Physiochemical Characterization of Seed, Oil and Meal from Bauhinia purpurea grown in moderate region of Pakistan

By

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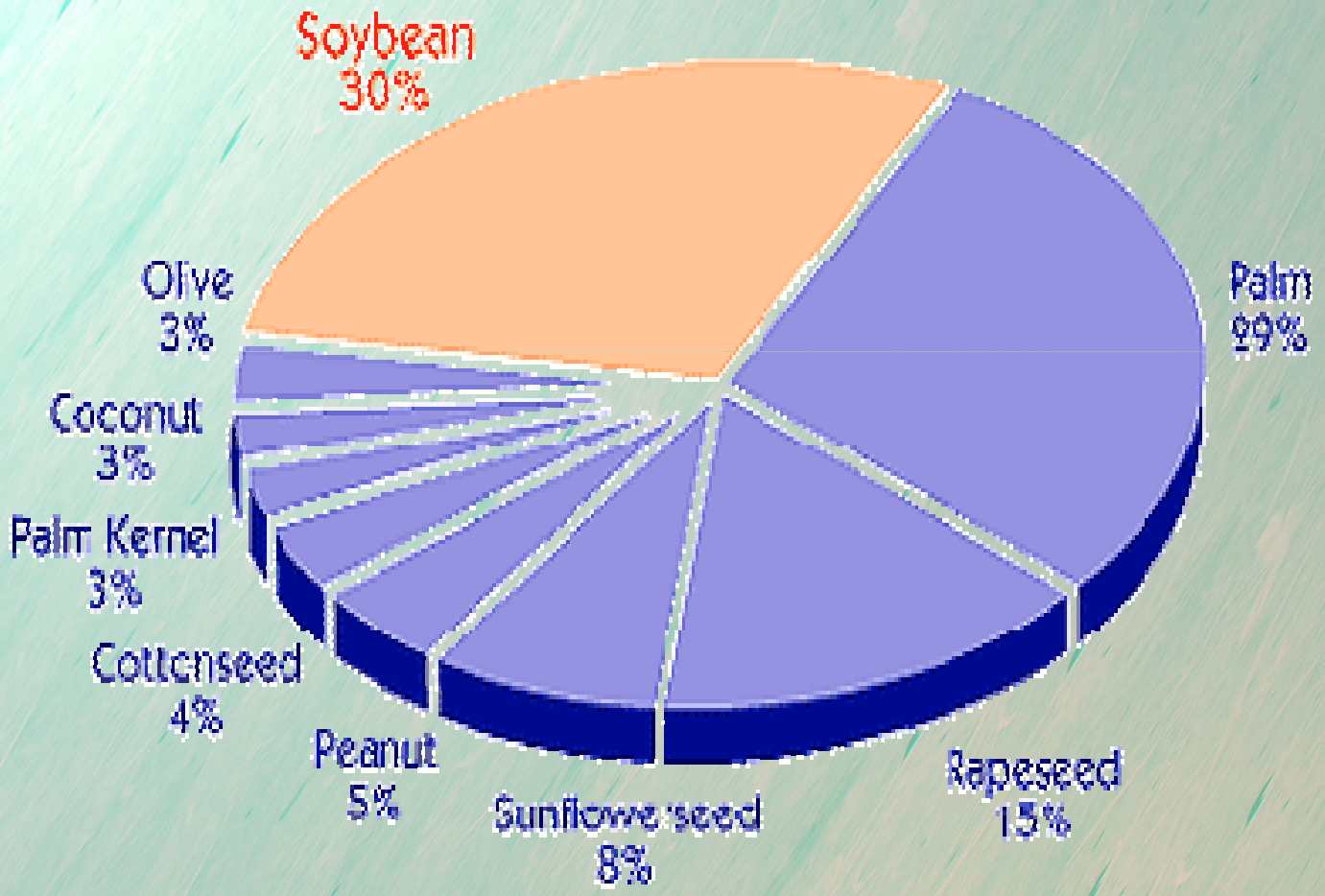
Introduction of Oilseeds

- Oil seeds are oil bearing crops, such as soybean, sunflower, cotton seed, palm and rape seed oil.
- Oil seeds are widely used in food, feed and industrial applications.

