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### Original Research Article

## Extraction and Determination of Active Composite in the Cultivated *Daucus Carota* (Carrot) Seed Oil

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### ABSTRACT

Chemical composition and physicochemical analysis were established in carrot (*Daucus carota*) seeds from Zaria, Kaduna to investigate their potential uses. The oil extraction from Carrot seed was achieved through Soxhlet extraction method. Some of the physicochemical analysis was iodine value (67.007 /100 gram of KOH), saponification value (78.94 KOH/ gram), acid value (9.54 mg), relative density (0.9797), and free fatty acid (4.797%). The analysis of the seed oil is performed by FTIR and has allowed us to identify 3 functional groups in the oil of *daucus carota* of cultivated seeds in Zaria. These chemical compositions are carbonyl group for a cyclic 5-membered ring (C=O), methylene group (C-H), and aromatic alkenes (C=C). From the analysis of the chemical composition, it can be concluded that the active composite of cultivated carrot seed oil are C=O, C=C, and C-H.

**Keywords:** Extraction, seed oil, FTIR, Physicochemical properties, Iodine value

## Introduction

About 80% of the population in developing countries depends on medicinal plants which plays a vital role in health care [1]. About 25% of medicines prescribed today are products produced directly or indirectly from these plants. Newman et al, (2016)[2] earned an assessment that 49% of the new 983 chemical entities approved as drug from 1981 to 2014 were natural product derivatives.

The demand for natural product has been in great increase being impact of medicinal plant to health and agricultural sector [3]. The rate at which people accept natural product is increasing in detriment to conventional drugs is due to the long usage history and prominent side effects of conventional drugs.

Recently, several researchers have been attracted to investigation on aromatic plant particularly essential oils [1]. According to the International standard organization on essential oils [4-6] and European pharmacopoeia council of Europe [7, 8], an essential oil is defined as the product obtained from plant raw material by hydro distillation, steam distillation or dry distillation by a suitable mechanical process. The chemical compounds obtained have been used for several purposes such as insecticides, anti-microbial, anti-parasites, fungicidal, bactericidal based on the beliefs of the users. Currently, essential oils play critical values in pharmaceutical, food, cosmetics and perfumes, sanitary, food, agronomic industries [1, 9, 10].

The species *Daucus carota* L. commonly known as carrot is particularly known worldwide due to general use in both food and medicinal purposes [11]. Carrot contains mostly the tap root which is mainly eaten and the green leafy part which is eaten as well but it is not common and is always crispy when fresh. The taproot is regarded as the consumable part of carrot which is the nutritious and healthy food supplement due to its high vitamins and fiber content [12]. Different biological activities are embedded in the carrot seed oil [1, 9, 10, 13-16]. The carrot seed oil has been described for hypotensive, anti-microbial, antihelminthic diuretic properties [17].

Carrot seed oil is an essential oil that has been likened to cheaper carrot oil obtained from maceration of carrot with carrier oil [12]. Carrot is not wildly grown in our region, it is more cultivated. Carrot seed oil is being imported into our geographical area, to the best of my knowledge the extraction of carrot seed oil and determination of the active composite has not been reported. Few researches have been carried out on carrot seed oil which was obtained from processed carrot seed which has gone through preservative in other to maintain its half life

before being used. Several years, research were been carried out on carrot seed oil obtained from local farmers or obtained from wild source.

## Experimental

### Sample Collection

Carrot seeds were obtained from a seed vendor at sabon gari local market, Zaria Kaduna state. The seed of the plant was dried, pulverized thereafter, extraction proceeded.

### Extraction Procedure

295 gram of the crushed carrot seed was weighed and placed inside a thimble made from thick filter paper, which was loaded into the main chamber of the soxhlet extractor. 490 cm<sup>3</sup> of n-hexane was poured into a distillation flask and the soxhlet extractor was placed onto this flask which was equipped with a condenser. The solvent was heated to reflux at constant temperature of 68 °C and the vapor travelled up a distillation arm into the chamber housing the thimble of solid where the vapor is condensed and the warm solvent flow under gravity and percolate through the bed of the crushed carrot seed to extract the oil. When the solvent chamber is almost full, the chamber is automatically emptied by a siphon side arm, with the solvent running back into the distillation flask. The cycle was repeated for 8 hours. After the extraction the extract was The following sections give the description of the different methods of preparation of reagents used in the characterization of the oils.

