeds paste.

As illustrated in **Table (7)** it could be observed that inflammatory markers (IL-6 & INF-Gamma) were significantly increased in the positive control group (23.51±0.84 & 37.58±0.61) respectively as compared with the negative control group (14.21±0.37 & 20.58±1.60) respectively. While INF-gamma & IL-6 were significantly decreased as compared with the positive control group with the lowest result for the group fed on 10 % seeds paste.

Table (8) illustrate our results of chemical compositions of *Cucurbita Pepo L.* seeds.

Table (1): Effect of *Cucurbita Pepe L.* Seeds and Paste on BWG %, FI and FER in Stressed Rats

Parameters / Groups	Initial BW	Final BW	BWG %	FI (g/d/rat)	FER
G_1: Control (-ve)	102.80±2.28^{a}	137.40±2.15^{a}	33.74±1.08^{a}	13.50	0.610±0.000^{a}
G_2: Control (+ve)	97.60±1.28^{a}	106.60±1.96^{c}	9.19±0.67^{b}	11.50	0.019±0.001^{c}
G_3: 5% Seeds	99.20±2.37^{a}	132.99±2.28^{ab}	33.15±1.97^{a}	12.50	0.062±0.000^{a}
G_4: 10% Seeds	100.80±1.93^{a}	129.20±2.35^{ab}	28.21±1.20^{a}	14	0.048±0.001^{ab}
G_5: 5% Seeds Paste	98.00±2.62^{a}	129.80±2.78^{ab}	32.86±2.68^{a}	14.50	0.052±0.006^{ab}
G_6: 10% Seeds Paste	98.20±3.15^{a}	125.60±1.74^{b}	28.47±2.70^{a}	15	0.043±0.006^{b}

All values are represented as means ± SD.
Means with different superscript are significantly different at ($P < 0.05$).

Table (2): Effect of *Cucurbita Pepe L.* Seeds and Paste on Liver Enzymes of Rats

Parameters / Groups	AST	ALT
	(μ /L)	
G_1: Control (-ve)	51.64±1.61^{e}	**46.28±1.03^{e}**
G_2: Control (+ve)	104.28±1.35^{a}	**90.08±1.46^{a}**
G_3: 5% Seeds	89.72±1.93^{b}	**79.75±1.85^{b}**
G_4: 10% Seeds	74.28±1.07^{c}	**56.48±1.17^{d}**
G_5: 5% Seeds Paste	63.75±1.27^{d}	**68.91±1.71^{c}**
G_6: 10% Seeds Paste	59.42±2.47^{d}	**56.44±1.99^{d}**

All values are represented as means ± SD.
Means with different superscript are significantly different at (P < 0.05).

Table (3): Effect of *Cucurbita Pepe L.* Seeds and Paste on Kidney Functions of Rats

Parameters / Groups	Creatinine	Urea
	(mg /dl)	
G_1: Control (-ve)	0.77±1.01^{d}	55.09±1.13^{d}
G_2: Control (+ve)	1.09±1.041^{a}	**8**6.02±1.66^{a}
G_3: 5% Seeds	0.94±1.01^{b}	**72**.18±**3**.75^{b}
G_4: 10% Seeds	0.87±0.91^{bc}	**6**6.20±1.99^{bc}
G_5: 5% Seeds Paste	0.77±1.04^{cd}	**62**.78±**2**.16^{bcd}
G_6: 10% Seeds Paste	0.73±1.00^{cd}	**58**.15±**2**.**18**^{cd}

All values are represented as means ± SD.
Means with different superscript are significantly different at (P < 0.05).

Table 4. Effect of *Cucurbita Pepe L.* Seeds and Paste on Lipid Peroxidation Expressed as Malondialdehyde of Rats

Parameters / Groups	MDA
	(mmol /ml)
G_1: Control (-ve)	0.83±0.02^{d}
G_2: Control (+ve)	1.90±0.02^{a}
G_3: 5% Seeds	1.13±0.03^{b}
G_4: 10% Seeds	1.09±0.04^{b}
G_5: 5% Seeds Paste	1.03±0.01^{bc}
G_6: 10% Seeds Paste	0.96±0.01^{c}

All values are represented as means ± SD.
Means with different superscript are significantly different at (P < 0.05).

