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Chemical Composition and Characterization of Hemp (*Cannabis sativa*) Seed oil and essential fatty acids by HPLC Method

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ABSTRACT

The hemp (cannabis sativa) seed oil is extracted from seed. It was examined for physical and chemical properties. It was analysed by Percentage ash, Refractive index, Specific gravity, Acid value, Iodine value, Saponification number, Unsaponifible mater content and Peroxide number. Hemp seed oil contains fatty acids that maintain healthy blood vessels, nerves and tissues. The high performance liquid chromatography (HPLC) is a suitable analytical method for seed oil of hemp. It was detected by HPLC chromatogram showing retention time.

Keywords: Hemp (*cannabis sativa*) seed oil, Chemical composition, Characterisation, Instrumental analytical techniques, HPLC.

INTRODUCTION

Hemp (cannabis sativa) is an angiosperm belonging to the cannabaceae family and cannabis genus. Hemp plant itself is easy to grow in temperate climates and requires good soil, fertilizer and water but no pesticides nor herbicides.A hemp crop is usually harvested in 120 days after reaching a height of 10-15 feet. The whole seed contains roughly 25 % protein, 30 % carbohydrates, 15% insoluble fibre, carotene, phosphrous, potassium, magnesium, sulphur, calcium, iron and zinc as well as vitamin E,C,B₁,B₂, B₃, B₆. Hemp seed is one of the best source of essential fatty acids with perfect 3:1 ratio of omega-3-linolenic acid and omega-6-linoleic acid, good for strengthening the immune system. It is also a good source of gamma linoleic acid. The high content of omega-6 & omega-3 fatty acids and the relatively high phytosterol content of hemp make them beneficial to cardiovascular health polyunsaturated for saturated fats can reduce the risk of sudden cardiac arrest and fatal cardiac arrhythmia as well as reducing blood cholesterol levels and decreasing the cellular proliferation associated with atheroscierosis. It is also a good source of gamma linoleic acid (GLA). The GLA and vitamin D of hemp beneficial in preventing and treating Osteoporosis. Essential fatty acids has been found capable of reversing scaly skin disorder, rheumatism, inflammation, diabetes, excessive epidermal water loss, itch and poor wound beneficial for atopic eczema and psoriasis. Traditional hemp formulas were applied topically to treat abscesses, boils, pimples and swellings. The seed folk remedy for tumour and cancerous ulcers. The seed oil is also used in paint, shampoos and soap.Oil is also used in cosmetics and body care product is antimicrobial, anti-inflammatory and antiageing balances, skin pH and moisture levels.

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MATERIALS AND METHODS

The dried clean seeds of cannabis sativa are extracted with n-Hexane in soxlet extractor. Solvent is removed by rotavapour and the oil is purified. The purified oil is tested by proximate analysis. Physical and chemical test methods are used to access the quality of seed oil.





Preparation of Extract

The seed powder prepared from shade dried seed of cannabis sativa was extracted with n-hexane (1:4 w/v) by continous extraction in a soxhlet extractor for 12 hrs. The solvent (n-hexane) was separated at 40° C with the help of rotavapour. The oil was purified and stored in disiccator and then stored in sealed & dark bottles. Proximate chemical composition, physical and chemical analysis of the extracted oil was done. The physical state, colour,taste and odour were evaluated by means of sensory organs.

Proximate Chemical Composition

The Proximate Chemical Composition of dried seeds of cannabis sativa as shown in Table 1. Recommended methods of Association of Official Analytical Chemists (AOAC, 2005) were used to determine the chemical composition of the cannabis sativa. Seeds including the moisture content, crude oil, protein, ash and fibre content and also content of carbohydrates.

Characterisation of seed oil

Cannabis sativa seed oil was a liquid at room temperature with bold yellow colour, pleasant nutty odour and bland taste congealing point were determined using ice and salt mixture. Refractive index was measured with an Abbe's refractometer equipped with a thermostated circulator. Specific gravity was determined at 20 ⁰c using 25 ml capacity specific gravity bottles. The chemical properties, acidvalue, iodine value, saponification number, unsaponifible matter content and peroxide number were determined by standard methods of AOAC (1990).

Table 1,2 & 3 shows that the proximate analysis, Physical Properties & of hemp seed, Physical & Chemical properties of hemp seed oil .

Sr.No.	Parameters	Content (%)
1)	Moisture	03.07 %
2)	Oil	32.21 %
3)	Protein	23.90 %
4)	Ash	04.32 %
5)	Fibre	17.30 %
6)	Carbohydrate	28.50 %

Table 1. Proximate Analysis of Hemp (cannabis sativa) Seed

Sr. No.	Parameters	Value
1)	Physical state	Liquid
2)	Colour	Bold yellow
3)	Odour	Pleasant nutty
4)	Taste	Bland
5)	Congealing point	15-72 ° C
6)	Refractive index (40°C)	1.4570
7)	Specific gravity (20°C)	0.8927

Table 2. Physical Properties of Hemp (cannabis sativa) Seed Oil

Table 3. Chemical Properties of Hemp (cannabis sativa) Seed Oil

Sr. No.	Parameters	Value
1)	Acid value (mg KOH / g of oil)	2.15
2)	Iodine value (g / 100 g of oil)	163.5
3)	Saponification number (mg KOH / g of oil)	190.2
4)	Unsaponifible matter (% of oil)	0.26
5)	Peroxide number (Meg / Kg)	7.2



1 PDA Multi 1 / 210nm - 400nm 4nm

Hemp (cannabis sativa) seed oil contains oil about 32.21 %. The content of moisture, protein, ash, fibre and carbohydrates were 03.07 %, 23.90 %, 04.32 %, 17.30 %, 28.50 % respectively. The whole seed moisture content were quite low. Hemp seeds are a good source of oil, protein & carbohydrates. The high oil content makes the seed a potential source of commercial vegetable oil. It is used for many purposes. The physical & chemical properties of hemp (cannabis sativa) seed oil were determined. It was found that the oil extracted from dried hemp seeds was liquid at room temperature bold yellow colour having bland taste & pleasant nutty odour. It has congealing point 15-72 °C. The refractive index at 40 °C and specific gravity at 20 °C were 1.4570 & 0.8927 respectively. The acid value (mg KOH / g of oil), iodine value (g / 100 g of oil), saponification number (mg KOH / g of oil), unsaponifible matter content (% of oil) and peroxide number (Meg / Kg) of oil were 2.15, 163.5, 190.2, 0.26 & 7.2 respectively. Acid value of oil is an indicator for edibility of oil & suitability for industrial use. Iodine value of hemp seed oil indicate a high composition of poly unsaturated fatty acid is an assest in nutrition as high content of saturated fatty acids is implicated in cardiovascular disease. It is also used in consmetics and body care products.hemp seed oil contains fatty acids that help to maintain healthy blood vessels & nerves. High performance liquid chromatography (HPLC) is a suitable analytical method for determing seed oil of cucurbita maxita was detected by HPLC chromatogram showing fatty acids with 35.74 % at 4.428, 3.79 % at 3.286, 5.08 % at 3.451 , 13.25 % at 3.832 , 2.40 % at 4.076 , 49.05 % at 4.490 , 3.59 % at 4.870 , 4.74 % at 5.113 , 5.32 % at 5.505 , 1.54 % at 5.767, 7.21 % at 7.989 and 3.99 % at 8.633 respectively. HPLC study revealed presence of number of constituents were detected and further investigations are in progress in the laboratory.

CONCLUSION

Hemp (cucurbita maxita) seed oil, which has medicinal as well as industrial applications.

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