Title

Biophysiochemical characterization of *Ricinus communis* seeds for biodiesel production.

Dissertation – II

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ABSTRACT

In today's world, as pollution and population is increasing, the requirement of petroleum based products also increases but these resources are non-renewable used in vehicles, machines and generators and due to higher consumption, depleted day by day. As prices of diesel and petrol reaches the highest level, the shortage of non-renewable sources increases, because they are exhausted in the earth's crust as utilization is increasing. The non-edible seeds of *Ricinus communis* are considered as one of the alternative resource for biodiesel production and have properties similar to those petroleum and diesel. In this present study, castor seeds of different regions were collected from India. Bio physiochemical analysis was performed in which morphological characterization was studied, including seed color, skin surface, and climatic conditions. The result of present study taken various samples were as, pH(6.24-7.33), oil yield(12.6-74.2%), fire point(110-190^{0}C), flash point (90-160^{0}C), saponification value (140.25-274.89mgKOH/g), iodine value (39.37-97.79mgKOH/g), acid value (0.602-3.520mgKOH/g), FFA(0.301-1.760mgKOH/g, specific gravity(0.432-1.240gm) and viscosity(184-300mm^2/s).The result obtained by the analysis confirms that oil sample obtained from Morena and Meerut were found to be the best quality for producing biodiesel. On the basis of characterization and parameter analysis, this is used for commercialization due to which it will bring profit. Hence, it has no harm, no emission of pollution in the environment as it is environment friendly and biodegradable. This may be utilized in the energy generation, soap making, cosmetics, and paints making it for industrial purpose. *Ricinus communis* is one of the best option to be used as an alternative fuel.

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Komal Sahlot

DECLARATION

I hereby declare that the dissertation entitled "**Biophysiochemical characterization of *Ricinus communis* seeds for Biodiesel production**" Submitted for M.Sc.(Hons.) Biotechnology Lovely professional University is entirely original works and All ideas and references have been duly acknowledged. It does not contain any work for the award of any other degree.

DATE: **KOMAL SAHLOT**

CERTIFICATE

This is to certify that Komal Sahlot student of M.Sc. (Hons.) Biotechnology Registration Number-11305803 has completed M.Sc. project entitled "**Biophysiochemical characterization of *Ricinus communis* seeds for Biodiesel production** "under my supervision. To the best of my knowledge the present work is the result of her original study and investigation. No part of the report has ever been submitted for any other purpose at any university/institute.

The project report is appropriate for the submission and partial fulfillment of the conditions of the conditions for the evaluation leading to award of Master of Science in biotechnology.

Dr. Kuldip Chandra Verma

(Supervisor)

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CHAPTER -1

INTRODUCTION

INTRODUCTION

The *Ricinus communis* plant belongs to Euphorbiaceae family, which is easily grown and resistant in adverse environment (drought). This species is called the castor oil plant and Palma Christi. *Ricinus communis* is a plant which is able to stand in arid environment and get adapted to that environment. The flowering in castor oil occurs after sowing about 45 days. The each part of this plant is capable for producing oil such as seeds, leaves and stem. It is found in subtropical and tropical countries in the world, both wild and cultivated states **(www.icrepq.com).**

The leaves of castor oil plant are used as an antibacterial and anti –inflammatory agent in the treatment of rheumatic pains. Concern towards depletion of petroleum & increasing of the environmental pollution therefore, finds an alternative the vegetable oil which easily derived from a plant like Castor which gives a non- edible vegetable oil **(Jain and Suhane, 2012).**

Castor oil have some unique properties hence it is used in cosmetic preparations and act as a vehicle for medicinal and it also remarkable the anti -dandruff effect therefore skin tolerated this in a well manner. In the presence of sodium salts when the distillation process taking place this castor oil solidifies or thickens to make a gel mass and this gel mass is used as a protective in the dermatitis and eczemas, consequently it is also used in the treatment of dermatosis **(Anandan *et al.*, 2005).**

The oil obtained from castor plant is used in several areas in making of explosives, lubricants, hydraulic fluids, soap synthesis and paints etc. It serves as a chemotherapeutic drug in case of cancer tumor in medical field. It is harmful in case of eating seed, which contains ricin and ricinine also which lead to the death if its intake is in excess amount and sometimes causes nausea. This plant is very ancient it is cultivated for their oil producing properties for centuries. Approx. more than 4000 years ago the Egyptians used this oil in their lamps **(Pashazadeh and Basalma, 2012).**

Castor oil is considered as GRASE (generally recognized as safe as effective) is approved by the united states, FDA (Food and drug administration) a derivative of castor oil are approved for the uses in the treatment of skin disorders, possibly it is used in the constipation but this treatment is not recommended. Drugs are not given in their pure chemical state; there are few drugs which rarely used in pure chemical form, some compound or additives which mixed with this medicine, and act as an active content. A derivate of castor oil is used in the numerous modern drugs such as Nelfinavir mesylate which act as an HIV protease inhibitor, Acijel its uses is found in maintaining the acidity of the vagina, Xenaderm ointment start up a uses in the skin ulcer treatment, Paclitaxel it works in a cancer chemotherapy as a mitotic inhibitor, and Miconazole used as an anti-fungal agent. In the food stuff and also in the food industry it used for flavorings and candy as chocolate. It also works as a mold inhibitor and packaging. The polyoxyethylated castor oil used in food industry as it is a non-edible oil it is not safe for consuming, but it make to be safe to eat by lower down its iodine and acid value by the procedure of the cold press process **(Burdock *et al.*, 2006).**

Castor oil is utilized as a raw material for the manufacturing and production of chemicals for example- chemical used in the jet as a lubricant (Dioctyl Sebacate), chemical which is used in the perfume formulation (Undecylenic acid) and castor oil is also used as a plasticizer (Sebacic acid) and it has a application to act as a raw material in the nylon-11 **(Das *et al.*, 1989).**

The castor is cultivated for its oil containing properties, the content of oil varies from ranging 40%-60%.Oil which is extracted from castor has a large variety of uses in the market such as in medicines, cosmetics, plastics, lubricants and it is known for its exclusive properties due to this a number of applications found in this technology world. They used to produce medicines which are used in skin cure problems, used in the problems linked with indigestion, menstruation irregularities, anti-fungal, anti-inflammatory, paints, plastics, transparent paper and brake fluid. The detergent after ordinary soap which is used in the many industries for the treatment of leather and industrial lubricant, it is first synthetic detergent which is called turkey red oil or sulfonated castor oil (sulfated) in preparing of solutions atropine, cocaine which is further used in ophthalmic surgery as a solvent of pure alkaloids. The fatty acid comprises 90% castor oil in this the content of ricinoleic acid is high (hydroxylated fatty acid). A poison (ricin) present in the bean, stem, and leaves of castor plant, it remains in the press cake and this cake is utilized as a manure and fuel for certain plant stalks. It also used for paper pulp preparation and it is harmful for human in taking of 4-8 beans kill the person, horse and ox **(Ozturk *et al.*, 2014).**

This castor oil is environmentally friendly and it is not a naturally occurring resource. It has a good shelf life in comparison with other vegetable oil due to this property and characteristics it is not freeze when it stored. The castor Oil is used in painting and in typewriter. It is used as a lubricant, protective covering and it is also used in textile industry for dying in addition also used in the preservation of leather. The castor oil has high viscosity which differs from the natural vegetable oil **(Salmon *et al.*, 2010).**

Fig 1: *Ricinus communis* plant

(www.cooltropicalplants.com)

Fig 2: *Ricinus communis* flower

(www.plant-worldseeds.com)

The aim of current study is for biophysico characterization of *Ricinus communis* seeds for biodiesel. The objective for this topic production of biodiesel has several reasons such as rising prices in petroleum increases their demand. In India, due to transportation, industrialization and mechanization consumption of energy increases with rapid rate.

This increasing price in terms of energy demand makes Indian economy insecure and a source of energy is a basic need for any country for their economic development. Environment is affected by this such as greenhouse effect, pollution therefore, to sort out this problem substitute should be chosen and it should be renewable and pollution free. The option for this comes to obtain it from plant, which found easily in Indian diversity.

Biodiesel (an alternative fuel) is produced when vegetable oil and animal oil reacting with alcohol and catalyst it produces the fatty acid which has similar properties as fossils fuels. It is eco-friendly, biodegradable and non-toxic. The production of biodiesel mainly done by crops, such as soybean, mustard and sunflower etc. in the world but if the edible oil is used here the nation faces the shortage of edible oil. To solve the problem non-edible oils are used for biodiesel production. The growing period of castor oil is short due to which farmers are aware for its cultivation. Biodiesel are fatty acids methyl or ethyl esters, which produced it from edible and non edible oil **(Shrirame *et al.,* 2011)**. The castor seeds cultivated in various regions in world which differ in size and outer external patterns but, most of them have oval shaped with shining brown brittle coats and marble grey in color and the height of castor is found to be different as plants belong to different regions 3m traditionally. In Europe 1.7m height was measured and the growing season 140 to 180 days, moderate rainfall required. Only grow in long periods of drought and storage of castor seed done at less than 6% moisture content **(www.oilseedcrop.org).**

There are various advantages by the use of biodiesel

Biodiesel contains low toxicity as compared with other diesel due to reduced emission of carcinogenic components their risk on health is low. It is obtained from vegetable oil of some plants although it is renewable. The emission of contaminants such as aldehydes, carbon monoxide and some particulate matter emitted through this is in a very low amount. Biodiesel has properties of lubricant; there is no emission of harmful gases sulphur dioxide. Over the advantages there are little disadvantages also such as in cold weather it is inconvenient because it is less stable in this condition due to higher freezing point and it is not recommended for long storage due to low calorific value it has slighter higher consumption than other diesel **(Romano and Sorichetti, 2011).**

Fig 3: Seeds of *Ricinus communis*
(www.camdengrey.com)

Fig 4: Fruit of castor bean (Cluster form)
(www.jaxshell.com)

For running the biodiesel in the engine no modification of engine is done it is stored pumps, tanks, transport, trucks etc. Biodiesel extracted out from castor is obtained from plant which can breathe CO_2 so no net gain CO_2.Therefore carbon dioxide residue emission is less often it is mixed with diesel fuel called as B20 similarly used by mixing it with petroleum diesel. This is biodegradable and non-toxic so, it stays safe for handling and transports well as also has an aroma of popping popcorn as compared with petroleum diesel fuel. Some problems also arise such as NO_2 emission is higher and consumption is higher as it contains O_2 **(Sreenivas *et al*., 2011).**

The uses of biodiesel are environmentally friendly and there are various oil crops which are used for production of biodiesels such as sunflower, soybean, oil palm, *Jatropha curcus* and *Ricinus communis* Castor oil plant seed known for its oil & it is a commercial crop in Ethiopia, a medicinal commodity. It is a shrub perennial can reach height of 12m.This one is fast growing non–hardy plant and now India is the one of the producer of castor oil plant. It is cultivated in coastal sandy belts and marginal lands of Indo–Pak under warm climate **(Chakarbarti and Ahmed, 2008).**

The fruits of castor oil are produced in the form of clusters the well-developed seeds in an each pod is sufficient to bearing oil 47-49%.The oil extracted from castor oil plant is pale yellow or viscous, colorless ,tasteless and no odor. The commercial importance of its oil is used as lubricants and also used in a number of industrial chemicals such as surface coatings, pharmaceutics and cosmetics **(Shrirmae *et al.*, 2011).**

As compared with other non-edible crops castor crop requires less inputs of water, fertilizer & pesticides, castor crop has ability to grown in the marginal lands frequent ally it grows at the day temp 20^{o}C the leaves of castor plant has insecticidal properties for this purpose it is grown in garden to resist flies and mosquitoes **(Abdulkareem *et al.*,2012).**

The profitability of castor is reduced because of its producing toxicity, its comparison with non-toxic oil crops the seed cake value of castor is low whereas other serves as a higher concentration of protein fodder supplement. By means of biofuel the Brazilian government supported castor as a biofuel crop in the north east of the country intended for the small farmers. On behalf of the settling of seeds of castor there is no too low or too high temperature & also avoided of humidity which increase the chances of pest problems. Castor plant is capable to mature in a stony land with a range of pH 4-8.3.This plant shows little response to phosphorous if there is the excess concentration of nitrogen it does not increased the seed yield but enhance their vegetative growth. The castor does not loss their economic importance until it is attacking by several pest & diseases castor crop is alternated with cotton ,groundnuts, horse gram, tobacco and finger millet in India, along this the leaves of castor is used to provide feed the silk worms in china.