Table (5): Effect of *Cucurbita Pepe L.* Seeds and Paste on Antioxidant Enzymes Activity Which Were Assayed in Blood of Rats

Parameters / Groups	Catalase	GPx	SOD
	U/ml		
G_1: Control (-ve)	839.09±3.46^{a}	812.19±4.35^{a}	891.15±8.28^{a}
G_2: Control (+ve)	345.28±4.89^{e}	356.03±4.93^{d}	535.15±11.13^{c}
G_3: 5% Seeds	461.41±5.18^{d}	361.38±4.42^{d}	571.67±5.45^{c}
G_4: 10% Seeds	551.00±4.46^{c}	427.14±4.29^{c}	743.83±6.31^{b}
G_5: 5% Seeds Paste	671.05±2.72^{b}	545.28±5.76^{b}	851.00±4.46^{a}
G_6: 10% Seeds Paste	685.43±2.35^{b}	560.32±4.25^{b}	852.64±5.63^{a}

All values are represented as means ± SD.
Means with different superscript are significantly different at ($P < 0.05$).

Table (6): Effect of *Cucurbita Pepe L.* Seeds and Paste on Antioxidant Enzymes Activity Which Were Assayed in Brain Tissue of Rats

Parameters / Groups	Catalase	GPx	SOD
	U/ml		
G_1: Control (-ve)	1149.14±4.35^{a}	1083.35±5.76^{a}	1033.86±3.42^{a}
G_2: Control (+ve)	865.89±4.25^{d}	752.92±3.65^{e}	729.26±4.12^{e}
G_3: 5% Seeds	879.50±5.71^{d}	758.64±5.67^{e}	741.95±2.33^{de}
G_4: 10% Seeds	959.69±4.46^{c}	850.91±4.39^{d}	767.28±4.66^{d}
G_5: 5% Seeds Paste	975.29±2.43^{c}	875.27±6.19^{c}	834.89±5.15^{c}
G_6: 10% Seeds Paste	1034.47±8.28^{b}	924.42±6.64^{b}	881.38±4.87^{b}

All values are represented as means ± SD.
Means with different superscript are significantly different at ($P < 0.05$).

Table 7. Effect of *Cucurbita Pepe L.* Seeds and Paste on Inflammatory Markers (IL-6 & INF-Gamma) of Rats

Parameters / Groups	IL-6	INF-gamma
	(pg /ml)	
G_1: Control (-ve)	20.58±1.60^{d}	14.21±0.37^{d}
G_2: Control (+ve)	37.58±0.61^{a}	23.51±0.84^{a}
G_3: 5% Seeds	29.57±0.48^{b}	19.32±0.47^{b}
G_4: 10% Seeds	24.26±0.78^{cd}	17.41±0.50^{bc}
G_5: 5% Seeds Paste	25.39±0.78^{bc}	16.97±0.80^{bcd}
G_6: 10% Seeds Paste	22.22±0.59^{cd}	15.25±0.45^{cd}

All values are represented as means ± SD.
Means with different superscript are significantly different at ($P < 0.05$).

Table (8): Chemical Compositions of *Cucurbita Pepe L.* Seeds (g/100 g Fresh Weight

Parameter	*Cucurbita Pepe L.* Seeds
Moisture (%)	6.72
Total lipids (%)	30.76
Protein (%)	35.11
Carbohydrate (%)	5.32
Ash (%)	5.1
Fiber (%)	16.99

According to the data in the **Table (1)** *Cucurbita Pepo L.* seeds and seed paste exhibit antioxidant and anti-obesity properties. *Cucurbita Pepo L.* seeds and paste have been shown to protect against obesity, which is linked to an increased risk of cardiovascular disease **(Ilkun and Boudina, 2013).**

According to the findings in **Table (2)** *Cucurbita* seed and seed paste show antioxidant and hepatoprotective properties. Pumpkin seeds (*Cucurbita Pepo L.*) contain antioxidants **(Nkosi *et al.*, 2006). Makni *et al.* (2008)** identified hypolipidemia as a condition. **Eraslan *et al.* (2013)** found it to be antiatherogenic and hepatoprotective. activities. Pumpkin seed oil (PSO) is high in unsaturated fatty acids, antioxidants, and sterols, according to phytochemical research **(Rabrenovic *et al.*, 2016). Procida *et al.* (2013)** identified amino acids, important fatty acids, -carotenes, certain triterpenes, phytosterols, zinc, and selenium. luteolin, tyrosol, vanillin, vanillic acid, ferulic acid **(Andjelkovic *et al.*, 2010).**