### Preparation of 0.1 M of Sodium thiosulphate Solution (Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub>)

$$\begin{aligned} \text{Molecular weight of Na}_2\text{S}_2\text{O}_3 &= 158 \text{ g/mol} \\ \text{Number of moles} &= \frac{\text{reacting mass}}{\text{molar mass}} = 0.1 = \frac{Rm}{158 \text{ g/mol}} \end{aligned}$$

$$Rm = 0.1 \text{ mol} \times 158 \text{ g/mol} = 15.8 \text{ grams}$$

Therefore, for conveniences 3.95 grams of Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub> crystals was accurately weighed and was quantitatively transferred into a clean 250 cm<sup>3</sup> volumetric flask. To this, a small amount of

distilled water (30 cm<sup>3</sup>) was added to the content shaken vigorously, so as to dissolve sodium thiosulphate. Distilled water was added to make up the volume to the marked point and the solution was used in the evaluation of iodine value.

### Preparation of 0.1 M of Potassium hydroxide solution (KOH)

Molecular weight of KOH = 58 g/mol

$$\text{Number of moles} = \frac{\text{reacting mass}}{\text{molar mass}} = 0.1 \text{ mol} = \frac{Rm}{58 \text{ g/mol}}$$

$$Rm = 0.1 \text{ mol} \times 58 \text{ g/mol} = 5.8 \text{ grams}$$

Since, minute amount of the solution is required, 1.45 grams was accurately weighed and quantitatively transferred into a clean 250 cm<sup>3</sup> volumetric flask containing 30 cm<sup>3</sup> of distilled water, and the solution was shaken vigorously until a clear solution was observed. Distilled water was further added to the marked point and the solution was used in the evaluation of acid value.

### Preparation of 0.5 M of Potassium Hydroxide (KOH)

Molecular weight of KOH = 58 g/mol

$$\text{Number of moles} = \frac{\text{reacting mass}}{\text{molar mass}} = 0.5 \text{ mol} = \frac{Rm}{58 \text{ g/mol}}$$

$$Rm = 0.5 \text{ mol} \times 58 \text{ g/mol} = 29 \text{ grams}$$

Since, minute amount of the solution is required, 7.25 grams was accurately weighed and quantitatively transferred into a clean 250 cm<sup>3</sup> volumetric flask containing 30 cm<sup>3</sup> of distilled water, and the solution was shaken vigorously until a clear solution was observed. Distilled water was further added to the marked point and the solution was used in the evaluation of acid value.

### Preparation of 0.5 M of Hydrochloric acid (HCl)

Molecular weight of HCl = 36.5 g/mol

$$\text{Number of moles} = \frac{\text{reacting mass}}{\text{molar mass}} = 0.5 \text{ mol} = \frac{Rm}{36.5 \text{ g/mol}}$$

$$Rm = 0.5 \text{ mol} \times 36.5 \text{ g/mol} = 18.25 \text{ grams}$$

Therefore for conveniences, minute amount of the crystals was required; 4.56 grams was accurately weighed and transferred into a clean 250 cm<sup>3</sup> volumetric flask containing 30 cm<sup>3</sup> distilled water.

## Characterization of the oil

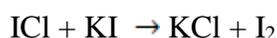
The following parameters normally used in evaluating the quality of the oil during storage or in use were determined. These parameters include iodine value (IV), acid value (AV), saponification value (SV), free fatty acid, relative density (RD).

### Iodine value (IV)

Using a weighing balance, 1 gram of the oil sample was weighed and transferred into 50 cm<sup>3</sup> of chloroform and allowed to dissolve. Using a pipette, 10 cm<sup>3</sup> of the oil sample in chloroform was pipette out to an iodination flask was labeled as “A”. 20 cm<sup>3</sup> of iodine monochloride reagent was added into the flask and mixed thoroughly. Then the flask was allowed to stand for 30 minutes in an incubated dark place. A blank was set up labeled as “B”, in an iodinated flask; 10 cm<sup>3</sup> of chloroform was transferred to the empty flask, and 20 cm<sup>3</sup> of iodine monochloride reagent was also added and mixed thoroughly, then incubated also in the dark for 30 minutes. After 30 minutes, flask A was taken out and 10 cm<sup>3</sup> potassium iodide solutions was added to the flask, 50 cm<sup>3</sup> of distilled water was added and used to rinse the stopper and the flask. Then flask A was titrated against standardized sodium thiosulphate solution until a pale straw color was observed. Then about 1 cm<sup>3</sup> of starch indicator was added to the content in the flask and a purple color was observed. Then titration was continued until the content in the flask became colorless, the endpoint reading was recorded. This procedure was repeated for the blank which is flask B. the endpoint was recorded.

The volume of 0.1 M of sodium thiosulphate by the blank (B) – volume used in the determination of the sample (A) gives the amount of sodium thiosulphate equivalent to iodine absorbed.

The equation of the reaction is given below



Using Hanus method, the iodine number was calculated

$$IV = \frac{(B - A) \times M \times 12.91}{\text{weight of the oil}}$$

### Saponification value

About 1 g of oil is transferred in to an R. B. flask and dissolved in 25 cm<sup>3</sup> of distilled ethyl alcohol. 25 cm<sup>3</sup> of 0.5 N alcoholic KOH is added into it and mixed well. Put it in the boiling water bath for 30 minutes. After 30 minutes allow cool to room temperature.