Fig 5: Biodiesel cycle (www.alternative.energy).

“Blown oil” the oil which remains as a fluid in low temperature it produces to allow the partial oxidation of castor oil at 100^{o}C it is used in making of leather, varnishes and hydraulic fluid. Engines like jet engines for airplanes it works under extreme conditions. The oil gotten from castor polymerized when there is increases in temperature due to rich in esters and these ester doesn’t decompose at high temp 650 F more exposure of castor in the temperature outcomes the better or improved lubricant which found the stability at room temperature. The first detergent coming after the ordinary soap is sulfonated castor oil or Turkey red oil which is used in the several industrial purposes moreover the oil of castor is used as a solvent of pure alkaloids & ophthalmic surgery **(Nielsen *et al.*, 2011).**

CHAPTER- 2

REVIEW OF LITERATURE

REVIEW OF LITERATURE

The chains of fatty acids comprise a medium length C16-C18 in which the mono alkyl fatty acid esters were noticed in the biodiesel fuel these are non-toxic, free from the sulphur and carcinogenic ring **(Yamane *et al.*, 2001).**

A studied was done by **Conceicao *et al.* (2007)** in which physical chemical properties and comparison between crude oil and its biodiesel was done which show a variation in the value of viscosity and flash point. The flash point of castor oil was 310^0C and castor biodiesel was 120^0C but the ASTM D 93 (93^0C).The density determination was done at 15^oC and 20^0C (g/cm^3) the value for castor oil was 0.957 and castor biodiesel was 0.927 and the ASTMD1298 according range 0.875-0.9 whereas, density at 20^0C ASTM1298 the ASTM limits 0.86 value of biodiesel was 0.924 and castor oil 0.958.The viscosity (mm^3/s) was observes castor oil (293.39) and biodiesel (13.75) and ASTMD445 have range of limits (1.9-1.6).

High hygroscopicity, high stability and its solubility (in alcohol) is a distinguishing of castor oil because of its transesterification reaction it was affected due to its less solubility in methanol **(Conceicao *et al.*, 2005 and Han *et al.*, 2005 Scholz *et al.*, 2008).**The Ricinoleic acid found in this is polar in nature and the transesterification is taking place in the presence of a strong base and strong acid when triglyceride is reacts with alcohol and it produces a glycerol and a mixture of fatty acid alkyl esters if the alcohol is used in excess amount, it favors the product formation and this transesterification is influenced by the molar ratio of alcohol and oil. It is a triglyceride, in which the 90%fatty acid chain is ricinoleic acid a monounsaturated 18-carbon fatty acid & other components such as oleic and linoleic acids **(Chakarbarti and Ahmad, 2008).**

Biodiesel production from the seeds of castor have an extraordinary demand in terms of market values due to carbon content which was to be low, the storage of biodiesel is safe providing the nation an energy security along with this it is biodegradable and also safe for environment, having high lubricating properties. It is a clean burning fuel through which biodiesel released from this have low sulphur & carbon dioxide rather than the petroleum diesel. sometimes for biodiesel production transesterification is done but to achieve a high yield of biodiesel enzymatic transesterification was perform but the enzyme has a high cost ex-lipases hence not followed the enzymatic transesterification and also in the transesterification the catalyst removal and purification of product is creating a problems so a process of BIOX preferred the properties of biodiesel was studied like specific gravity which gives the value(0.87-0.89), flashpoint having the value between (120-130) ^{0}C iodine number a range is followed (60-135) & the viscosity was estimated at 40^{0}C to give value (3.7 to 5.8) in mm^2/s(**Demirbas 2003, Balat 2005, Nezahat *et al.*, 2009**).

The properties of castor seed oil was examined by **Deligiannis *et al.* (2009)** and the various of physical and chemical parameters results obtained such as kinematic viscosity was measured at 40^{O}C (240.12) ,density at 15^{0}C was(967.3kg/m^3),the flash point (greater than 260^{0}C) acid value was obtained around (0.7 mg KOH/g), free fatty acid value in percentage was (0.35) the iodine value was measured in cg I/g oil 84.4 and 181.4 was the saponification value in (mg KOH/g).

According to **Akpan, (2009) and Bengali *et al.* (2010)** for the quality of castor oil ASTM specification was found which says about result of oil yield percentage i.e. 48.75 % the property of castor oil considering acid value range matching to ASTM 0.4-4.0 but experimental value was 2.2. Saponification value obtained 181 and ASTM range was in the line of between 175-187, the iodine value gets from the experiment was 87 and ASTM followed the range between 82-88, whereas specific gravity and viscosity at 25^{0}C the value obtained after experimental determination was 0.95 and 6.9 on the contrary of ASTM range 0.957-0.968 and viscosity 6.3-8.8. The pH value obtained was 6. The study was done on the Malaysian castor for physiochemical properties such as the saponification value was 182.9 mg/g, iodine value mg/g was 84.5 mg/g viscosity was measured in cP 332 & the acid value of Malaysian castor seed oil was 4.9mg/g **(Salimon *et al.*, 2010).**

Ricinus communis oil is produced by the method which is known as soxhlet extraction method and solvent used in this is n- hexane. The result finds that the oil extraction from castor oil for biodiesel production is more viscous as compared with other oil for this reason the viscosity creates a problem. Sulfonated castor oil form of this oil is used in the dyeing of clothes a particular kind of cotton fabric and oil of castor is more viscous so viscosity problem is eliminated by mixing the castor biodiesel with other biodiesel, density and viscosity of castor oil is higher than other biodiesel. The result obtained was kinematic viscosity of castor biodiesel at 40^{0}C 11.5 mm^2/s density was determined at 15^{o}C 920kg/m^3 and the flash point was greater than 130^{o}C.when castor biodiesel was blended with soybean and examined the density and kinematic viscosity which varies from 5% castor biodiesel and

95% soybean gives the viscosity 4.5mm^2/s and density 887kg/m^3.if the 10% castor biodiesel and 90% soybean was blended the result of viscosity and density was 4.9 mm^2/s and 888 kg/m^3 then 20% castor biodiesel was taken with 80% soybean biodiesel it gives 5.6mm^2/s viscosity and 891 kg/m^3 density. The value increases when 50% of soybean and 50% of castor biodiesel blended 7.3mm^2/s value of viscosity and 902 kg/m^3 value of density was obtained. The biodiesel which is pure obtained from castor cause obstacle due to its highly viscous nature in the injection system **(Saribiyik *et al.*, 2010).**

Properties of castor oil was studied by **Encinar *et al.*, 2010** where the density was determined at 15^{0}C (961.20kg/ m^3),acid value (1.09 mg KOH/g),molecular weight(918.12 g /mol),viscosity was measured at 40^{0}C (0258.01cst.), Iodine value was obtained (84.19gI2/g) and (180.33mgKOH/g) was the saponification value.

The different physical properties of castor oil was studied and observed the various parameters the acid value was 0.022 mg KOH/g ,density was 993kg/m^3 and viscosity at 20^{O}C was 961 × 10^{-6} m^2/s, while estimation at the 40^{0}C its value decreases 268 as the temp increase the kinematic viscosity value decreases when the viscosity was measured at 70^{0}C obtained value was 61.9 the study was done also on the basis of the properties of methyl ester of castor which gives the acid value 6.24×10^{-3}mg KOH/g, density 956 kg/m^3 and kinematic viscosity was 10^{-6}m^2/s at 70^{0}C it was 15.8 whereas at 40^{0}C 19.4 was the value which recorded while at the 20^{0}C the value was recorded 40.6 **(Shrirame *et al.*, 2011).**

The outcome for the viscosity of crude castor oil was 0.96kg/m^3 then it was treated to esterification the value decreased to 0.89kg/m^3 and the value of biodiesel was 0.85kg/m^3 which is lower than castor hence the high value of castor for viscosity was reduced further by doing blending of oil. The flash point of biodiesel was around 50^{0}C and for B100, B20 and B10 value was 190^{0}C, 89^{0}C, 85^{0}C similarly the kinematic viscosity of castor oil was 200mm^2/s at temperature 30^{0}C reduced further for B100 15mm^2/s, B20 the value was 4.97mm^2/s and B10 4.45mm^2/s but after blending the ASTM limits (1.99-1.6) for biodiesel **(Bello and Makanjo, 2011).**

Ricinus communis is a plant whose oil is hardy and non- drying oil which neither become thick with Cold and nor thin with heat this is the reason that it is used as a lubricants in racing car engines and in jet.

The consumption of fuel is 10% which is very low in comparison to diesel fuel this comes because of the difference in heating values of the fuel the specific gravity is lowered for biodiesels. About 97.7% unsaturation is present in castor oil whereas saturation is 2.30%. It is soluble in methanol due to this solubility without heating and uses of catalyst it gives the higher yield of biodiesel **(Bello and Makanjo, 2011).**

According to the study **Sreenvias *et al.* (2011)** the method of trans esterification is done for the conversion of crude oil or oil in presence of NaOH catalyst with the help of methanol and ethanol convert it into useful product biodiesel, a fatty acid alkyl ester, the dye used for cracking of vegetable oil molecule is lye work as a catalyst .A successful survey is done in 2002 year in which a report of reduction in fuel bill is found by Indian railway by the 5% use of biodiesel. The bio physical properties determined by this method the value of castor oil density before esterification is $950kg/m^3$ and after the esterification was done its value is $925kg/m^3$.This is totally differ when it is esterified by using of mineral turpentine oil it is reduced more and reaches $878kg/m^3$.The kinematic viscosity determined is $220mm^2/s$. After esterification the value is $24mm^2/s$. It get better characteristic when it mineral turpentine oil is used and the value comes is $5.5mm^2/s$ and the iodine amount absorbed by the castor oil ’’iodine value” is 100 along with the using of mineral turpentine oil its value is reduced to 80.

