

## Study the effect of watermelon white rind extract as decreased the detrimental of soybean oil in vivo.

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### ABSTRACT

Dried watermelon white rind was subjected to mycotoxin determination to assure its safe usage. Results ensured that the rind was free from aflatoxin and ochratoxin. Watermelon white rind aqueous extract contained iron, copper and potassium at concentration range of 3.4, 0.53 and 45.51 ppm, respectively. While, the amount of Chrome and Selenium were 14.2 and 98.51 µg/Kg, respectively. Soybean oil had free fatty acid, peroxide value, iodine number and anisidine value of 0.43%, 13.62 meq O<sub>2</sub>/Kg, 132 and 0.7, respectively. GC-MS analysis of soy oil ascertained the presence of twenty-four compounds: linoleic acid, methyl ester (25.27%), monensin (15.75%), elaidic acid (9.24%), nonadecanoic acid, methyl ester (7.04%), cis-13-eicosenoic acid (4.92%), cis-vaccenic acid (4.68%), linoleic acid (4.67%), palmitoleic acid (4.46%), 9-tetradecenal (4.42%) and cysteine (4.18%) were the most predominant. Fatty acid profile of the oil showed that the ratio of saturated fatty acid to unsaturated fatty acids was 1:5. Biological experiment was designed to evaluate safety usage and impact of soybean oil on different blood parameters of rats for two months' interval period. Rats fed diet prepared by soybean oil had a decreased calcium level in comparison with negative control ( $p < 0.05$ ). Supplementation with watermelon white rind aqueous extract rendered calcium level to normal status as negative control. Phosphorus level wasn't affected by soya oil.

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*Keywords: watermelon; GC-MS; blood parameters; calcium level.*

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### 1. INTRODUCTION

Watermelon (*Citrullus lanatus* var. *lanatus*, family Cucurbitaceae) is a flowering plant originally from southern Africa. The white rind is thrown as unused-agro waste. Rind constitutes 30% of the weight of whole watermelon fruit.

[33] cited that ethanolic and aqueous extracts of watermelon white rind possessed antibacterial activity against *E. coli* and *Salmonella* sp., Gas Chromatography-Mass Spectrometry analysis revealed the existence of methionine, L-Aspartic acid, Glycyl-D-asparagine, 9-Cis-Retinoic acid, Stearic acid allyl ester and Ascorbic acid permethyl that contributed to its antibacterial activity.

The rind had total antioxidant activity of 297 mg AAE/100g, total phenols content of 139 mg GAE/100g and total flavonoids of 40.4 mg QE/100g. FRAP assay indicated the high reducing ability of the rind. Crude protein content amounted to 13.3%, crude fiber (14.7%) and fat (2.11%). The rind is a source of iron (30.4 mg/kg), potassium (6.94%), copper (9.4 mg/kg), chromium (85 µg/100g) and selenium (542 µg/100g). Unsaturated fatty acid amounted to 81.2%. Vitamins A and E valued 383.44 µg/100g and 4.72 mg/100g, respectively [ 10]. Wastes are source of sugars, minerals, organic acids, dietary fiber, and bioactive compounds [ 6 ] .

Soybean oil affected negatively bone structure as reported by [ 5].

A study investigated the adverse effect of soybean oil in rat found that oil induced significant fatty liver [ 13].

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## 2. MATERIAL AND METHODS

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Watermelon white rind was cut into small pieces, dried at 40°C and pulverized into fine powder.

### Preparation of white rind aqueous extract:

One gram of dried powder was mixed with one liter of hot water, stirred, filtered and used as the sole source of fluid.

### Determination of Aflatoxin and Ochratoxin

Total Aflatoxin and Ochratoxin were determined according to AOAC [ 3].

### Elemental analysis of rind aqueous extract

Iron, copper, potassium, chrome and selenium were determined according to AOAC [2].

### **Chemical analysis of soybean oil**

Quality of oil was assessed by determining anisidine value, iodine number, peroxide value and free fatty acid according to AOAC [4]. Fatty acid composition was determined according to AOAC [4].

GC-MS analysis of soybean oil was carried out using GC (Agilent Technologies 7890A). The components were verified by matching their mass spectra and retention time with the database of National Institute of Standard and Technology (NIST) library.

### **Biological experiment**

Eighteen rats were distributed into three groups:

Group (1) served as negative control and fed normal diet [ 9] and supplied with drinking water

Group (2) served as positive control fed normal diet to which 150 ml soybean oil was added per kilo and supplied with drinking water.

Group (3) fed diet as group (2) supplied with aqueous watermelon white rind extract.

Diet and fluids were supplied *ad-libitum* for all groups.

At the end of the experiment, blood samples were collected, centrifuged at 4000 rpm and serum was subjected to the analysis of calcium and phosphorus.

### **3. RESULTS AND DISCUSSION**

93 elemental analysis of watermelon white rind aqueous extract (Table 1) ensured the  
94 presence of iron (3.4 ppm), copper (0.53 ppm), potassium (45.5 ppm), chrome (14.2  
95  $\mu\text{g/Kg}$ ) and selenium (98.5  $\mu\text{g/Kg}$ ).

96 Data in Table (2) revealed that soybean oil had anisidine value of 0.7, iodine number  
97 131.97, free fatty acid 0.43% and peroxide value of 13.62 meq  $\text{O}_2/\text{Kg}$ .