From the above mentioned data in **Table (3)** it could be observed that, *Cucurbita Pepo L.* seed and seed paste have antioxidant and nephroprotective effects. The seed's nephroprotective qualities might be linked to its antioxidant capabilities, which suppress the action of arginase. Nonetheless, as compared to raw *Cucurbita Pepo L.* seeds supplemented diet, roasted *Cucurbita Pepo L.* seeds supplemented food had a much stronger effect on all examined parameters. Arginase has been found to be an essential biomarker in disorders associated with renal injury/harm **(Anadozie *et al.*, 2018; You *et al.*, 2013).** Arginase plays an important function in renal tissue protection, and decreased enzymatic activity has been linked to decreased albuminuria, oxidative stress, plasma creatinine levels, and kidney macrophage recruitment **(You *et al.*, 2013).**

Free radicals, such as superoxide radical, can combine with NO to form peroxynitrite, reducing NO bioavailability. Expanded arginase activity in the kidney might thus reduce L-arginine accessibility to nitric oxide synthase, resulting in a decrease in NO generation and an increase in superoxide formation owing to eNOS uncoupling, as discovered in the current study **(Kim *et al.*, 2009).**

A roasted *Cucurbita Pepo L.* seed diet may increase the availability of arginine, which is required for NO generation. As a result, arginine availability modulates the NO/cGMP pathway **(Anadozie *et al.*, 2018; Reddy *et al.*, 2015).** This study backs up previous findings that dietary constituents and plant products decrease arginase activity in toxic chemicals or xenobiotics-induced kidney injury **(Akinyemi *et al.*, 2017; Akomolafe *et al.*, 2019; Anadozie *et al.*, 2018).**

From the above mentioned data in **Table (4)** it could be observed that, *Cucurbita Pepo L.* seed and seed paste contain antioxidant and anti-obesity properties, which are important because obesity, insulin resistance, and type 2 diabetes are all linked to an elevated risk of CV disease **(Ilkun and Boudina, 2013)**. Several critical phenomena associated with CV illness can be induced by the connected OS, including lipid buildup, increased fibrosis and stiffness, altered calcium homeostasis, aberrant autophagy, changed substrate consumption, and mitochondrial dysfunction **(Ilkun and Boudina, 2013; Rani *et al.*, 2016).** In hypercholesterolemic rats, pumpkin seed oil was tested for its cardiovascular preventive impact. The delivery of pumpkin seed oil resulted in a decrease in the ratio of triglycerides to high-density cholesterol, indicating antiatherogenic (CV protecting) properties. This protective effect was linked to the presence of phytosterols, unsaturated fatty acids, phenolics, and carotenoids, which combined decreased the lipids as a result of pumpkin seed oil's strong antioxidant nature.

From the above mentioned data in **Tables (5 & 6)** it could be observed that, *Cucurbita Pepo L.* seed and seed paste can increase the activity of invitro antioxidants, which is important because oxidative stress has been linked to a variety of chronic illnesses and associated consequences, including diabetes, obesity, CVD, and cancer **(Yadav *et al.*, 2017).**

Several laboratory investigations have demonstrated antioxidant activity **(Caili *et al.*, 2006).** The phenolic components in *Cucurbita Pepo L.* seed methanolic extract are more concentrated. Their overall phenolic content influences their radical scavenging ability. *Cucurbita Pepo L.* seed extract injection raised the serious and hepatic activities of superoxide dismutase and glutathione peroxidase in mice while decreasing malonaldehyde content **(Yadav *et al.*, 2017).** Pumpkin polysaccharide has

also been shown to improve superoxide dismutase and glutathione peroxidase activity while decreasing malondialdehyde concentration in tumor-containing mouse serum.