Castor oil plant has low solidification point (-12^0C-18^0C),boiling point is 313^0C density is $961kg/m^3$ low melting point (5^0C) high molecular weight (298KD).This is the only vegetable oil which has high proportion of hydroxyl fatty acids. The viscosity of castor oil is 100 times more the diesel oil. The functional group in ricinoleic acid and castor oil makes it unusually polar, a hydroxyl group which makes valuable in chemical feedstock. In the carbon 12 the group present (hydroxyl, carboxyl and double bond) is a source for the biodiesel production study. In the castor oil the viscosity value is high, for this reason the obstacle is coming from the production of oil yield to eradicate this problem the alternative of transesterification is taken for high increasing yield of biodiesel production and improves its quality.

The oil attained from the castor oil plant for the production of biodiesel is not used in the cold countries without adding any additives because the cloud point is very high in castor oil plant which is not suitable for making biodiesel, but with the use of additives the castor oil biodiesel is preferred in cold countries as the cloud point of castor oil is lower in comparison to other biodiesel plant such as Jatropha, additives are used to improve their cold filter plugging point, i.e. CFPP **(Okullo *et al*., 2012).**

Table 1: Comparison of Physiochemical properties of crude oil and neutralized oil (Okullo *et al.*, 2012)

Property	**Crude Oils**	
	Jatropha	**Castor**
Acid Value [mg KOH/g]	3.38±0.23	2.41±0.03
FFA [%]	1.70±0.46	1.21±0.02
Kinematic Viscosity [mm^2/s]	33.86±1.92	222.00±6.93
Flash Point [^0C]	292±1.00	294±0.58
Density [Kg/m^3]	910±2.64	940±3.00
Cloud Point [^0C]	2±0.21	14±1.00
Oil Yield [% wt.]	52±1.13	56±3.13

From the above Table 1, it is concluded that as the acidic value of crude oil of Jatropha is higher than that of the castor oil but the production yield of the castor oil is higher than the Jatropa oil but the oil yield of castor oil is high.

Castor oil has a number of industrial applications like a coating agent as well as also a component of plastic products subsequently it acts as a fungicide and known importance in making biodiesel fuel. The more number of increasing the requirement of biodiesel more the cultivation and production of castor plant which also give result a large amount of byproduct which obtained after oil extraction .The castor cake by-product have high protein content and fiber Along with this its usage is found in the fertilizers on the other hand a limit is found in the use of this byproduct the finding of toxic compounds due to which there is restriction to use it for animal feed. Another problem also originate related to the causing of health problems for those peoples who work with the castor seeds because of the presence of allergenic proteins and ricin a toxic compound which is found in the endosperm portion of castor seed. some process were carried to remove the toxicity and allergen effect such as, castor cake was treated with some chemical solution Cao and Ca $(OH)_2$ after the treatment of castor cake there is no risk to use the castor cake as the byproduct is freeing from toxicity **(Fernandes *et al.*, 2012).**

According to the experimental analysis done by **Penugonda and Venkata, (2012**) the vegetable oil used in this has high smoke emission due to their high viscosity and carbon residue so by the trans esterification reaction modification is done reaction with ethanol and methanol and convert in alkyl esters which is fatty acids 'biodiesels' and it was done in the presence of catalyst NaOH. The estimation of castor oil flash point is 230^0C and after the esterification in presence of catalyst its get value 200^0C but when it was treated with mineral turpentine oil it is 125^0C as compared with normal diesel its value is higher and this is the reason it is safe to store. Kinematic viscosity determination was done in refer to thickness of the oil and its measure as the time taken by an oil to passes through a specified size of orifice. The viscosity of castor oil is more as compared with other biodiesel i.e. 100 times more than other oil. At 40^OC its value is 240mm^2/s and after doing transesterification it was 24mm^2/s then treatment with mineral turpentine oil it gives the value 5.5mm^2/s the value obtained is comparable with the viscosity value of biodiesel 2.27mm^2/s along with these fuel characterization density is also a parameter it defines as mass per unit volume as more energy therefore oils should be more denser and density measurement was done at 15^OC value obtained 950kg/m^3 this value reduces when esterification is done value obtained was 925kg/m^3 and it further reduced when castor oil is treated with mineral turpentine oil 878kg/m^3 and the density biodiesel (880kg/m^3)with this it is comparable. The iodine value treated with mineral turpentine oil was 80, esterification its value reaches at 90 and for castor oil the iodine value is 100.

The biodiesel was obtained from both the resources edible and non-edible and also from the animal fat if the biodiesel were produced from the edible oil by taking consideration of sesame oil, rapeseed oil, sunflower oil, and soybean oil. This situation rise the human nutrition chain imbalance and economically wastefully than the fuels which derived from petroleum, therefore non-edible oil was recommended for the commercial use of biodiesel for their production castor and jatropha seed capable to fulfill the source for biodiesel manufacture **(Antony *et al.*,2011 , Sruthi 2013).**

According to **Bale *et al.* (2013)** the various physical and chemical parameters were taken into consideration such as specific gravity, density, saponification value, acid value, free fatty acid value and iodine value. The observation of density value was recorded 880g/cm^3 for castor seed oil, the saponification value was 327.4 mg/g, the iodine value results 1.061 I_2 g/100g, the value of acid was 14.42 mg/g and percentage of free fatty acid was 7.21.

According to **Bale *et al*. (2013)** the acid value of the castor oil was obtained (14.4mg/g) and the acid value obtained by **Ogunniyi (2006)** was (10mg/g). Its saponification value (327.4mg/g) which is almost two times more as compared to the **Jumat *et al.*(2010)** which get value 182.9 mg/g and **Ogunniyi(2006)**value obtained was 177-182mg/g .It may be due to the quality of oil and environmental conditions which may be unfavorable.

The most advance research was done on castor oil plant that the leaves and stem of *Ricinus communis* is utilized for the Bio methane generation, when it goes under anaerobic condition. This involve in the production significantly high amount of biogas. By taking a reference of cow dung, *Ricinus communis* has potential for the biomethane production **(Yauri *et al*., 2013).**

According to **Nagbes *et al.* (2013)** the determination of physical and chemical properties were done in which the percentage of oil content found in the castor sample was approximately 48% the pH value of castor oil was obtained 5.8 & the viscosity (cps)was determined at 28^0C and results the 0.425± 0.120.12 the flash point lie in the range of 256 ^{0}C±1.10 and fire point 225 0±2.10 the density found was 0.948± 0.0 and the saponification value found in an average line (180.770± 0.32) mg KOH /g whereas iodine value or Wij's value obtained 58.64±0.71.

Castor oil having the density about 961kg/m^3 generally, it is pale yellow liquid 90% of the carbon chains of fatty acid was the ricinoleate & other than ricinoloate, oleate and linoleates were correspondingly the compound which was found in the fatty acid chain of castor oil. To diminish the health risk issues of toxic substance ricin, investigators undertaking this for doing the modification genetically to stop the production of ricin synthesis **(Abdelaziz *et al.*, 2014).**The biophysicochemical characterization was done under which the analysis of refined and crude oil taking place in CLSER(central laboratory of science, environmental and soil research) crude oil gave the specific gravity 0.963 and refined oil 00.96. The determination of viscosity was measured in cost at temperature 40^0C & 100^oC the crude oil recorded value was 234.07 and 18.79 however refined oil results the value 209.6 and 18.30. Measuring of some chemical parameters and thermal properties of castor oil were also done according to ASTM and British pharmacopeia 2007, the iodine number of crude castor oil was 86.98 and refined oil get value 84.23, the flash point of crude oil was 305^0C According to ASTMD93 acid value of crude castor oil was 1.231 and refined oil was 0.916 whereas saponification value recorded for crude oil & refined oil was 179.33

and 178.56 the obtained results was comparable with the castor oil quality and followed under the ASTM range (**Akpan *et al.*, 2006, Jose, 2011 and Abdelaziz *et al.*, 2014).**

The properties of castor oil that was extracted from castor seeds were experimented by hot method and cold method. When the hot method was select these are the parameters and obtained value of crude oil & refined oil. The pH of crude castor oil resulted was 6.3 and refined oil was 6.5 as well as the pH value of hot castor oil was 6.7, the specific gravity of crude oil was 0.960 and after refining the value obtained was 0.952 and get value 0.959 after applying hot method. Kinematic viscosity (mm^2/s) measurement was performed at 40^0C the crude oil having value 243 after refining of oil it was 241 and by the hot method value evaluate was 238. The acid value (mg KOH/g) and FFA (%) value of crude oil calculate was 5.68 and 2.48 in the manner of refining treatment get a reading 5.92 and 2.96 and by the involvement of hot method there is dropping in the values 3.12 and 1.56. The iodine value (g I_2/100g) estimated by hot method results 74.08 while refined oil get 62.23 and crude oil 71.12 and the saponification value (mg KOH/g) of crude oil was 185.04 ,after considering refining get a value 173.16 and after doing the hot method the value was 181.52.

The outcome achieved by doing the physicochemical properties, examined the three types of biodiesel which was produced from crude oil, refined oil and cold methods. The wide range of pH values of these three was recorded (6.5,6.7 and 6.1) the kinematic viscosity measurement was completed at 40^oC results coming for crude 24.9, refined oil its value was 24.6 and hot method biodiesel 23.8 and the free fatty acid (%) obtained for crude was 2.18,refined was 2.74 and value obtained after performing hot method biodiesel was 1.90 whereas acid value (mg KOH/g) was (4.36,5.48 and 3.80) The iodine value of these three type of oil obtained from castor seeds was (62.23,71.12,and 58.34 g I_2/100g) saponification value recorded was (181.10,171.04 and 179.06) in mg KOH/g. The differences in the value was getting because of the different types of procedures was used for oil extraction, various solvents and chemical used for refining process. Therefore it was concluded that cold extraction method was not better as such hot extraction method because in industry good quality and high oil yield was recommended **(Harbawy and Mallah, 2014).**

Castor oil the density was 961kg/m^3 mostly it is pale yellow liquid 90% of the carbon chains of fatty acid was the ricinoleate and other than ricinoloate oleate and linolaetes were also the compound which was found in the fatty acid chain of castor oil. There are also some health risk issues of toxic substance ricin but investigators doing the modification genetically to stop the production of ricin synthesis **(Abdelaziz *et al.*, 2014).**The biodiesel has an advantage because of its nature which is slightly viscous along this its viscosities lie near by the petroleum diesel the problem begins in the ignition, engine starting and drop in the thermal efficiency, A droplet like form was found in the injection due to which spreading in the exhaust smoke and combustion happens was poor so, the poor atomization results due to high viscous nature **(Alnumami *et al.*, 2014).**

The study done by **Okechukwu *et al*.(2015)** showed in the Table the work on characterization of castor oil the physical and chemical properties was studied by them such as pH specific gravity, fire point ,flash point, color ,acid value, iodine value ,viscosity, and saponification value. The properties were characterized from extracted castor oil crude and refined biodiesel. In the chemical properties the acid value in NaOH mg/g of castor oil was 15.71×10^{-3} the resulted iodine value was 75.82 g I_2 /100g of oil and percentage of FFA value obtained from castor oil was 7.855×10^{-3}.

