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Characterization and Utilization of castor bean seed oil extract for production of medicated soap.

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ABSTRACT: *The research work is to investigate the potential utilization of castor bean seed oil extract in the production of medicated soap. The oil was extracted via soxhlet extractor using hexane as solvent. The characterization analysis reveals the acid value and saponification value of the oil which were between the ranges of values specified by ASTM. The soap produced gave a pH of (8.9), foam height (16cm), alcohol insoluble (3.45%), moisture content (4.2%) and free acidity of (0.10). The antibacterial activity of soap produced from castor oil on bacteria isolate (Staphylococcus Aureus) was promising with an inhibition zone of 15.5mm but at dilutions of 10^{-1}, 10^{-2} and 10^{-3} , were found to be 11.5mm, 9.5mm and 6mm respectively. This shows that as concentration decreases, the sensitivity of the soap to the bacteria isolate also decreases. The sensitivity of the medicated soap to the bacteria isolate is as a result of the presence of ricinoleic acid present in large proportion in the fatty acid composition of castor oil. It can be concluded that a highly effective soap can be produced from castor bean seed oil extract.*

Keywords *- castor bean seed, characterization, medicated soap, solvent extraction, and staphylococcus aureus.*

I. INTRODUCTION

Castor plant, *Ricinus communis*, is a specie of flowering plant in the spurge family, Euphorbiaceae. Its seed is the castor bean which, despite its name, is not a true bean. Castor plant is indigenous to the southeastern Mediterranean Basin, Eastern Africa, and India, but is widespread throughout tropical regions (Phillips & Martyn, 1999). Castor bean is cultivated for the seeds which yield viscous, pale yellow non-volatile and non-drying oil. It has been used only for industrial and medicinal purposes (Ogunniyi, 2006; Ramos *et al*, 1984). Castor oil is one of the few naturally occurring glycerides with high purity, since the fatty acid portion is nearly 90% of ricinoleic (Akpan *et al*., 2006).The crude oil has district odour but can easily be deodorized in the refining process like any other vegetable oil. Castor oil has an advantage over other mineral oils in that it is biodegradable, eco-friendly and renewable resource (Ogunniyi, 2006).

The presence of ricinoleic acids, oleic acid, palmitic acid, stearic acid and dihydroxylstearic acid in castor seed oil is an indication of good quality that can be utilized for use in cosmetics and soap industries. Compared with the common vegetable oils, castor oil is more viscous, less soluble in hexane and more soluble in ethanol, all as a consequence of the presence of the hydroxyl acid (Abitogun *et al*., 2009). Apart from soap, it is the earliest anionic surfactant (Gunstone, 2005). One of its uses is in the manufacture of transparent soaps (Kochhar, 1998). Its major fatty acids are the unsaturated fatty acid, hydroxylated 12-hydroxy, 9-octadecenic acid known familiarly as the ricinuleic acid (RNA). The fatty acid composition of a typical castor oil contains between 87-90% ricinoleic acid. Thus the oil can be used in the production of vanishes, lacquers, protective coatings, lubricants, soaps, paints, inks and it is a primary raw material for the production of nylon and other synthetic resins (Wiley and Oeitmann, 1991). The ricinoleic acid is anti-microbial and has antibacterial, antiviral, anti-fungal, anti-inflammatory and analgesic properties. Its antifungal properties in particular have been traditionally used to inhibit the growth of fungus and fight yeast infections (Robertus, 1991).

Seeds other than castor bean seed had been a source for production of medicated soap but with different anti-microbial properties as carried out by Abdulrasheed et al. (2015) where carrot seed was used due to anti-fungal properties specifically on Trichophyton rubrum. The objectives of this research are to extract oil from castor bean seed through solvent extraction using hexane as the solvent, determine physicochemical characteristics of the oil extracted, produce medicated soap from the extracted oil and carry out quality control assessment on the soap produced to ensure it is within stipulated industrial and market standard. The research gap is the utilization

of oil from castor bean seed which is non edible not just for soap production but one with medicinal efficacy specifically for bacteria growth inhibition.

A large variability of seed oil percentage was observed, ranging from 39.6% – 59.5%. Whereas Ogunniyi (2006) found out that mechanical pressing will remove about 45% of the oil present and the remaining oil in the cake can be recovered only by solvent extraction.

II. MATERIALS AND METHODS

A. Preparation of beans

The castor beans obtained were first cleaned which was done manually. Hand cleaning was performed to remove foreign materials such as sand, sticks, stems and leaves. The cleaned beans were dried in the sun until the casting splits and the seeds are shed. These beans were further oven dried at 90^{o}C to a constant weight which further reduces the moisture content initially by 5-7% (Doan, 2004). The next step is winnowing, where the chaff was separated from nibs (Cotyledon) by blowing in a tray to promote high oil recoveries. The seeds were then crushed and ground using mortar and pestle to rupture cell walls for easy and efficient oil extraction.