From the above-mentioned data in **Table (7)** it could be observed that, *Cucurbita Pepe L.* seed and seed paste have antioxidant and anti-inflammatory effects. TNF- α and IL-6 have both been implicated in the pathophysiology of depression **(Taraz *et al.*, 2015; Pedraz-Petrozzi *et al.*, 2020).** Mice lacking IL-6 or TNF- α receptors have been shown to be resistant to depressive behaviors **(Viana *et al.*, 2010).** As a result, they were evaluated in this study, and it was discovered that TNF- α and IL-6 levels were considerably higher in the serum of rats exposed to CUMS for 4 weeks, indicating that they were depressed. **Numakawa *et al.* (2014)** found this in rats with behavioral despair, while **Taraz *et al.* (2015)** found it in people with depression. In this investigation, treatment of pumpkin extract was related with improved depressed behavior, as evidenced by a substantial reduction in FST immobility time and a significant decrease in corticosterone and inflammatory cytokines TNF- α and IL-6 in the blood. **Kim *et al.* (2016)** confirmed these findings by reporting that SSP for 28 days reduced the number of inflammatory cytokines in depressed rats. Pumpkin's anti-inflammatory action may be ascribed to active components such as oleic and palmitic acids, as well as estradiol.

Our Results for Chemical Compositions of *Cucurbita Pepe L.* Seeds: Moisture: 6.72%, Total lipids: 30.76%, Protein: 35.11%, Carbohydrate: 5.32%, Ash: 5.1% and Fiber: 16.99%. While chemical compositions of *Cucurbita Pepe L.* seeds according to **Kim *et al.* (2012)** were: Moisture: 6.41%, Total lipids: 37.98%, Protein: 26.88%, Carbohydrate: 10.22%, Ash: 5.5% and Fiber: 12.84%. We found that DPPH Antioxidant activity was 93.4 % and (TPC) total phenolic compounds: 205 mg GAE/100 g.

4. Conclusion

In conclusion, the results suggested that ingestion of high concentrations from *Cucurbita Pepo L.* seeds and seeds paste increases their protective effect against oxidative stress hazards so the current study recommends intake of appropriate amounts from high concentrations from *Cucurbita Pepo L.* seeds and seeds paste Daily particularly in patients suffering from oxidative stress disorders.

The results showed that using (5% & 10%) levels of *Cucurbita Pepo L.* seeds paste has a more potent anti-oxidant effect than using (5% & 10%) *Cucurbita Pepo L.* seeds.

References:

Aebi, H. (1984): Catalase in vitro, isolation, purification, characterization, and assay of antioxygenic enzymes. *Methods Enzymol*. 105:121-126.

Akinyemi, A., Onyebueke, N., Faboya, O., Onikanni, S., Fadaka, A., and Olayide, I. (2017): Curcumin inhibits adenosine deaminase and arginase activities in cadmium-induced renal toxicity in rat kidney. *Journal of Food and Drug Analysis*, 25(2), 438–446.

Akomolafe, S., Olasehinde, T., Adewale, O., and Ajayi, O. (2019): Curcumin improves biomolecules associated with renal function and attenuates oxidative injury and histopathological changes in potassium-induced toxicity in rats' kidney. *Biological Trace Element Research*.

Anadozie, S., Akinyemi, J., Agunbiade, S., Ajiboye, B., and Adewale, O. (2018): Bryophyllum pinnatum inhibits arginase II activity and prevents oxidative damage occasioned by carbon tetrachloride (CCl4) in rats. *Biomedicine & Pharmacotherapy*, 101, 8–13.

Andjelkovic, M., Van-Camp, J., Trawka, A., and Vehre, R. (2010): Phenolic compounds and some quality parameters of pumpkin seed oil. *Eur J. Lipid Sci Technol*. 112(2):208-17.

AOAC. (2000): Official methods of analysis, 3rd ed.; Washington, DC, 2000.

Bartels, H., Bohner, M., and Heierli, C. (1972): "Serum creatinine determination without protein precipitation." *International Journal of Clinical Chemistry*; 37:193-196.

Benalia, M., Djeridane, A., Gourine, N., Nia, S., Ajandouz, E., and Yousfi, M. (2015): Fatty acid profile, tocopherols content and antioxidant activity of algerian pumpkin seeds oil *(Cucurbita pepo L.). Mediterranean Journal of Nutrition and Metabolism,* 8(1), 9-25.

Caili, F., Huan, S., and Quanhong, L. (2006): A Review on Pharmacological Activities and Utilization Technologies of Pumpkin. 73–80.