Table-2: Comparison between the crude and refined oil of castor on the basis of physical properties studied by Okechuku *et al.* (2015).

Properties	pH	Color	Viscosity (40 ^{0}C)	Specific gravity	Flash point (^{0}C)
Crude oil	7.8	Amber	6.48mm^2/s	0.9595	135
Refined oil	9.2	Yellowish	6.48mm^2/s	0.8658	140

From the above Table 2, the characterization performed on the basis of viscosity, flash point, specific gravity, pH and color between the two forms crude oil and refined oil. It was observed that flash point of refined oil higher as comparison with refined oil consequently the specific gravity of crude oil was higher.

CHAPTER -3

SCOPE OF STUDY

SCOPE OF THE STUDY

In today's world, the demand of petroleum products is very high as time passes day by day it is expected that in the coming years these resources is depleting. The fuels (petroleum based) are stored in the earth, but in a short period of time these coal pits are vacant because today's world are very dependent on the petroleum based products such as increasing number of vehicles & machines , more the depletion of petroleum diesel products which is not renewable. With the limitation of this the prices of diesel and petrol increases regularly. Along with it causes the harmful impact on environment to create pollution. A large amount of toxic gases emitted which have impact on plants ,animals and environment due to these consumption fossils fuels giving toxic gases and some compounds which is organic such as CO_2, NO_2, SO_2, CO, Aldehyde, polycyclic aromatic hydrocarbon etc. All these are responsible to disturb the maintenance of environment causes acid rain, pollution and global warming. Due to these toxic effects number of problems or complications arises like heart disease, cancer and respiratory problems take consideration of these problems search for the energy sources which decrease effects for that reason biodiesel are produced from biomass (plant and animal) used as an alternative source.

Therefore, this study is the finding of the alternative source which is used in a diesel engine, running in automobiles, Generators, etc. It matches with the similar properties of the traditional fossil fuel which is petrol and diesel. The production of biodiesel is done by both edible and non-edible plants but in our study the production of biodiesel is completed by using of non-edible plants. Hence, in future there is no declining of natural flora of edible plants and to recognize the data on the cost & demand of edible oils in the world. Subsequently the non- edible oil is preferred for biodiesel production in today's world.

Certain advantage for the production of biodiesel results that it has less environment impact in regards with increasing pollution, environmental friendly as it is biodegradable in nature. The complete combustion taking place which emits the emission of CO (hydrocarbons) is significantly low and fuel obtained is renewable, low toxic, low health risk so there is no emission of SO_2, aldehydes, aromatic hydrocarbon while it has a property of good lubricant. Castor oil is also used in the production of nylon6, coating, ink, varnishes, soaps, surfactants, synthetic resins etc.

To solve out the problem of increased oil demand in this world, continuous exhaustion of petroleum resources energy security problem arises, which is resolved by implanting the new technologies, improved by government policies and funding in discovering of renewable energy resources. Later importance will be given to non–edible oil such as *Ricinus communis*. Towards preventing the world from food shortage problem, environmental degradation and protect the agricultural lands from pre–exploitation.

CHAPTER -4

OBJECTIVE OF THE STUDY

OBJECTIVE OF THE STUDY

(1) Extraction of oil and biodiesel formation from *Ricinus communis* seeds from different geographical locations

(2) Bio physiochemical characterization of *Ricinus communis* seed oil for biodiesel production.

CHAPTER- 5

RESEARCH METHODOLOGY

RESEARCH METHODOLOGY

5.1. Sample collection process

Ricinus communis seeds were collected from the different regions in India (Table 3).

Table-3: Different regions where sample of castor seed was collected

S.no	Sample	S.no	Sample
1.	Ludhiana (Punjab)	2.	Sangrur(Punjab)
3.	Nawasehar (Punjab)	4.	Udaipur(Rajasthan)
5.	Jammu	6.	Gwalior(M.P)
7.	Haryana	8.	Bikaner (Rajasthan)
9.	Meerut (U.P)	10.	Nampalley
11.	Morena (M.P)	12.	Ghaziabad(U.P)
13.	Mandi (H.P)		

Fig 6: Sample collected From (A) Nawanshahr,(B) Ludhiana, (C)Ghaziabad and(D) Gwalior

5.2. Preparation of seed sample (Castor bean processing)

The seeds which were collected from different regions, only healthy and clean seeds were selected whereas, the damaged seeds were discarded and the castor seeds were exposed to dried under sunlight, before using it in a lab it was dried in Hot air oven at temperature 60^oC-65^oC, for 6-7 hour to eliminate the moisture content Seeds were crushed, in which cell walls rupture. And make a castor bean paste and separated into seeds and shells, grinded to fine powder using the mortar pestle and electric grinder.

Fig 7: Homogenized seeds of *Ricinus communis*

5.3. OIL EXTRACTION

50 gram fine powder was weighed and packed into a filter pouch which was transferred into a thimble of soxhlet apparatus for oil extraction.300 ml n-hexane used as a solvent was poured into a round bottom flask which was attached to the soxhlet extractor. For the continuous oil extraction heating is supplied by the mantle heater at constant temperature at 60^0C-65^oC for 8 hours. After completion, the crude oil from different samples collected from different regions taken and going to distillation process.

Fig 8: Soxhlet Apparatus For oil extraction

In the soxhlet apparatus, homogenized sample through which the product is formed for further use, the sample which is homogenized is put into the thimble which was made up of filter paper for commencement of the extraction process. The solvent which was extracted coming into the flask onto which solvent extractor was placed that was attached with a condenser. For supply heating placed the flask in a heating mantle the solvent was heated for reflux after that heating vaporizes the solvent to distillation arm and revert back. Then, solvent vapor cools down with the condenser and warm condensate solvent fill in the chamber. Compound is then dissolved in solvent, the cycle allowed and repeated many times.

The sample from different regions were run in a Soxhlet apparatus and collected for further distillation:-

Fig 9: Extracted out crude sample of (A) Ghaziabad and (B) Sangrur (Punjab).

Fig 10: Extracted out sample of oil of different regions (A)Hyderabad,(B)Udaipur,(C)Jammu,(D)Ghaziabad,(E) Sangrur,(F) Nawasehar,(G) Bikaner,(H)Ludhiana.

5.4. DISTILLATION OF EXTRACTED OIL

After the extraction of crude oil, distillation process was done at 60^0C.In this process the oil was separated out from the organic solvent. All the samples were distilled out and the oil was collected.

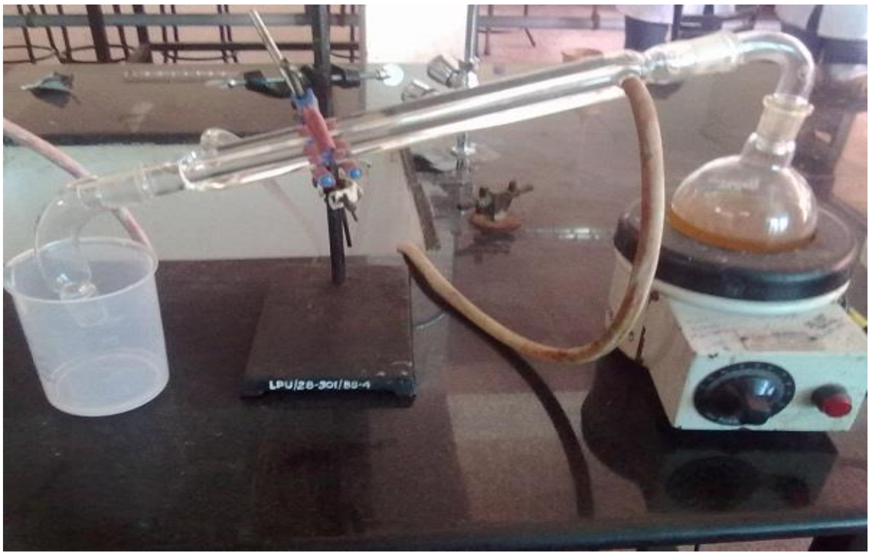

Fig 11: Distillation for separating oil and organic solvent (n-hexane).

5.5. TRANSESTERIFICATION PROCESS

A process includes in the presence of catalyst fat or oil is reacted with methanol to produce glycerin and methyl esters or biodiesel. Transesterification of different samples was done step by step as follows-

1 ml of oil sample was taken into a round bottom flask and adds the 25ml methanol into it, after that weighed the 0.4gm of sodium metal quickly and added in the round bottom flask which contains the sample. For removing the impurities and salt content these were refluxed for 2 hours at 40-60^{0}C by kept it on heating mantle. When 2 hours completed switched off the reflux and allowed the sample to cool. Sample is used for next step for the production of biodiesel

Fig 12: Transesterification Oil sample kept for Reflux.

5.6. TRANSESTERIFICATION REACTION

$$\begin{array}{l} CH_2\text{-}COOR_1 \\ | \\ CH\text{-}COOR_2 \\ | \\ CH_2\text{-}COOR_3 \end{array} + 3ROH \xrightarrow{\text{catalyst}} \begin{array}{c} CH_2OH \\ | \\ CHOH \\ | \\ CH_2OH \end{array} + \begin{array}{l} R_1COOR \\ | \\ R_2COOR \\ | \\ R_3COOR \end{array}$$

TRIGLYCERIDE+METHANOL $\longrightarrow$ GLYCEROL+BIODIESEL

(1)

(2)

(3)

(4)

Fig 13: (1) Methyl Red is added into the Refluxed sample and distilled water. (2)50% conc.H_2SO_4 is added in the funnel for giving the pink color. (3)Petroleum ether is added and mixed it well, ether is on the upper phase having biodiesel. (4)The lower layer contains the water and impurities and upper layer containing Biodiesel.

5.7. PRODUCTION OF BIODIESEL

A product which one gets from different samples, oil is extracted from that and biodiesel is produced chemically reacts with methanol to produce fatty acid methyl esters and glycerol. A catalyst is used to improve the rate of reaction sodium metal was used as a catalyst which is a strong base. Transesterification is performed for producing biodiesel.

These are the following steps

- 250ml separatory funnel is taken and kept it on a funnel stand for holding the funnel. Add the refluxed sample and 25ml of distilled water to the separatory funnel. Mix it gently.
- A 0.2ml methyl red indicator is added which indicate the reaction by producing yellow color.
- 50% H_2SO_4 (conc.) was added into it until it changes into the pink color and shake it properly for mixed the contents present into this separatory funnel.
- After that 100ml petroleum ether was added into a separatory funnel again shake it for mixed the contents.
- Two layers was seen ether forms upper layer at the top whereas glycerin forms the lower layer at the bottom of the separating funnel allowed these two layers to stand without disturbing it .by removing the lower layer and take the upper layer which was washed by distilled water (2-3 times) this upper layer containing the biodiesel.
- For settle down the impurities allowed it to stand for overnight and washing was done with 3-4times with distilled water after washing, this biodiesel containing upper layer is taken into a beaker then 4-5gm of anhydrous sodium sulphate is added into a beaker and kept it overnight.
- Kept it open in beaker until only biodiesel is left and store the biodiesel in bottle.