- The oil content of different oilseeds are as follows.
- Soybean 18-20%
- Cotton seed 15-18%
- Palm oil 45-48%
- Sunflower 42-46%
- Canola oil 42-44%

World Vegetable Oil Consumption

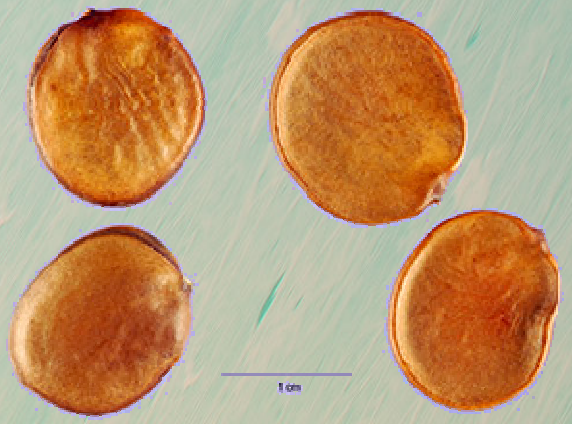


Aims of work

- **To evaluate Physiochemical Characteristics of *Bauhinia purpurea* seed oil for various commercial exploration for oil industry and improvement in crops economics.**

- **To investigate the quality of Seed, Oil and meal for their use in poultry and animal feed industry**

Bauhinia Pupurea



General Information of Bauhinia Species

- **Scientific name:** Bauhinia variegata, B. candida, B. purpurea
- **Family: Febaceae (Caesalpinioideae)**
- **Flower color:** white, purple, Pink
- **Fruit shape:** elongated; pod, 12 inches or more in length, fruit covering dry, hard and brown in color, seeds are rounded flat and shiny brown
- **Light requirement:** tree grows in part shade/part sun in full sun
- **Soil tolerances:** slightly alkaline; acidic; well-drained
- **Watering Needs:** Moderate water
- **Propagation:** Easy by seeds and cutting

Occurrence

- **About 600 species of bauhinia grow in the tropical regions all over the world (Southeastern Asia, India, China, Africa, Florida and Hawaii). Widely planted in the tropics and warm regions of the world.**

- **In Pakistan Bauhinia is naturally found in the sub-Himalayan tract. Northern areas, In all over the Punjab, in Sindh (Tandojam, Hyderabad, Karachi, Jamshoro) and outer hills and valleys of river Indus in Pakistan.**

Importance of Bauhinia plant

- **It is used in traditional medicine for the treatment of various ailments, Saveral Auyurvedic Medicine Formulation.**
- **The stem bark of the plant is used in the treatment of headache, fever, skin diseases, tumors, diseases of the blood, dysentery and diarrhea.**
- **Pharmacological studies of the plant revealed that the ethanol extract of leaves have analgesic, antipyretic, anti inflammatory and antimicrobial activity.**

Experimental Work

Sampling

- **Seeds of Bauhinia purpure were collected from Hyderabad, Tandojam, Karachi and Abbotabad University Campus during year 2007-2008.**

- **Seeds were packed and sealed in polythene bags and stored in dark until subjected to analysis.**

Analysis of Extracted Oil

- Color of oil
- Refractive index
- Free fatty acids, Peroxide value, Saponification value, Iodine value, Unsaponifiale matter, Refrective index and color of oil.
- Fatty acid composition by capillary Gas Chromatography
- Tocopherol content by high performance liquid chromatography, Sterol by GC/MS

Extraction Method of oil

The oil was extracted from the ground material by extraction with n-hexane (b.p 50–60 ^{0}C) in a Soxhlet apparatus for 6 h following the (AOCS Aa 4–38). The oil was weighed to calculate the percentage of extracted yield. Oil and meals were stored at 5^{0}C under nitrogen atmosphere for further analyses.