Chapman, G., Castilla, R., and Campbell, A. (1959): Evaluation of protein in food. Method for the determination of protein efficiency ratio. *Canadian Journal of Biochemistry and Physiology.*; 37:679-686.

Eraslan, G., Kanbur, M., Aslan, Ö., and Karabacak, M. (2013): The antioxidant effects of pumpkin seed oil on subacute aflatoxin poisoning in mice. Environ Toxicol. 28(12):681-8.
Fawcett, K., and Scott, J. (1960): "A Rapid and Precise Method for The Determination of Urea." *Journal of Clinical Pathology.;* 13:156-159.
Hu, J., Liu, Z., and Zhang, H. (2017): Omega-3 fatty acid supplementation as an adjunctive therapy in the treatment of chronic kidney disease: a meta-analysis. *Clinics*.;72(1):58-64.
Ilkun, O., and Boudina, S. (2013): Cardiac dysfunction and oxidative stress in the metabolic syndrome: An update on antioxidant therapies. *Current Pharmaceutical Design*, 19(27), 4806–4817.
Kim, J., Bugaj, L., Oh, Y., Bivalacqua, T., Ryoo, S., Soucy, K., and Berkowitz, D. (2009): Champion HC, Berkowitz DE. Arginase inhibition restores NOS coupling and reverses endothelial dysfunction and vascular stiffness in old rats. *Journal of Applied Physiology*, 107, 1249–1257.
Kim, M., Kim, E., Kim, Y., Choi, C., and Lee, B. (2012): Comparison of the chemical compositions and nutritive values of various pumpkin (Cucurbitaceae) species and parts. Nutrition Research and Practice, 6(1), 21–27.
Kim, N., Kim, H., Kim, M., Kim, H., and Jeong, H. (2016): Improvement of Depressive Behavior by Sweetme Sweet Pumpkin™ and its Active Compound, β-carotene. *Life Sci.* 147, 39–45.
Lemus-Mondacal, R., Marin, J., and Rivas, J. (2019): Pumpkin seeds *(Cucurbita maxima).* A review of functional attributes and by-products. *Revista Chilena de Nutricion*, 46(6): 783-791.
Makni, M., Fetoui, H., Gargouri, N., Garoui, E., Jaber, H., Makni, J., Boudawara, T., and Zeghal, N. (2008): Hypolipidemic and hepatoprotective effects of flax and pumpkin seed mixture rich in omega-3 and omega-6 fatty acids in hypercholesterolemic rats. Food Chem Toxicol. 46(12):3714-20.
Nishikimi, M., Roa, N., and Yogi, K., (1972): The occurrence of superoxide anion in the reaction of reduced phenazine methosulfate and molecular oxygen. *Biochem. Bioph. Res. Common.*, 46: 849-854.
Nkosi, C., Opoku, A., and Terblanche, S. (2006): Antioxidative effects of pumpkin seed (*Cucurbita Pepo L.*) protein isolate in CCl4- induced liver injury in low-protein fed rats. Phytother Res. 20(11):935-40.
Nnadozie, J., Chijioke, U., Okafor, O., Olusina, D., Oli, A., Nwonu, P., Mbagwu, H., and Chijioke, C. (2019): Chronic toxicity of low dose monosodium glutamate in albino Wistar rats. *BMC Research Notes*, 12, (593);1-7.
Numakawa, T., Richards, M., Nakajima, S., Adachi, N., Furuta, M., and Odaka, H. (2014): The Role of Brain-Derived Neurotrophic Factor in Comorbid Depression: Possible Linkage with Steroid Hormones, Cytokines, and Nutrition. *Front. Psychiatry* 5, 136.
Ohkawa, H., Ohishi, N., and Yagi, K. (1979): Assay for Lipid Peroxides in Animal Tissues by Thiobarbituric Acid Reaction; Anal. *Biochem*., 95: 351-358.
Paglia, D., and Valentine W. (1967): Studies on the quantitative characterization of erythrocyte glutathione peroxidase. *Journal of Laboratory and Clinical Medicine* 70:158-169.
Pedraz-Petrozzi, B., Neumann, E., and Sammer, G. (2020): Pro-inflammatory Markers and Fatigue in Patients with Depression: A Case-Control Study. *Sci. Rep.* 10, 66532–66536.
Penttinen, M., Virtanen, J., Laaksonen, M., Erkkola, M., Vepsäläinen, H., Kautiainen, H., and Korhonen, P. (2021): The Association between Healthy Diet and Burnout Symptoms among Finnish Municipal Employees. *Nutrients*, 13, 2393.
Procida, G., Stancher, B., Cateni, F., and Zacchigna, M. (2013): Chemical composition and functional characterisation of commercial pumpkin seed oil. *J Sci Food Agric*. 93(5):1035-41.
Rabrenovic, B., Vujasinovic, V., Novakovic, M., Vujasinović, V., Čorbo, S., and Basić, S. (2016): Comparative review of the nutritional value of cold-pressed pumpkin (*Cucurbita Pepo L.*) seed oil of different origins. Hem Ind. 70:59-65.
Rani, V., Deep, G., Singh, R., Palle, K., and Yadav, U. (2016): Oxidative stress and metabolic disorders: Pathogenesis and therapeutic strategies. *Life Sciences*, 148, 183–193.
Reddy, Y., Kiranmayi, V., Bitla, A., Krishna, G., Srinivasa-Rao, P., and Sivakumar, V. (2015): Nitric oxide status in patients with chronic kidney disease. *Indian Journal of Nephrology*, 25(5), 287–291.
Reeves, P., Nielsen, F., and Fahey, G., (1993): AIN-93 purified diets for laboratory rodents: final report of the American Institute of Nutrition ad hoc writing committee on the reformulation of the AIN-76A rodent diet." *Journal Nutr.* 123, 1939-51
Reitman, S. and Frankel, S., (1957): Colorimetric method for aspartate and alanine amino transferases. *Am. Journal of Clinical Pathology.* 28, 56-63.
Resurreccion, A. (2007): Physicochemical and sensory characteristics of peanut pastes affected by processing conditions. *Journal of Food Processing and Preservation* 17(5):321 - 336.
Shibli1, S., Siddique1, F., Raza, S., Ahsan, Z., and Raza, I. (2019): Chemical Composition and Sensory Analysis of Peanut Butter from Indigenous Peanut Cultivars of Pakistan. *Pakistan Journal of Agricultural Research*, (32)1; 159-