5.8. FLOW CHART

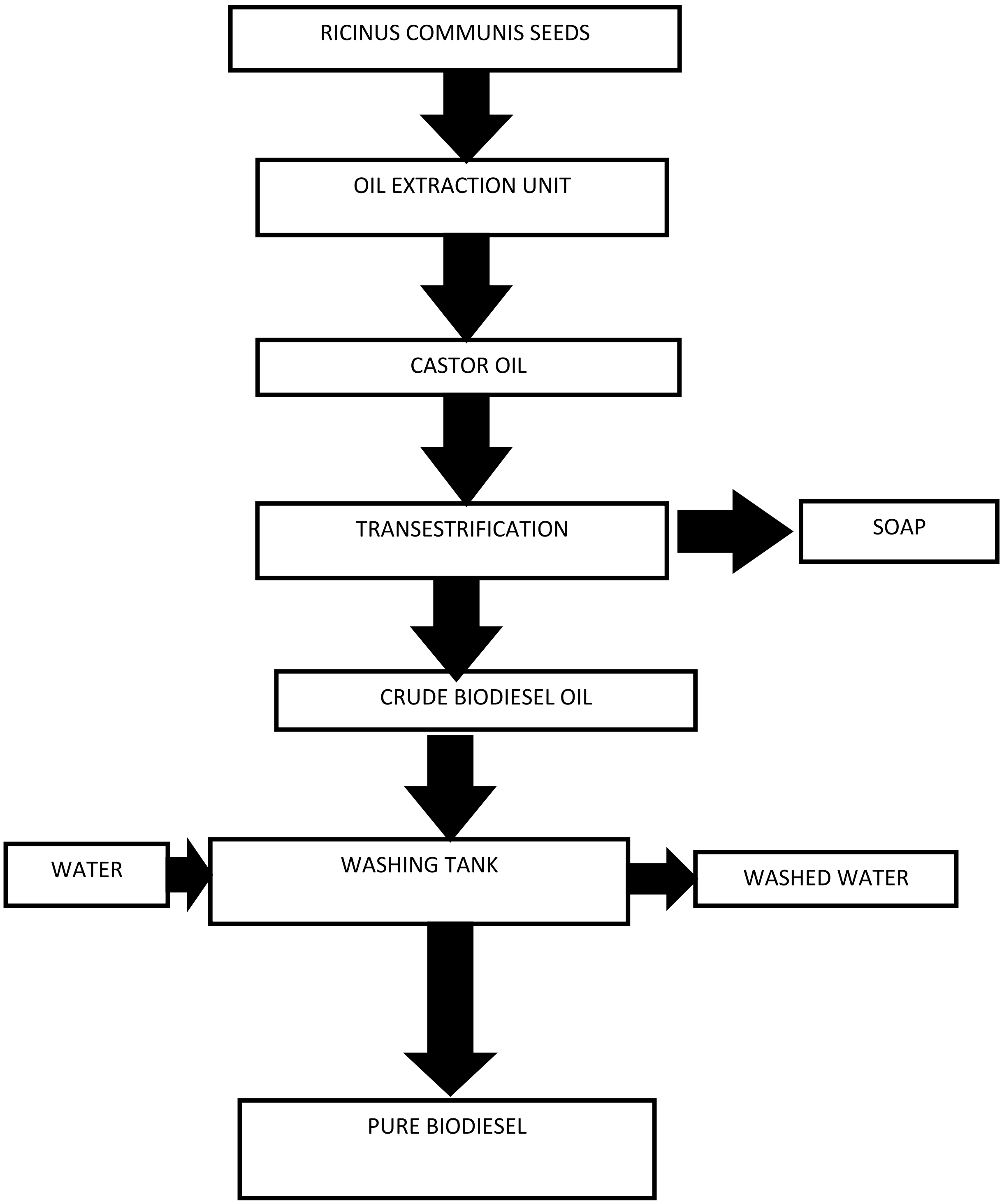

Fig 14: Flow chart of Biodiesel Production

5.9. BIOPHYSIOCHEMICAL TEST

1. Determination of oil content (Haque *et al.*, 2010)

The oil was extracted after the distillation of the sample and this percentage of oil was calculated from this formula.

$$\textbf{Percentage of oil content} = (w_2 - w_1) \times 100/w$$

Where, w = weight of sample, w_1= weight of the beaker with glass ball, w_2 = weight of the beaker with glass ball and oil, $w_2 - ws_1$ = weight of oil.

2. Determination of viscosity (Chandra *et al.*, 2013)

For viscosity measurement of the biodiesel sample, the viscometer was used using water as reference sample for interpreting the appropriate calculation. However the time was analyzed using the stop watch.

$$\eta 1 = \rho 1 \times t1 \times \eta 2 / t2$$

Where, η1=(viscosity of biodiesel),

ρ1=(density of b/d),

t1=(flow time of b/d),

η2=(viscosity of water) and,

t2=(flow time of water).

3. Determination of Specific Gravity (Akpan *et al.*,2014, Okechukwu and Anuforo, 2015)

Oil density estimation was done using a clean and dry density bottle (25ml). Weight of empty density bottle was taken and dried weight was taken to fill with distilled water. Bottle was dried weight was weighed was recorded after filling it with oil. While the specific gravity was estimated using the formula:

$$S.G = (W1-W0)/(W2-W0)$$

Mass of the substance / Mass of an equal volume of water.

Where,

W0 is the weight of empty density bottle

W1 is weight of the density bottle filled with oil

W2 is the weight of density bottle filled with water

4. Determination of saponification value (Akpan *et al.*, 2014)

In a conical flask 2g of sample was taken and followed by addition of 0.1N ethanolic potassium hydroxide and constantly stirred to boil gently for 60 min. A reflux condenser was placed on the flask containing the mixture. Few drops of phenolphthalein were added to the solution as an indicator and 0.5M HCL was added to the solution as a titrating agent until the light pink appearance was disappeared. The same procedure was used for other samples and blank.

The expression for saponification value is given by:

$$S.V = 56.1N\ (V0-V1)/M$$

Where,

V0 = the volume of the solution used for blank test;

VI = the volume of the solution used for determination;

N = Actual normality of the HCl used;

M = Mass of the sample.

5. Determination of Acid value (Nayak and Patel, 2010, Abdulkareem *et al*., 2012)

Two gram of the pure oil was weighed and poured in to the 250 mL conical flask. Neutral ethanol (20 mL) was pipetted in to the flask and heated on a steam bath for 3-min then the flask was allowed to cool down. The entire content was titrated with 0.1N alcoholic potassium hydroxide solution using indicator (phenolphthalein).A blank titration was also conducted side by side.

$$\text{Acid value} = 56.1 \times V \times N/W$$

V=volume of standard chemical used

N=normality of standard chemical

Weight of sample (g) = mg KOH/g

6. Determination of iodine value (Akpan *et al.*, 2014)

Conical flask was taken with 0.4 g of sample and to dissolve the oil we have used 20 ml of carbon tetra chloride in the sample. 25 ml of Wij's reagent was suspended in to the flask placed in to the fume chamber. The flask was sealed completely and the contents were swirled properly. Further, the flask was kept in the dark surrounding for 2.30 hrs. Finally, by using measuring cylinder we have added 20 ml of 10% aqueous potassium iodide and 125 ml of water. To perform the titration we used 0.1 M sodium thiosulphate solution until the yellow color was disappeared. Later, few drops of 1% starch indicator was added and the process continued by adding the thiosulphate drop wise until the blue color was disappeared. To avoid the biasing we have also perform the same procedure for blank and other samples.

The iodine value is given by the expression = 12.69C (V1-V2)/M

Where, C = Concentration of sodium thiosulphate used

V1 = Volume of sodium thiosulphate used for blank

V2 = Volume of sodium thiosulphate used for determination

M = Mass of the sample.

7. Determination of free fatty acid value

The amount of free fatty acid (FFA) was calculated as being equivalent to half the value of acid value, the formula is

FFA=Acid value/2=mg KOH/g

8. Determination of pH value (Akpan *et al.*, 2014)

Weigh the 2g sample of oil pour it into a clean and dried Flask .Add 13-15ml of hot distilled water in a beaker which contain the oil sample stirred it slowly. Allow it for Cooling. To measure the pH value standardized the pH electrode by using the buffer solution. After that electrode immersed into the sample and pH was recorded.

9. Determination of Flash point (Garba *et al.*, 2013)

Flash point is the minimum temperature at which the fuel or oil will ignite with the application of ignition source. In measuring the flash point a conical flask or beaker was taken and pour 2ml oil sample .Heated at low temperature at constant rate on a heat plate.

10. Determination of fire point (Shambhu *et al.*, 2013)

It is the point at which vapor of fuel catches fire and stays approx. to 5 sec. this point at which vapor rises is recorded.

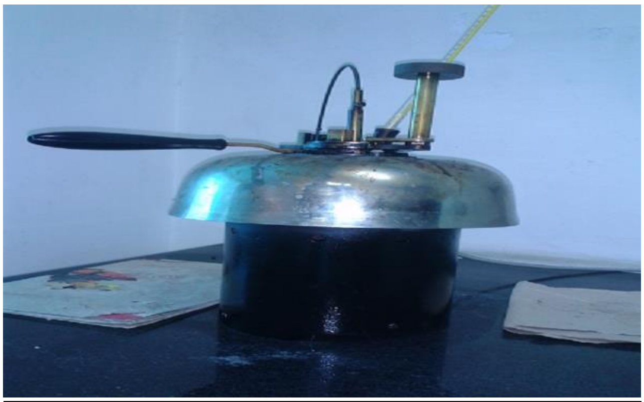

Fig 15: Pensky Martin apparatus used for the determination of fire and flash point.

Pensky Martin is a closed apparatus which was used for the flash point and fire point determination. In this sample of oil was allowed to tested was put into test cup and stirred it at slowly with constant rate simultaneously heating allowed.

Thermometer was attached at a side of apparatus which gives the temperature for flash and fire point as the temperature increases with the help of shutter flame was introduced for a moment. The sample placed in a cup catches fire was appeared and it stays for few seconds and the temperature at which the sample of oil catches fire is the fire point and a pooping like sound and lighting appeared which indicate the temperature of flash point **(Shambhu *et al.*, 2013).**

CHAPTER -6

RESULT AND DISCUSSION

RESULT AND DISCUSSION

Fig 16: Biodiesel produced from castor oil seeds

A(Gwalior), B(Nawasehar), C(Ghaziabad), D(Hyderabad), E(Sangrur).

6.1. Physical characterization of castor oil

The castor oil has generally pale yellow in color, high viscous, no odor and no taste. It is clear liquid at 27°C (room temperature).The different samples of crude oil shown in the figure (17) appears different in color such as dark brown, light brown, yellowish, pale yellow and yellow white.