Determination of physical parameters

Several analytical methods were used to evaluate oil composition, quality and oxidative stability. Standard AOCS and IUPAC official methods were used to determined the following Parameters.

Color Index. A Lovibond Tintometer Model-E Salisbury, England was used for the reading of color of oil.

Refractive index. measured by refractometer (IUPAC 2.102).

Acid value. Acid value is a measure the amount of free acids present in a given amount of fat. The number of milligrams of potassium hydroxide required to neutralize the free acid in 1 gram of the sample (AOCS Cd 3d-63).

Iodine value

Iodine value of the oil is the number of grams of iodine absorbed by 100grams of the oil determined by using Wijs method (IUPAC 2.205).

Saponification value

Hydrolysis of ester under alkaline condition. It is the number of KOH required to saponify 1 gram of oil. Saponification is the hydrolysis of ester under alkaline condition (AOCS Cd 3-25).

Peroxide value

Peroxides (R-OOH) are the primary products formed in the initial stages of oxidation. It is determined by measuring the amount of iodine liberated from KI by the oxidative action of peroxides present in the oil (AOCS Cd 8-5).

Unsaponifiable matter

Those substances dissolved in fatty acids which cannot be saponified by caustic treatment, but which are soluble in the normal fat solvents. Included higher aliphatic alcohols, sterols, pigments, and hydrocarbons (IUPAC 2.401) .

Fatty Acids Analysis by GC

1. **Prepare methyl ester prepared by standard IUPAC method (2.301).**

2. **Fatty acid methyl ester were analyzed by Gas Chromatography, using Perkin- Elmer Model 8700 Gas Chromatograph equipped with capillary column and Flame ionization detector. The following conditions were used**

- **Mobile phase:** **Nitrogen 3.5ml/min**
- **Temperature:** **130/4/220/2min**
- **Column:** **Supelco SP-2340**
- **Length** **60 m x 0.25 mm**
- **Split ratio** **1:18**

Tocopherol Analysis by HPLC

- **Tocopherols (α, β,γ and δ) analysis was carried out by High performance liquid chromatography (HPLC) following the method of Anna Gliszezynska-swiglo and Sikorska (2004).**

- **2g of oil was dissolved in 2-propanol and diluted in a 10ml volumetric flask wrapped in foil to inhibit oxidation.**

- **Quantification was done by external calibration method. The tocopherol were identified by comparing their retention times with those of corresponding standard.**

- **A 10µl portion was injected onto Hitachi model 6200 HPLC unit equipped with Licrosorb octadecylsilane (ODS) column. Hitachi F-1050 florescence detector was set at emission wavelength of 325nm with an excitation at 295 nm. Mobile phase: 50% acetonitrile and methanol with the flow rate of 1ml/min.**

Determination of sterol by GC/MS

- **Sterol analysis.** Separation of sterols (ST) was performed after saponification of the oil sample without derivatisation.

GC-MS parameters for sterol analysis

- **Agilent Technology GC- model 6890 N fitted with a polar capillary column HP-5MS (5% phenyl methylsiloxane) 30m x 0.25mm i.d x 0.25 *micron* film thickness and a MS detector.**
- **Helium was used as a carrier gas at a flow rate of 1.2 mL/min.**

Initial oven temperature	**150 °C**
Ramp rate 1	**15 °C/min**
Second temperature	**250 °C**
Ramp rate 2	**10 °C/min**
Final temperature	**310 °C (10 min stay)**
Injector temperature	**290 °C**
Detector temperature	**330 °C**

Analysis of oil seed and Seed residue

- **The oil seed residue (meal) obtained after the extraction of oil from the seeds were analyzed by official methods for fiber, ash and protein contents.**
- **Ash content of seed and meal was determined according to the ISO method**
- **Fiber content of seed and meal was determined by the ISO method (749).**
- **Protein content was determined by ISO method (1983).**