Taraz, M., Taraz, S., and dashti-Khavidaki, S. (2015): Association between Depression and Inflammatory/anti-Inflammatory Cytokines in Chronic Kidney Disease and End-Stage Renal Disease Patients: A Review of Literature. *Hemodial. Int.* 19, 11–22.

Verdi, L., Brighente, I., Schripsema, J., Braz Filho, R., Pizzolatti, M. (2004): Kaurene diterpenes and flavonoids from Baccharis illinita flowers. *Biochemical Systematics and Ecology;* 32:837–840.

Viana, A., Maciel, I., Dornelles, F., Figueiredo, C., Siqueira, J., and Campos, M. (2010): Kinin B1 Receptors Mediate Depression-like Behavior Response in Stressed Mice Treated with Systemic *E.Coli* Lipopolysaccharide. *J. Neuroinflammation* 7, 98.

Yadav, M., Jain, S., Tomar, R., Prasad, G., and Yadav, H. (2017): Medicinal and biological potential of pumpkin: an updated review Nutrition Research Reviews Nutrition Research Reviews. 184–190.

You, H., Gao, T., Cooper, T., Morris, S., and Awad, A. (2013): Arginase inhibition mediates renal tissue protection in diabetic nephropathy by a nitric oxide synthase 3-dependent mechanism. *Kidney International*, 84(6), 1189–1197.

Zhan, J., Yang, H., Jiang, X., Lu, H.; Guo, X., and Hong, Y. (2019)**:** Analysis of Students' Nutritional Status of in the "Nutrition Improvement Program for Rural Compulsory Education Students" Area. *Advances in Social Science, Education and Humanities Research,* 371; 363-367.

Zhang, H., Chen, X., Bi, M., Yang, J., Cai, J., and Zhu, Y. (2022): Cadmium exposure triggers oxidative stress, necroptosis, Th_1/Th_2 imbalance and promotes inflammation through the TNF-α/NF-κB pathway in swine small intestine. *Journal of Hazardous Materials*, 421, 126704.