B. Oil extraction

About 5g of dry ground seeds were first oven-dried at 105°C for 1 hour. The samples were refluxed for 6 hours in mild temperature (55-60°C) in a Soxhlet extractor using hexane as solvent. The solvent was then separated from the oil extracted by evaporation in a rotary evaporator. The extracted oil was kept in an oven at 60°C for 30 minutes after which was accurately weighed. The extracted seed oil was kept in a closed container and stored in a refrigerator at 4°C for further use.

C. Oil Characterization

In the characterization of extracted oil, the following physicochemical properties of the castor seed oil extract were determined:

(1) Specific Gravity

The specific gravity was calculated by dividing the oil density by the density of water (1000 kg/m^{3}) as presented in Equation (1) (TSE, 1971).

$$Specific\ gravity = \text{density of oil} / \text{density of water} \quad (1)$$

(2) Oil yield

5g of the sample was placed in the thimble and about 250ml of normal hexane was poured into the round bottom flask. The apparatus was heated at 60^{o}C and allowed for 10 hours continuous extraction. The solvent was finally distilled and the percentage of oil extracted was determined.

(3) Boiling Point

The boiling point of the oil was determined by heating the oil in a beaker placed on a heating mantle. The oil was observed carefully in the presence of a thermometer, immediately the oil started agitating and bubbling, the temperature on the thermometer was read and recorded as the boiling point of the oil.

(4) Refractive index

Few drops of oil sample were transferred into the glass slide of the Refractometer. Through the eyepiece of the Refractometer, the dark portion viewed was adjusted to be in line with the intersection of the cross. At no parallax error, the pointer on the scale pointed to the refractive index. This was repeated thrice using different oil samples. The average was taken and recorded.

(5) Peroxide value

This value is determined by titration. 5g of oil sample was transferred into a conical flask. 30 mL solution of Chloroform and Acetic acid mixture in the ratio 2:3 were added to it. The test was performed in an oxygen starved environment by passing nitrogen across the sample to remove residual oxygen. The solution was then saturated with potassium iodide to release free iodine which was titrated against 0.01 mol/L sodium thiosulphate. The peroxide value was determined from the titration volume of sodium thiosulphate as calculated in Equation (2).

$$\text{Peroxide Value (meq/g)} = (EP1 - BL1) \times TF \times R/SIZE \quad (2)$$

Where EP1 = Titration volume (mL)
BL1 = Blank Level (0.00mL)

TF = Factor of Reagent (1.006)
R = Constant number (10)
Size = Sample size (g).

(6) Acid Value

5g of oil sample was weighed into clean conical flask and mixture of 25 ml diethyl ether and 25 ml ethanol was added and used to dissolve the oil in the mixed neutral solvent. 1 ml of phenolphthalein added and the solution was carefully titrated with 0.1N KOH.

The acid value is calculated as thus in Equation (3);

$$Acid\,Value = MW\ X\ N\ X \frac{V}{M} \quad (3)$$

Where MW= molecular weight of KOH (g).
N = normality of KOH (mol/L)
V = volume of KOH used (L).
M = mass of the sample (g).

(7) Saponification Value

Using Indicator method, 2g of oil sample was weighed into a conical flask; 25ml 0.1N KOH was then added. The content was constantly stirred and allowed to boil gently for 60minutes. A reflux condenser was placed on the flask containing the mixture. Few drops of phenolphthalein indicator was added to the warm solution and then titrated with 0.5N HCL to the end point until the pink colour of the indicator just disappeared. The same procedure was used for the blank (Saad *et al*, 2007). The expression for saponification value (S.V) is shown in Equation (4).

$$\text{Saponification Value} = MW \times N\ (V_0 - V_1)/M, \quad (4)$$

Where
V_0 = the volume of the solution used for the blank test (L).
V_1 = the volume of the solution used for determination (L).
M = mass of the sample (g).
N = Actual normality of KOH (mol/L)
MW = Molecular Weight of KOH (g).

D. Soap Production

Two types of soap: Test soap (TS) and control soap (CS) were produced following the same steps only that castor seed oil extract was incorporated into TS while CS was not fortified with the extract. A commercial Antibacterial soap (Dettol) regarded as standard soap (SS) was purchased.