RESULT AND DISCUSSION

Physiochemical Characteristics of Bauhinia purpurea seed oil

Characteristics	Mean ± SD
Refractive index (40 ^{0}C)	1.4645 ± 0.001
Color (red unit)	2.52 ± 0.07
Color (yellow unit)	50.5 ± 0.16
Iodine value (g of I_2/100 g oil)	99.19 ± 0.79
Saponification number (mg of KOH/g oil)	189.02 ± 1.39
Acid value (as oleic acid g/100 g)	0.16 ± 0.02
Unsaponifiable matter (g/100 g)	1.41 ± 0.34
Peroxide value(meq/kg oil)	0.02 ± 0.01

Fatty acid composition of Bauhinia purpurea seed oil

Fatty acid	Mean ± SD (g/100 g)
Myristoleic acid (C14:1)	0.18±0.02
Palmitic acid (C16:0)	17.47±0.98
Palmitoleic acid (C16:1)	0.16±0.01
Stearic acid (C18:0)	11.40±0.64
Oleic acid (C18:1)	11.84±0.97
Linoleic acid (C18:2)	55.34±0.72
Arachidic acid (C20:0)	0.92±0.01
Alpha linolenic (C18:3)	0.47±0.02
Gama linolenic (C18:3)	0.36±0.02
Behenic acid (C22:0)	0.34±0.02
Eicosadienoic acid (C20:2)	0.36±0.01
Eicosapentaenoic (C20:5)	0.38±0.02
Lignoceric acid (C24:0)	0.14±0.02
Nervonic acid (C24:1)	0.51±0.04
Σ SFA	30.27
Σ MUFA	12.79
Σ PUFA	56.94

Tocopherol profile *Bauhinia purpurea* seed oil

Tocopherols (mg/Kg)	**Mean±SD (Range)**
α-tocopherol	996.05 ± 64.78
(β+γ)-tocopherol	644.89 ± 39.79
δ-tocopherol	17.33 ± 1.03

Sterol profile (% of total sterol) of *Bauhinia purpurea* seed oil

Sterol composition (%)	**Mean ± SD (Range)**
Compesterol	10.71 ± 0.75
Stigmasterol	18.83 ± 0.74
β-sitosterol	62.24 ± 1.13
Δ5-avenasterol	4.05 ± 0.47
Δ7- avenasterol	1.62 ± 0.34
Δ7- stigmasterol	2.53 ± 0.58

Composition of Bauhinia purpurea Seed and Seed Meal

Constituents	Seed (g/ 100 g)	Meal (g/100 g)
Moisture	6.45 ± 0.21	5.69 ± 0.79
Oil	18.16 ± 1.11	Nil
Protein	35.69 ± 1.74	43.72 ± 1.01
Fiber	6.17 ± 0.15	8.04 ± 0.43
Ash	3.13 ± 0.14	5.78 ± 0.72
Carbohydrates	32.40 ± 0.75	33.93 ± 0.88

Conclusions

- **The results of present analysis indicate that Bauhinia seeds have great potential for the nutraceutical application and economical utility as a new source of edible oils.**

- **Bauhinia seed oil had higher amount of Palmitic acid content than sun flower oil and is suitable for margarine production.**

- **The high amount of PUFA make the Bauhinia seed oil special component for nutritional application . Further the low level of free fatty acid is an indicator of good quality of bauhinia oil.**

- **High tocopherol contents detected in bauhinia seeds oil contribute to protect oil against oxidation during storage, so blending of Bauhinia oil with other consumable oil can enhance the stability and shelf life of these oils.**

Conclusions

- **The bauhinia seed oil could be used directly as a natural antioxidant in functional food products to increase the shelf life.**

- **Furthermore bauhinia purpurea seed meal quality analysis indicate that it can be used as a good source of protein, which could be used in poultry feed as replacement of soybean and sunflower meal for the local poultry industry.**


Thanks
A view of Mushk purri top Nathia Gali , NWFP , Pakistan