98 Twenty-four compounds were detected in the GC-MS chromatogram of soybean oil.  
99 Linoleic acid (25.27%) was the most predominant in the tested oil, followed by  
100 monolinolenic acid (15.75%), elaidic acid (9.24%), nonadecanoic acid (7%), cis-vaccenic acid  
101 (4.67%), linoleic acid (4.67%), palmitoleic acid (4.46%), 9-tetradecenol (4.42%) and  
102 cysteine (4.18%) and accounted for 59.34% of oil constituent (Table 3).

103 In Table (4), fatty acid profile of soybean oil showed the existence of linoleic  
104 acid (44.28%), oleic acid (22.85%), linoleic acid (6.2%) and gadolic acid (0.21%) as  
105 unsaturated fatty acids accounting for 83.54% of total oil content. Saturated fatty acids  
106 comprised palmitic acid (10.99%), stearic acid (4.82%), arachidic acid (0.36%) and  
107 behenic acid (0.29%) representing 16.46% of soybean oil content. These results are in  
108 accordance with [ 7] who stated that soybean had low level of saturated fat and high  
109 content of linoleic acid [1 ].

110 As shown in Table (5), a significant difference ( $p<0.05$ ) existed between negative  
111 control (G1) and rats group fed diet with soybean oil (G2). A decrease in calcium level  
112 was observed indicating that soybean oil affected calcium blood level.

113 Soybean had high phytate level [ 12]. Phytates can block the uptake of essential  
114 minerals as calcium, copper, iron, zinc and magnesium in intestinal tract that may  
115 contribute to mineral deficiencies [ 8].

116 There was non-significant difference between negative control (G1) and Group 3 fed  
117 soybean oil and drunk rind extract, nor between G2 and G3.

118 Data revealed that phosphorus blood level was not affected by any treatment and non-  
119 significant differences existed between G1 and both groups G2 and G3.

120 The extraction of plant material and isolation of biologically active compounds are  
121 essential to understand their role in disease prevention and treatment.

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**Table (1): Elemental analysis of watermelon rind aqueous extract**

Element	Result
Copper (ppm)	0.5
Iron (ppm)	3.4
Potassium (ppm)	45.5
Chrome ( $\mu\text{g/Kg}$ )	14.2
Selenium ( $\mu\text{g/Kg}$ )	98.5

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**Table (2): Chemical evaluation of soy oil**

Tested parameters	Result
Free fatty acid (%)	0.43
Peroxide number (meq O <sub>2</sub> /Kg)	13.62
Iodine number	131.8
Anisidine value	0.7

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**Table (3): GC-MS analysis of soy oil**

RT	Compound name	Area sum (%)
3.88	Chicoric acid	0.29
5.7	Phytanic acid	0.59
6.187	3,2',4',5'-Tetramethoxyflavone	0.27
8.04	Gardenin	0.49
8.96	Isovitexin	0.59
11.7	Lutein	1.33
12.03	Stevioside	0.57
13.23	Hexadecanoic acid, methyl ester	2.63
13.43	Pentadecanoic acid	0.73
13.5	Monensin	15.75
13.9	Zearalenone	1.59
14.17	Oleic acid	2.83
14.35	Cis-vaccenic acid	4.68

14.52	Linoleic acid, methyl ester	25.27
14.59	Elaidic acid	9.24
14.66	Cis-13-eicosenoic acid	4.92
14.75	Nonadecanoic acid, methyl ester	7.0
14.93	Linoleic acid	4.67
15.14	Quinine	0.5
15.33	3-(3,4-dimethoxyphenyl)-4,6-dimethylcoumarin	0.98
15.9	Di- $\gamma$ -linolenin	1.97
16.009	Palmitoleic acid	4.46
16.04	Cystine	4.18
16.79	9-tetradecenal, (Z)-	4.42

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**Table (4): Fatty acid analysis of soybean oil**

Fatty acid	Classification	Relative distribution
Palmitic acid C16:0	Saturated fatty acid	10.99%
Stearic acid C18:0	Saturated fatty acid	4.82%
Arachidic acid C20:0	Saturated fatty acid	0.36%
Behenic acid C22:0	Saturated fatty acid	0.29%
Oleic acid C18:1n9	Unsaturated fatty acid	22.85%
Linoleic acid C18:2n6	Unsaturated fatty acid	54.28%
Linolenic acid C18:3n3	Unsaturated fatty acid	6.2%
Gadolic acid C20:1n9	Unsaturated fatty acid	0.21%

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**Table (5): Serum calcium and phosphorus levels in treated rat groups**

Parameters \ Groups	Group 1 (n=6)	Group 2 (n=6)	Group 3 (n=6)
Calcium (mg/dl)	13.2 $\pm$ 0.64	11.3 $\pm$ 0.48 *	12.8 $\pm$ 0.62
Phosphorus (mg/dl)	10.5 $\pm$ 0.66	10.38 $\pm$ 0.76	11.96 $\pm$ 0.44

\*Significant difference (p<0.05) in comparison with negative control

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#### 4. CONCLUSION

Soybean oil decreased blood calcium level, while phosphorus was stable in all treated groups. Supplementation with watermelon white rind aqueous extract rendered calcium level to normal status as negative control.

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