Production Steps

The fully boiled process was applied in the process of the soap preparation. 100 g of oil mixture (40% castor oil, 60% coconut oil) was measured and placed into a 500cm^3 beaker. It was warmed at 60^0C in order to quicken the reaction between alkali and fat. A calculated amount of NaOH was weighed and a fixed amount of distilled water was added to prepare aqueous NaOH solution. The caustic soda was stirred well using a stirring rod until it blends and completely dissolves in the distilled water. The aqueous NaOH solution was poured gradually into the oil mixture in the beaker and stirred gently in one direction to enhance thorough mixing of the alkali and fat. The beaker was insulated to prevent the fat from hardening and then heated at 60^0C for 30 minutes in order to achieve complete saponification. Once the alkali-oil mixture becomes pasty and starts to boil, it was poured into the mould and left to solidify.

II. SOAP CHARACTERIZATION

Standard analyses carried out on the soaps are:

- ***PH VALUE***

The pH meter was calibrated using buffer solution of pH between 4.0 and 7.0, thereafter it was dipped directly into the sample while the reading was taken immediately (Moulay et al., 2005).

- ***MOISTURE CONTENT***

10g of soap was weighed and reweighed after open heating for about 30minutes. The difference in weight gives the moisture content which is expressed in percentage (Moulay et al., 2005).

- ***FREE ACID CONTENT***

6g of soap sample was dissolved in 70ml hot neutral alcohol and titrated against 2M H_2SO_4 using phenolphthalein indicator (Moulay et al., 2005).
The free alkali/acidity was calculated as;

$$\text{Free acid content} = \frac{3.1MV}{W} \quad (5)$$

where:
M – Molarity of H_2SO_4 solution, $mol \cdot L^{-1}$;
V – Volume of H_2SO_4 solution used in titration, ml;
W – weight of the soap sample

- ***FOAM HEIGHT***

2g of soap was dissolved in a liter volumetric flask and made to mark with tap water, 50ml of the solution was introduce into a measuring cylinder such that it followed the walls of the column to avoid foaming. 200ml of the solution was taken in a conical flask and poured into a funnel, which was already clamped with the outlet closed. The measuring cylinder was then put directly beneath the funnel while the level (height) of the foam generated was read from the cylinder immediately the funnel outlet was opened (Moulay et al., 2005).

- ***ALCOHOL INSOLUBLE***

5g of soap sample was dissolved in 50ml hot alcohol and quantitatively transferred to already weighed filter paper; the residue was dried in oven at 105^0C for 30minutes, cooled in desiccators and weighed again (Moulay et al., 2005).

- ***ANTIMICROBIAL TEST***

The microorganism used in this study was staphylococcus aureus and the isolate was obtained from Abubakar Tafawa Balewa University Teaching Hospital Bauchi, Nigeria. Three soap samples (TS, CS and SS) 1 g/10mL were prepared and dilutions to (10^{-1}, 10^{-2} and 10^{-3}). Mueller Hilton aggar media was prepared for 9 plates (three plates for each test sample). From each prepared concentrations, 2ml of soap solution was transferred into a 6 mm bored well in a solidified Mueller Hinton Agar (MHA) plate which has been inoculated with the test isolate via streaking technique. Plates were kept for 30 min before incubation at 37^oC for 24 hours.The extent of susceptibility was recorded as clear zones of inhibition around the wells. The experiment was repeated three times for each test and average recorded.

III. RESULTS AND DISCUSSION

A. EXPERIMENTAL RESULTS

Table 1: Physiochemical Characteristics of castor bean seed oil.

Parameter	Value	Standard (ASTM, 2002)
Specific gravity	0.910	0.957-0.968
Refractive index	1.469	1.476-1.479
pH	6.11	-
Boiling point (^{O}C)	131	131
Oil yield (%)	47	-
Colour	Amber	Amber
Moisture (%)	0.30	-
Acid value (mgKOH/g of oil)	1.14	0.4-4.0
Saponification value (mgKOH/g of oil)	177	175-187
Peroxide value (meq/kg)	9.73	-

Table 2: Characteristic tests on produced Test soap (TS)

Parameter	Value	Standard (SON) (Nangbes *et al*, 2013)
Moisture content (%)	4.2	-
Hardness	Very hard	-
pH	8.9	6.5-8.50
Alcohol Insoluble (%)	3.45	≤2
Free Acidity	0.1	-
Foam Height (cm)	16	-

Table 3: Antimicrobial Effect of Soaps on Staphylococcus Aureus.