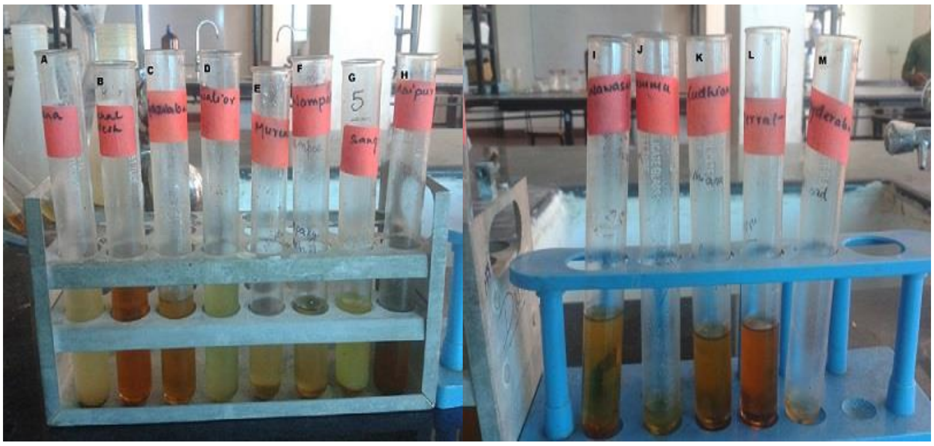

Fig 17:Showing the color of crude castor oil after distillation from different regions.A(Haryana),B(H.P),C(Ghaziabad),D(Gwalior),E(Morena),F(Nampalley),G(Sag-rur),H(Udaipur),I(Nawasehar),J(Jammu),K(Ludhiana),L(Merrut), and M(Hyderabad).

Table-4: Morphological characterization of castor seeds.

S.no	Sample	Colour, skin surface,kernel	Odor	Soil characteristics	Climatic conditions
1	Meerut	Dark brownish, rough, soft	Imperceptible	Sandy/loamy	Humid subtropical
2	Hyderabad	White/dark spots, smooth, soft	Imperceptible	Red soil	Wet and dry
3	Nampalley	Chocolate brown, smooth, hard	Imperceptible	Red and black	Semi-arid climate
4	H.P	Red/brown spot, rough, hard	Imperceptible	Forest/hill soil	Hot and sub humid
5	Morena	Dark brownish, smooth, soft	Imperceptible	Alluvial soil	Sub- tropical
6	Gwalior	Dust brown /black, rough, soft	Imperceptible	Alluvial soil	Sub-humid/hot
7	Ludhiana	Brown with black, rough, hard	Imperceptible	Deeploamy/alluvial	Extreme hot/cold
8	Ghaziabad	Dust-brown/black, rough, soft	Imperceptible	Sandy and loamy	Humid/subtropical
9	Sangrur	White and brown, smooth, hard	Imperceptible	Sandy and loamy	Extreme hot-cold
10	Bikaner	Light/brown/spot, rough ,soft	Imperceptible	Desert and alkaline	Hot
11	Jammu	Yellowish brown, smooth, soft	Imperceptible	Clay loam/ red clay	Semi –arid
12	Udaipur	Red/ brownish, smooth,	Imperceptible	Clay loam/ red	Semi –arid

		soft		clay	
13	Nawasehar	Dust/ black spots, rough, hard	Imperceptible	Sandy/clay loam	Extreme hot-cold

The above Table 4 shows the morphological characteristic of castor oil on the basis of seed color, shape, skin surface of seeds, and kernel. The soil characteristics into which the sample of different castor taken, the climatic condition favor the growth of the castor plant, seed ripening, etc.

Table-5: Percentage % of oil content and biodiesel produced after transesterification and pH value of sample of castor.

S.NO	Sample	pH	Oil Yield (%)	Biodiesel (gm)
1	Meerut (U.P)	6.80	57.6%	39.29
2	Ghaziabad (U.P)	6.98	43.7%	32.17
3	Udaipur (Rajasthan)	7.31	54.8%	36.25
4	Hyderabad	6.81	57.5%	46.57
5	Sangrur (Punjab)	6.43	39.7%	37.24
6	Ludhiana (Punjab)	6.78	39.2%	58.11
7	Nawasehar (Punjab)	6.19	72.0%	55.62
8	Haryana	7.33	74.2%	53.89
9	Jammu	6.58	43.3%	56.24
10	Morena (M.P)	6.32	23.9%	54.22
11	Nampalley	6.24	25.8%	42.16
12	Gwalior	6.83	12.6%	48.66
13	Himachal Pradesh	6.77	13.9%	38.16

*Above value is the mean of three replicates.

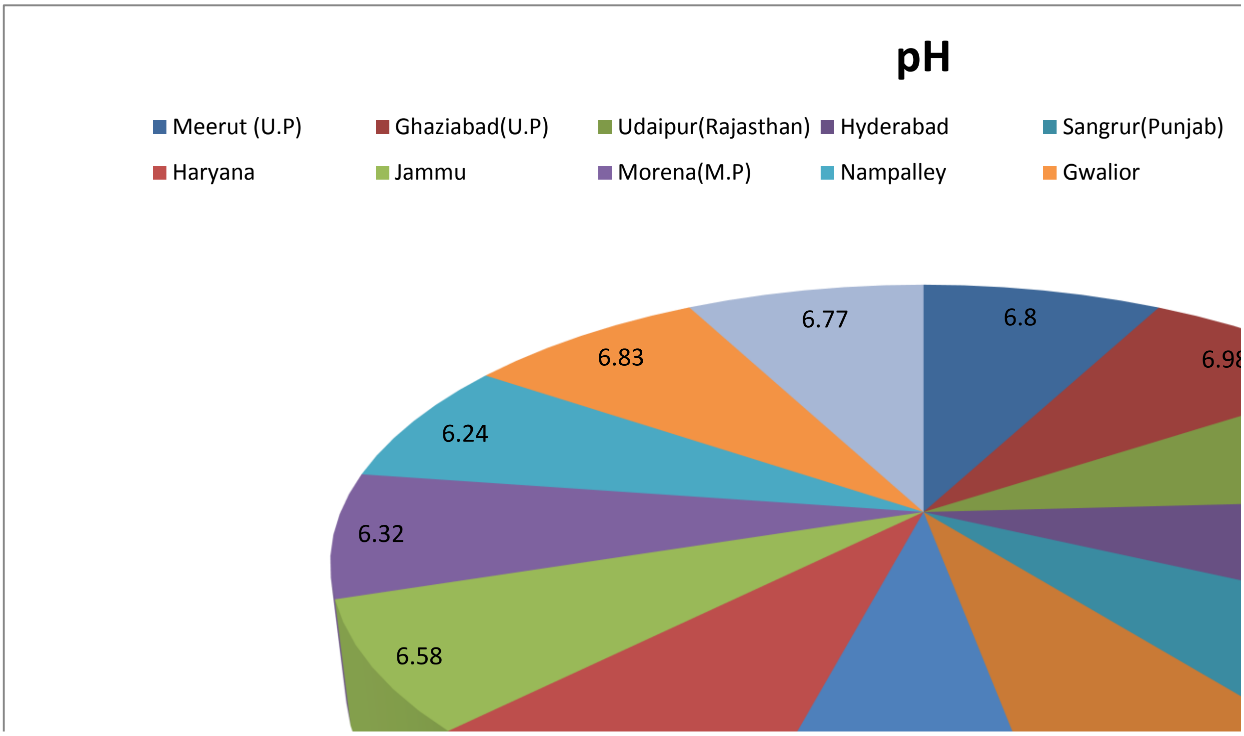

Fig 18: Recorded pH of castor oil samples from different regions.

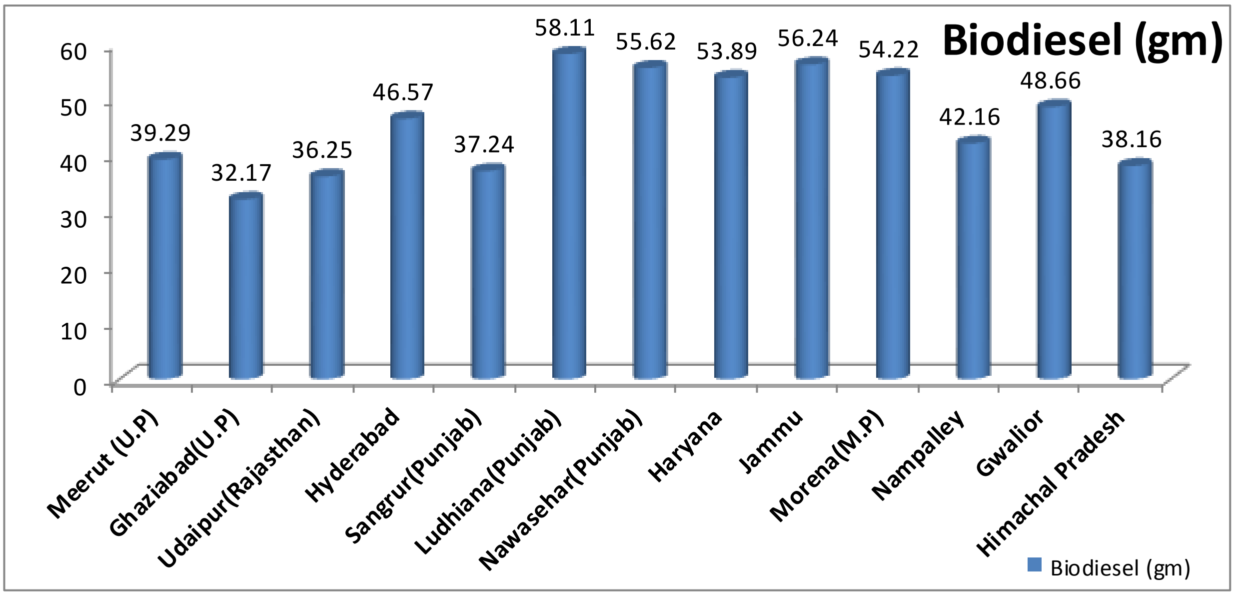

Fig 19: Biodiesel observation of biodiesel produced from different samples of castor.

