Original research paper

Watermelon white rind extract decreased the detrimental effect of soybean oil *In-Vivo*

Aim: To study the effect of white rind extract on decreasing soybean oil impact on calcium and phosphorous blood levels *In-Vivo.*

Method: Dried watermelon white rind was directed to mycotoxin and elemental determinations to assure its safe usage. Soybean oil was subjected to fatty acid and GC-MS analysis. Biological experiment was conducted using male albino rats fed diet prepared by soybean oil and supplied with aqueous watermelon white rind extract for two months' interval period. At the end of the experiment, blood calcium and phosphorus were determined.

Results: The rind was free from aflatoxin and ochratoxin. Watermelon white rind aqueous extract contained iron, copper, potassium, chromium and selenium at concentration ranges of 3.4, 0.53, 45.51, 0.0142 and 0.0985 ppm, respectively.

Soybean oil had free fatty acid, peroxide value, iodine number and anisidine value of 0.43%, 13.62 meq O_2/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.

Conclusion: 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 white rind, fatty acid and GC-MS analysis, calcium and phosphorous blood levels.

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.

Ola *et al.* [1] 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 1394 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 (6.95%), copper (9.4 mg/kg), chromium (85µg/100g) and selenium (542µg/100g). Unsaturated fatty acid amounted to 81.2%. Vitamins A and E valued 383.44 µg/100g and 37.72 mg/100g, respectively [2]. Wastes are source of sugars, minerals, organic acids, dietary fiber, and bioactive compounds [3] .

Soybean oil affected negatively bone structure as reported by [4].

A study investigated the adverse effect of soybean oil in rat found that oil induced significant fatty liver [5].

In the present work, biological experiment was designed to evaluate safety usage of watermelon white rind extract on decreasing soybean oil impact on calcium and phosphorous blood levels *In-Vivo.*

MATERIAL AND METHODS

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 Okratoxin

Total Aflatoxin and Ochratoxin were determined according to AOAC [6].

Elemental analysis of rind aqueous extract

Iron, copper, potassium, chrome and selenium were determined according to AOAC [7].

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 [7]. Fatty acid composition was determined according to AOAC [7].

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 [8] 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.

RESULTS AND DISCUSSION

Elemental analysis of watermelon white rind aqueous extract (Table 1) ensured the presence of iron (3.4 ppm), cupper (0.53 ppm), potassium (45.5 ppm), chromium (0.0842 ppm) and selenium (0.0985 ppm).

Data in Table (2) revealed that soybean oil had anisidine value of 0.7, iodine number 131.89, free fatty acid 0.43% and peroxide value of 13.62 meq O_2/Kg.

Twenty-four compounds were detected in the GC-MS chromatogram of soybean oil. Linoleic acid (25.27%) was the most predominant in the tested oil, followed by monensin (15.75%), elaidic acid (9.24%), nonadecanoic acid (7%), cis-vaccenic acid (4.68%), linoleic acid (4.67%), palmitoleic acid (4.46%), 9-tetradecenol (4.42%) and cystine (4.18%) and accounted for 59.34% of oil constituent (Table 3).

As in Table (4), fatty acid profile of soybean oil showed the existence of linoleic acid (54.28%), oleic acid (22.85%), linoleic acid (6.2%) and gadolic acid (0.21%) as unsaturated fatty acids accounting for 83.54% of total oil content. Saturated fatty acids comprised palmitic acid (10.99%), stearic acid (4.82%), arachidic acid (0.36%) and behenic acid (0.29%) representing 16.46% of soybean oil content. These results are in accordance with Friedman and Brandon [9] who stated that soybean had low level of saturated fat and high content of linoleic acid [10].

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

Soybean had high phytate level [11]. Phytates can block the uptake of essential minerals as calcium, copper, iron, zinc and magnesium in intestinal tract that may contribute to mineral deficiencies [12].

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

Data revealed that phosphorus blood level was not affected by any treatment and non-significant differences existed between G1and both groups G2 and G3.

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

CONCLUSION

Watermelon white rind aqueous extract is a source of iron, copper, potassium, chromium and selenium. 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.

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

Element	Result
Copper (ppm)	0.5
Iron (ppm)	3.4
Potassium (ppm)	45.5
Chromium (ppm)	0.0142
Selenium (ppm)	0.0985

Table (2): Chemical evaluation of soy oil

Tested parameters	Result
Free fatty acid (%)	0.43
Peroxide number (meq O_2/Kg)	13.62
Iodine number	131.8
Anisidine value	0.7

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-γ-linolenin	1.97
16.009	Palmitoleic acid	4.46
16.04	Cystine	4.18
16.79	9-tetradecenal, (Z)-	4.42

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 C20:1ω9	Unsaturated fatty acid	0.21%

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±0.64	11.3±0.48 *	12.8±0.62
Phosphorus (mg/dl)	10.5±0.66	10.38±0.76	11.96±0.44

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

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