A blank experiment is simultaneously conducted in the same way without taking oil. 2 drops of phenolphthalein is added and titrated against a standard solution of 0.5 M hydrochloric acid until the pink color disappeared. The difference between the test (A) and blank (B) gives the volume of 0.5 M HCl equivalent of KOH used in saponifying W g (here 1 g) of the oil. 56 is the equivalent mass of KOH.

$$\text{Saponification value} = \frac{(B - A) \times 0.5 \times 56}{W}$$

### Acid value

About 500 mg of the oil sample is taken in a conical flask and dissolved in 50 cm<sup>3</sup> of distilled alcohol by gentle warming. It is then titrated against 0.1 M KOH using phenolphthalein as indicator until a slight pink color is appeared. For this titre value, the acid value is calculated by using the following equation

$$AN = (V_{eq} - b_{eq})N \frac{56.1 \text{ gmol}^{-1}}{W_{oil}}$$

### Percentage of free fatty acid

To see the relationship between AV and FFA%, we can solve both equations for common values or:

$$AV/56.1 = (v - b) \times N / w \quad (1)$$

$$FFA\%/28.2 = (v - b) \times N / w \quad (2)$$

(The percentage of free fatty acid is usually calculated in terms of oleic acid, 1000 g of sample contains 282 g of oleic acid)

Now by combing the equations (1) and (2), we can mention

$$AV/56.1 = FFA\%/28.2$$

$$AV = 1.99FFA\%$$

$$\%FFA = AV/1.99 = AV \times 0.503$$

## Infrared Analysis

The infrared spectral analysis of carrot seed oil was recorded using fourier transformed infrared genesis model angilent. The results are shown below in table 4.

## Results and discussion

The following results were obtained from the experiments conducted on carrot seed oil;

### Results Obtained (Percentage of oil yielded)

The result for the percentage oil yield was as follows:

Sample weight= 295 grams

Empty beaker weight = 48.504 grams

Beaker + oil weight = 85. 06 grams

$$\% \text{yield} = \frac{\text{oil weight} - \text{empty beaker}}{\text{sample weight}} * 100$$

$$\% \text{yield} = \frac{85.06 - 48.50}{295} * 100$$

$$= 36.556 * 100/295$$

$$= 0.1239 * 100$$

$$= 12.39\%$$

The percentage yield of carrot seed oil is 12.39%.

The percentage yield of carrot seed oil (12.39%) is lower than the carrot seed oil (23.4%) which was investigated from Jos as reported from Abdulrasheed et al. [12]. These differences can probably be due to different seeds, soil characterization and environmental factor.

## Relative Density

The result of the relative density of oil is as follows:

Mass of the oil= (mass of oil + density bottle + stopper) – (mass of density bottle + stopper)

$$\text{Mass of oil} = 36.44 - 32.53 = 3.91$$

$$\text{Density of the oil} = \frac{\text{mass of oil}}{\text{volume of oil}}$$

$$\text{Density of oil} = 3.91/4.0 = 0.9775$$

Mass of water = (mass of water + density bottle + stopper) – (mass of density bottle + stopper)

$$\text{Mass of water} = 36.56 - 32.53$$

$$= 4.03$$

Density of water = mass of water/ volume of water

$$= 4.03/4.0$$

$$= 1.01$$

Relative density = density of oil/ density of water

$$= 0.9775 / 1.01$$

$$= 0.9678$$

The relative density of carrot seed oil 0.97 approximately. Relative density usually explains the buoyancy of a substance in a fluid or gas. Therefore, if a substance's relative density is less than one then it is less dense than the reference; if it is greater than one then it is denser than the reference. In the case of carrot seed oil (0.97), this indicates that the carrot seed oil sample was not adulterated with water or alcohol.

## Acid Value

The result obtained for the acid value analysis is as follows:

First titre value (cm<sup>3</sup>)

**A**

Final volume	0.85
Initial volume	0.00
Volume used	0.85

Second titre value (cm<sup>3</sup>)

**B**

Final volume	0.85
Initial volume	0.00
Volume used	0.85

$$\text{Acid value A} = \frac{56.1 \times 0.85}{5} = 9.537$$

$$\text{Acid value B} = \frac{56.1 \times 0.85}{5} = 9.537$$

$$\text{Average acid value} = \frac{9.537 + 9.537}{2} = 9.54$$

Acid value = 9.54 mg KOH / gram

The acid value of 9.54 mg KOH / g of oil of the carrot seed oil indicate a low level of long chain carboxylic acids in the carrot seed oil sample.