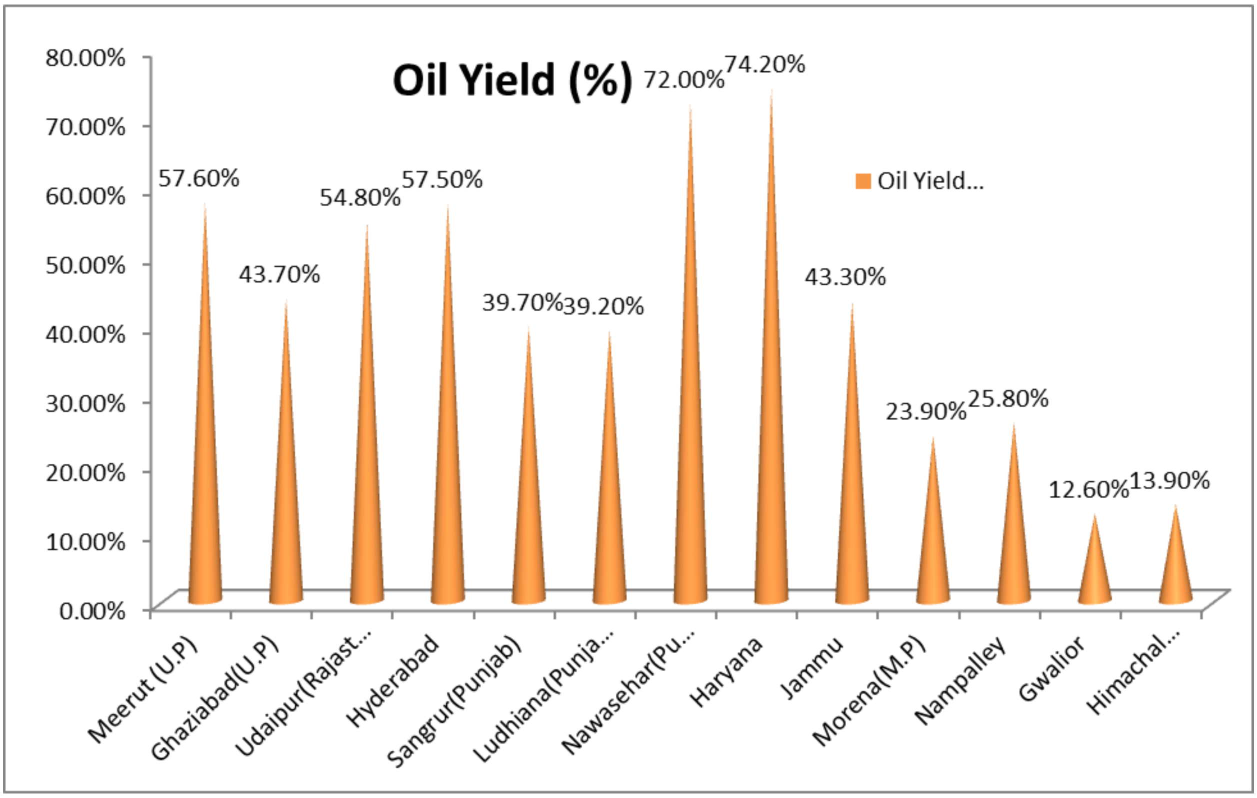

Fig 20: Bar graph showing the relationship between oil yield and geographical region of seed sample.

Result obtained from the Table (5) shows the variation of oil content in castor samples we have collected from different regions. The highest value of oil yield was found in the sample of castor seeds taken from Haryana having value 74.2%, followed by the Nawasehar sample 72.0% value whereas, seeds from Gwalior obtained lowest value of oil yield 12.6%. The value obtained from the sample of Meerut (57.6%), Hyderabad (57.5%) and Udaipur (54.8%). Similar values were reported by the **Jain and Suhane,(2012)**.These obtained values are lying between the values observed by **Okullo *et al.* (2012)** i.e., 56 ± 3.13 shows better results of oil extraction. The highest value of oil yield was observed here, may be the method of extraction which is an important parameter.

Oil content properties were dependent upon the locations and genotype of castor and environmental conditions, time of harvesting also affected the oil yield. High oil yield may be due to the environmental factors which enhance the quality and growth **(Nagbes *et al.*, 2013, Akpan *et al.*, 2014 and Abdelaziz *et al.*, 2014).**

The highest value of biodiesel was 58.11gm in the Ludhiana castor sample and the second highest value was observed from the Jammu sample (56.24gm) whereas, Nawasehar have biodiesel value 55.62gm and Gwalior have a moderate value 48.66gm. The lowest value of biodiesel recorded from castor crude oil was 32.17 recorded in Ghaziabad sample. For producing the biodiesel the crude oil of castor was allowed to transesterification process with the requirement of NaOH and methanol where, NaOH acts as a catalyst.

The pH value of castor oil was determined by the pH meter which has a pH electrode. The pH varies from regions to regions shown by the result from the above Table that the pH of the castor sample was approx. 6-7 range. 7.33, the highest pH value recorded in the Haryana castor sample and Udaipur has the value of 7.31, minimal pH value was optimized in the sample of Ghaziabad and Nawanshahr shows the value of 6.98 and 6.19. Approx. the same type of result of pH value analyzed by **Okechukwu *et al*.** (**2015)** in which value of crude oil and refined biodiesel was taken for analysis.

Same observations were reported by **Akpan *et al*.(2014)**. The present determination was falling in the same line determined by **Harbawy and Mallah, 2014.**Value below 7 indicate the acidic characteristics of oil the value of pH is affected during refining process done by neutralization of crude oil. Here, the pH value indicate an extraordinary properties utilized in soap making due to presence of equitable amount of FFA in the oil where it acts as a good indicator **(Nangbes *et al*., 2013).**

Table-6: Determination of different Biochemical characters

S. No.	Sample	Fire point (^{0}C)	Flash point (^{0}C)	Saponification value (mg KOH/g)	Iodine value (mg KOH/g)
1.	Haryana	120	100	241.23	82.55
2.	Nawasehar	100	90	274.89	59.69
3.	Jammu	150	130	173.91	69.85
4.	Morena	190	160	266.48	50.8
5.	Ghaziabad	110	90	218.79	80.01
6.	H.P	160	140	179.52	59.69
7.	Meerut	180	160	238.42	44.45
8.	Udaipur	160	120	196.35	97.79
9.	Gwalior	140	120	274.89	76.2
10.	Hyderabad	150	130	230.01	39.37
11.	Ludhiana	110	100	227.20	76.2
12.	Nampally	140	120	145.86	85.09
13.	Sangrur	110	100	140.25	46.99

*Above value is the mean of three replicates.

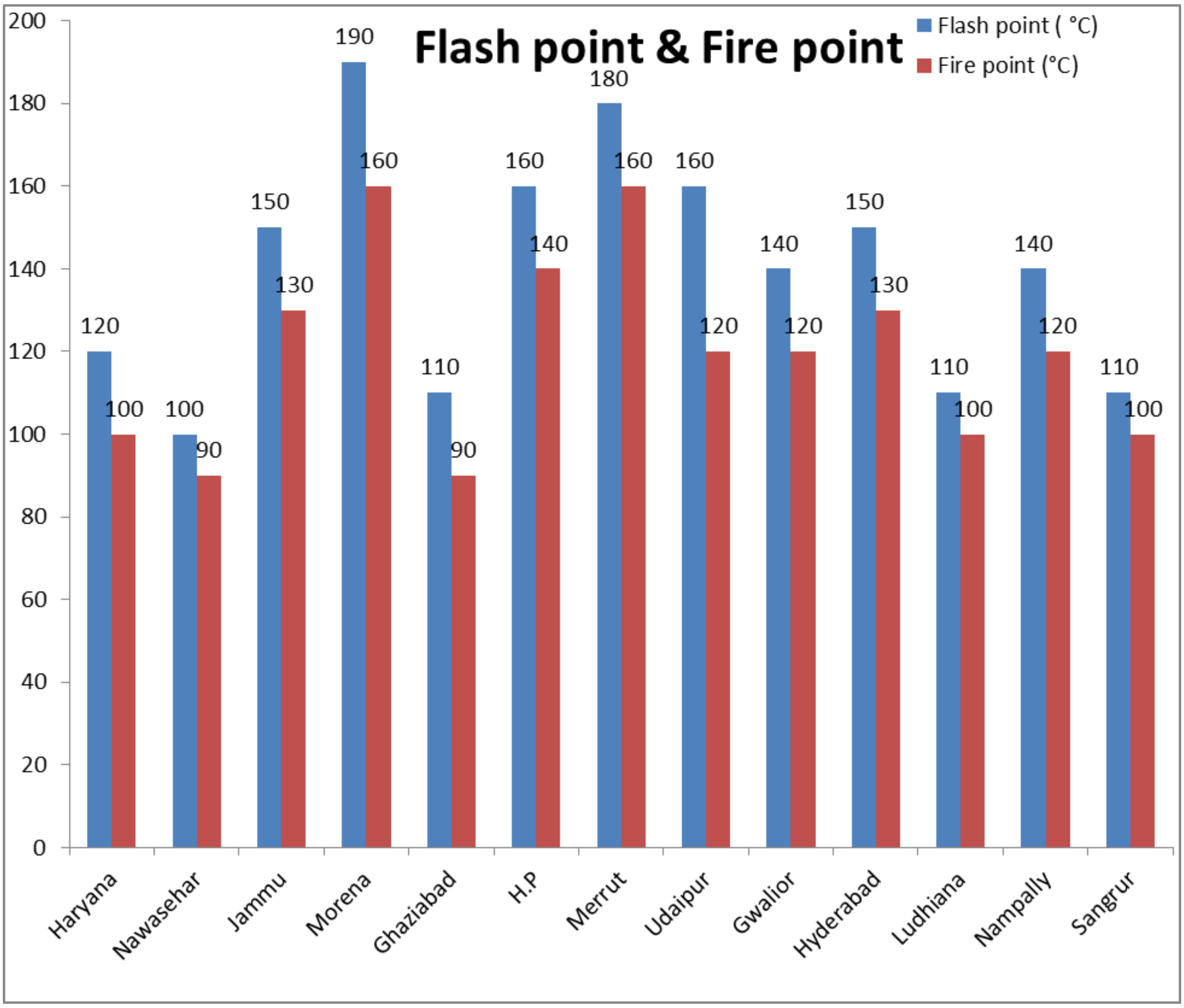

Fig 21: Bar graph showing the parameters of flash point and fire point respective with the geographical regions.

The flash point and fire point was determined by the Pensky-Marten apparatus which gives the popping sound and catch fire on the two different temperatures. The highest fire point was given by the Morena castor sample (190^{0}C) and Meerut (180^{0}C) whereas the Nawasehar sample gives the lowest valve. The highest flash point was given by the oil sample of Meerut and Morena having 160^{0}C. On the other hand the Ghaziabad and Nawasehar have the lowest value 90^{0} C. A moderate range of flash point was observed in Himachal Pradesh was 140^{0}C, Hyderabad was 130^{0}C.Same result for flash point reported by **Okechukwu *et al*.(2015)**. Whereas, nearby value of flash point observed by **Harbawy and Mallah, (2014)** and **Nagabes *et al*. (2013)**.

Fire point and flash point was giving a linear relationship with FFA, which was parameter which indicates the combustion .Castor having greater the flash point, the biodiesel obtained was not explosive , safe for handling and also non-hazardous **(Okechukwu *et al*., 2015, Nagabes *et al*., 2013)**.

Higher level of flash point decreases the threat of fire and this value help in storage as well as transportation **(Alnumi *et al*., 2014).** For the standard the diesel was tested which gives the fire at 90°C and flash at 70°C. The cotton oil gives fire at 130°C and flash at 110°C on the other hand the mobil oil give fire at 180°C and flash at 170°C.