	Diameter of zone of inhibition (mm)			
	Stock (1g/10ml of soap)	Dilutions		
		10^{-1}	10^{-2}	10^{-3}
TS	15.5	11.5	9.5	6
CS	9.5	5.5	2	
SS	19.5	14.5	13	10.5

B. DISCUSSION OF RESULTS

Table 1 presents the result of the yield and physic-chemical parameters of castor bean seed oil. The result obtained for the percentage oil yield was 47%. This value falls within the range value of 30-55% as reported by Aldrich, (2003). The moisture content of the crude oil was 0.30% the low moisture content is as a result of effectiveness of the distillation apparatus used for oil recovery. Again, the low moisture content is an indication of good shelf life characteristics. The specific gravity was 0.910, which was a little below the value (0.948) as reported by Salunke & Desai (1992). The refractive index analysis showed a value of 1.4686. Comparing this result with the ASTM values that ranges from 1.476-1.479 (ASTM, 2002), a little difference is noticed. However, this little difference can be considered being within an acceptable experimental error range that can be attributed to the presence of some impurities and other components of the crude oil mixture. Thus, the refractive index of the crude castor oil was in accordance with ASTM specification.

Also, the pH value of the crude oil which was found to be 6.11 indicates that the oil is a little slightly acidic. Its boiling point was found to be 131^0C. The saponification value of oil was 177 mg KOH/g oil. The saponification value reveals how many milligrams of base required to completely saponify 1 gram of the oil or fat. The oils with high saponification value are considered to make better quality soaps than those with low saponification value. This value means that 177 mg of KOH is required to saponify 1000 mg of castor oil. This projects the oil as good in areas of soap making and in detection of adulteration in the oil. However, it was within the range value of 156 to 185 mg KOH/g oil reported by Weise (1983) and is found within the range 175-187 as specified by ASTM. The peroxide value was found to be 9.73 Meq/kg. Acid value was also determined as 1.14. The low peroxide value of the oil shows that the oil is less prone to rancidity and thus, stable. The lower the acid value, the lower the deterioration or rancidity of the oil. An increase in acid value results to increase in rancidity. The saponification value and acid value were in conformity with the range of values specified by the American standard for testing and materials (ASTM, 2002).

Table 2 presents the characteristic properties and quality criteria of medicated soap produced. The pH value of the medicated soap produced was 8.9 indicating the alkalinity of the soap, this value was slightly above the range 6.5-8.5 as specified by standard organization of Nigeria (SON) (Nangbes *et al*, 2014), foam height in castor oil soap was 16 cm, and this could be trace to the type of oil (coconut oil) which is known for its high formability due to the presence of lauric acid which constitute 50% of the total fatty acid in the oil. The value of alcohol insoluble is 3.45 which show the crudity of castor oil soap because no much builder, ingredients was

incorporated in the soap production process, it was above the SON recommended value of ≤2.00 (Nangbes *et al*, 2014). The moisture content was 4.2% and the value of free acidity was also found to be 0.10. The soap produced was hard due to presence of coconut oil.

Table 3 shows the microbial sensitivity of different soaps considered on (Staphylococcus Aureus) bacteria that is a common cause of jock itch, and Ringworm. The diameter of zone of inhibition of the soaps were measured and subsequently at various dilutions. For the stock solution (1g/10ml of each soap sample), the Test soap was sensitive with diameter of zone of inhibition 15.5mm but at a dilution of 10^{-1} , 10^{-2} and 10^{-3} it was found to be 11.5 mm, 9.5mm and 6mm. This shows that as concentration decreases, the sensitivity of the soap to the bacteria isolate also decreases. Moreover, the sensitivity of the medicated soap to the bacteria isolate is also as a result of the presence of ricinoleic acid present in large proportion in the fatty acid composition of castor oil.

`Also from Table 3, the microbial sensitivity of Coconut oil which is the control soap on (Staphylococcus Aureus) was evaluated. The diameter of zone of inhibition of the control soap was measured and subsequently at various dilutions; the soap was sensitive with diameter of zone of inhibition 9.5mm for the stock solution but at a dilution of 10^{-3} it was not sensitive to the bacteria isolate. Comparing the results of Test soap and Control soap, it was observed that castor oil has antibacterial properties due to increment observed in the inhibition zone of the medicated soap as compared to that observed control soap (coconut oil soap) with insignificant zone of inhibition. The microbial sensitivity of standard soap (Dettol antibacterial soap) on Staphylococcus Aureus was shown in Table 3. The diameter of zone of inhibition of the soap was measured and subsequently of various dilutions; the soap was sensitive with diameter of zone of inhibition 19.5mm for the stock solution but at a dilution of 10^{-3} it was found to be 10.5mm. The high sensitivity of the standard soap is as a result of chemicals such as triclocarban used as antibacterial agent in the soap which is effective in fighting infections by targeting the growth of bacteria such as (Staphylococcus Aureus).

IV. CONCLUSION

Castor oil soap was prepared from oil extracted from castor bean seed due to its high antiseptic properties. Quality parameters such as, alcohol insoluble, free acidity content, pH value and hardness were found suitable for production of Toilet/Bathing Soap. Antimicrobial activity of castor oil soap was tested against a bacteria isolate (staphylococcus Aureus) and was found sensitive.

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