### Saponification Value

Sample containing oil(S)		blank (cm <sup>3</sup> )	
A		B	
Final volume	20.0	Final volume	22.81
Initial volume	0.00	Initial volume	0.00
Volume used	20.00	Volume used	22.81

$$\text{Saponification Value} = \frac{56.1 \times \text{Molarity of KOH} \times (\text{BL} - \text{S})}{\text{Weight of oil}}$$

$$\text{Saponification Value} = \frac{56.1 \times 0.5 \times (22.81 - 20.00)}{1}$$

Saponification Value= 78.82 mg KOH / g

The saponification value SV is used to determine the saponification number of fat or oil which is an index of the average molecular weight of triglyceride in the sample. Saponification value is important in soap production. The higher the SV, the longer the average fatty acid chain length. From the result obtained, SV 78.82 mg KOH / g indicates that it is suitable for soap production.

### Percentage of Free Fatty Acid

The result for percentage free fatty acid is as follows:

$$\begin{aligned} \% \text{ Free Fatty acid} &= \text{Acid value} \times 0.503 \\ &= 9.537 \times 0.503 \end{aligned}$$

$$= 4.797\%$$

Fats and oil are known to be triglycerides; therefore the free fatty acid should be very low in highly graded lipid sample. The free fatty acid percent was 4.797%. This indicates a low value therefore it has a good storage quality.

### Iodine value

Sample with oil (S)

First titre volume (cm<sup>3</sup>)

(A)

Final volume	45.70
Initial volume	0.00
Volume used	45.70

Second titre value (cm<sup>3</sup>)

Sample with oil (cm<sup>3</sup>)

(B)

Final volume	45.80
Initial volume	0.00
Volume used	45.80

Blank (Bl)

First titre volume (cm<sup>3</sup>)

Final volume	97.70
Initial volume	0.00
Volume used	97.70

Second titre value (cm<sup>3</sup>)

Blank (Bl)

Final volume	97.60
Initial volume	0.00
Volume used	97.60

$$IV = \frac{(Bl - S) \times M \times 12.91}{\text{weight of the oil}}$$

For A,

$$\text{Iodine Value} = \frac{(97.7 - 45.7) * 0.1 * 12.91}{1}$$

$$\text{Iodine Value} = 67.132$$

For B,



**Table 1.** Infrared spectroscopic data of the cultivated carrot seed oil

Compound	Functional group	Signal	Strength
Cultivated carrot seed oil	C-H (Alkane)	2922.2	Strong
	= C – H <sub>3</sub>	2855.1	Medium
	= C – H <sub>3</sub>	1379.1	Weak
	C = C (aromatic)	1461.1	Medium
	C = C – H	1610.1	Weak
	C=O (cyclic 5-membered) Aldehyde (CHO)	1744.4	Strong

The infrared spectra analysis of carrot seed oil as interpreted and reported in table 4.2 above shows that a strong signal at 2922.2, a medium signal at 2855.1 and a weak signal at 1379.1 for methylene. For an aromatic alkene, it was shown as a weak signal at 1610.1 and medium signal at 1461.1. For a carboxylic group a strong signal was shown at 1744.4.

## Conclusion

Iodine value of the carrot seed oil was calculated to be 67.0025 which fall within the range (67-82) of natural waxes that simulates fatty oils. The calculated iodine value of carrot seed oil classifies it as a natural wax which behaves similarly to solvent. In its liquid condition, carrot seed oil leaves a grease spot on paper making it a useful lubricant.

The percentage oil yield of the carrot seed obtained was 12.39% which is within the range of non-drying oil (12-15) such as Olive Kernel which are useful oils in soap making and edible.

The calculated relative density of carrot seed oil shows identical result to the relative density of essential oils which ranges from 0.850 - 1.142. It is therefore concluded that carrot seed oil is also an essential oil which have smaller relative density than water.

The acid value 9.54 mg KOH/g and free fatty acid 4.797% obtained from the carrot seed oil is low. The higher the acid value, the lower storage quality and vice versa. This shows that carrot seed oil have an excellent storage quality.

From the saponification value of carrot seed oil obtained 79.84 mg KOH/g, which means it has a higher potential for soap production. This indicates that the oil could be used in soap making since its saponification value is high.

The IR spectra shows results classifying carrot seed oil as essential oil due to the presence of various functional groups which are important in identifying essential oil. Such functional groups present are ketone, aldehyde and hydrocarbons.

Conclusively, the results in this analysis indicated good quality of the carrot seed oil. Physicochemical analysis presented, showed that saponification value, acid value, free fatty acid, relative density, and iodine value that fell within the range of those acceptable as having good potential for lubricant, soap production and a good storage quality. The infrared interpretation, iodine value and relative density showed that the carrot seed oil is also an essential oil.

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