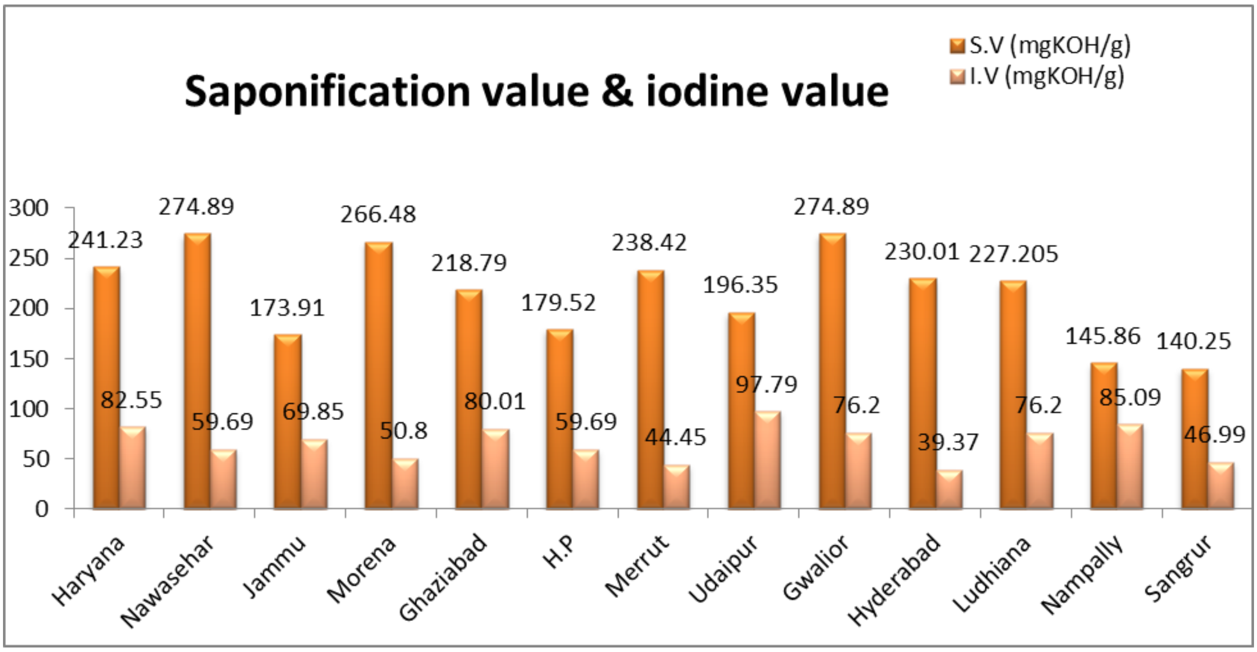

Fig 22: Bar graph showing comparative study of saponification value and iodine value regarding with geographical region.

The measure of the alkaline reactive group in oil and fatty acids is expressed in milligrams of potassium hydroxide which react with 1 gm of sample gives the saponification value. The value of saponification differs in different sample of castor. The highest saponification value was observed in Nawasehar and Gwalior 274.89 mg KOH/g which was similar to result reported by **Aldelaziz *et al.*(2014).** Whereas the value determined from the Hyderabad, Jammu and Himachal Pradesh sample was found to be followed the range estimated by ASTM (175-187) and approx. Value was estimated by **Akpan *et al.*(2014)**. 196.35mgKOH/g was the value obtained from the Udaipur castor oil which was same as

Reported by **Jain and Suhane, 2012.** The lowest saponification value was found in Sangrur sample of castor oil. The variation in the saponification value was due to the environmental conditions and the oil quality **(Bale *et al*., 2013).**

The highest iodine value was observed in Udaipur sample 97.79 mg KOH/g and the second highest value was observed in Nampalley sample 85.09 mg KOH/g whereas, the lowest value was reported in Hyderabad (39.37 mg KOH/g) and Meerut sample having 44.45mgKOH/g. The Iodine value observed by **Sreenivas *et al***.,**(2011)** was same to the presented work value existing in the range for iodine value shows better optimization.

Iodine value observed by **Harbawy and Mallah, (2014)** was same type whereas iodine value recorded by **Abdulkareem *et al*. (2012)** same to the result obtained in Nampalley sample, AOCS (Americian oil chemist's society) standard value for iodine in *Ricinus communis* was 83-88.The present work value follows the result observed by **Penugonda and Venkata, (2012)** whereas the I.V 58.64±0.71 by **Nagabes *et al*.(2013).** Iodine value defines to measure the unsaturation degree present in the oil sample due to its unique characteristics it should be used in the industries of cosmetics and soap making).proving a good raw material (**Abayeh et al., 1998).**

The iodine value measures the quantity of double bond present in the castor oil, which redirects the susceptibility of oil when exposure was allowed to oxidation. So due to this reason oil comes in the non-drying classes which known as liquid oil

In the industrial scale utilization of this oil was found to be used in hydraulic fluids ,coatings and in the manufacturing of lubricant and the value of iodine found high was due to presence of fatty acid in the oil in good concentration. Higher the value means higher unsaturation found in fats and oils **(Akpan *et al.*,2014, Ibiyemi *et al*., 1992 and Abitogun *et al*., 2008).**

Fig 23: Result after titration of saponification value.

Fig 24: Starch indicator is added which gives blue black color for iodine value determination.

Table-7: Determination of biochemical parameters of different castor sample

	Sample	Acid value(mg KOH/g)	FFA value(mg KOH/g)	Specific gravity (gm)	Viscosity (mm^2/s)
1.	Haryana	1.111	0.555	0.784	300
2.	Nawasehar	0.602	0.301	0.765	252
3.	Jammu	1.806	0.903	0.862	294
4.	Morena	3.520	1.760	0.874	190
5.	Ghaziabad	3.182	1.591	0.826	230
6.	H. P	2.838	1.419	0.648	246
7.	Meerut	1.548	0.774	1.240	274
8.	Udaipur	1.634	0.817	0.946	184
9.	Gwalior	1.462	0.731	0.724	230
10.	Hyderabad	0.860	0.430	0.987	290
11.	Ludhiana	1.290	0.645	0.886	282
12.	Nampally	1.118	0.559	0.432	264
13.	Sangrur	1.892	0.946	0.686	234

*Above value is the mean of three replicates.

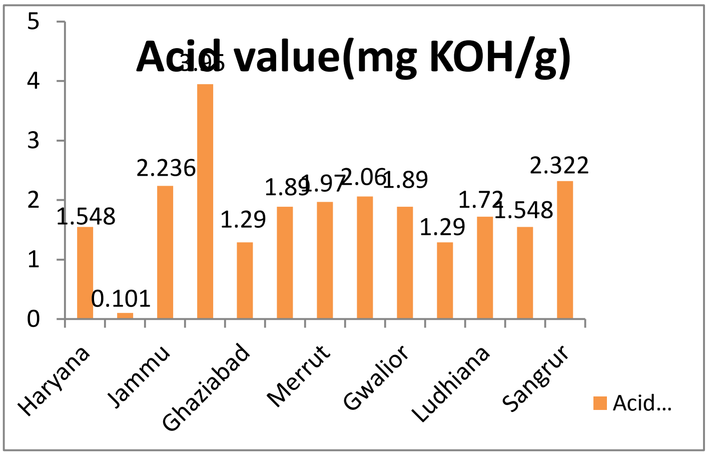

Fig 25: Graph showing the relationship between the acid value and geographical region of seed sample.

By observing the graph, the highest acid value was found 3.520 mg KOH/g in Morena sample and it was followed by Ghaziabad (3.182 mg KOH/g) on the other hand lowest value was obtained from a Nawasehar sample (0.602 mg KOH/g) the value observed from present work was same done by **Okullo *et al*., 2012** (2.41±0.03) A.V obtained by **Harbawy and Mallah, (2014)** was also giving same type of results. The ASTM standard range for acid value (0.4-4.0).

The acid value difference found in the different regions castor oil was due to quality of oil and some factors related to poor storage condition and immature seeds **(Salimon *et al*., 2010).** High acid value means large number of fatty acids present in the sample.

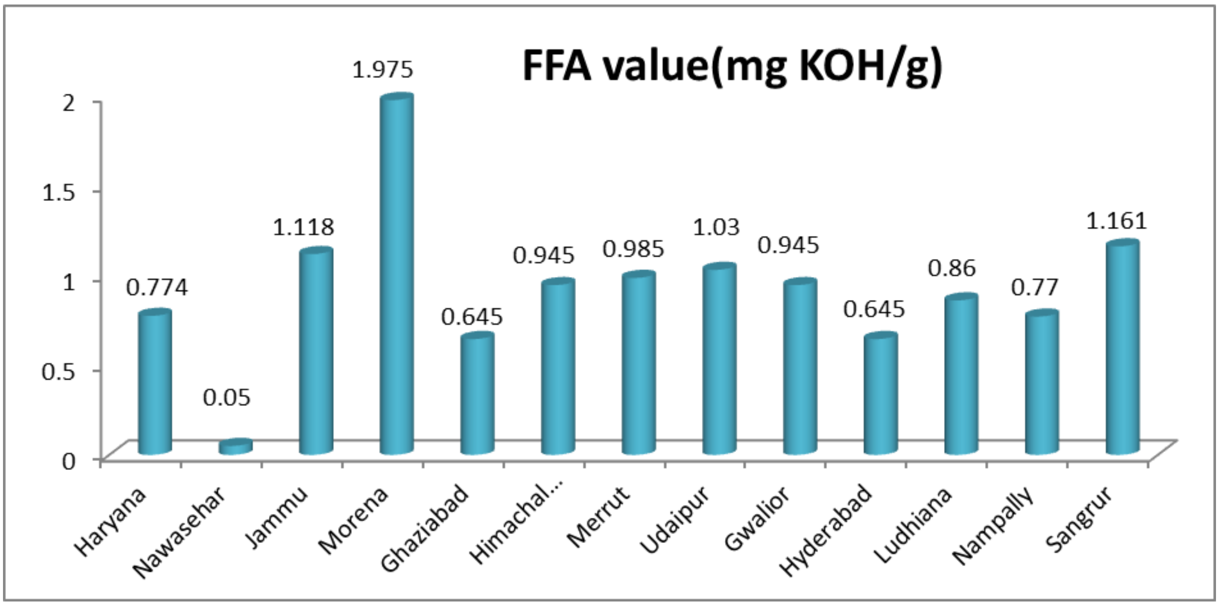

Fig 26: Showing the observation of FFA value by considering the different geographical regions

The FFA value observed from present work was 1.760 mg KOH/g highest value and lowest value was found in the Nawasehar (0.301mg KOH/g) moderate FFA value was observed in the Meerut sample 0.774 mg KOH/g. The similar observation of FFA value observed by **Harwary and Mallah , 2014** and **Okullo *et al*, 2012** and present work found similarity in FFA values reported in the above figure (26) the values of different samples fall in this range to check the oxidative deterioration by some chemical reactions and Enzymatic reactions.

High FFA value found in castor oil was due to the hydroxyl group.one of the reason for FFA value might be impurities present in the crude oil of castor. This is most important parameter for the energy industries and biodiesel properties also be influenced by the fatty acid analysis **(Nagabes *et al*., 2013).**

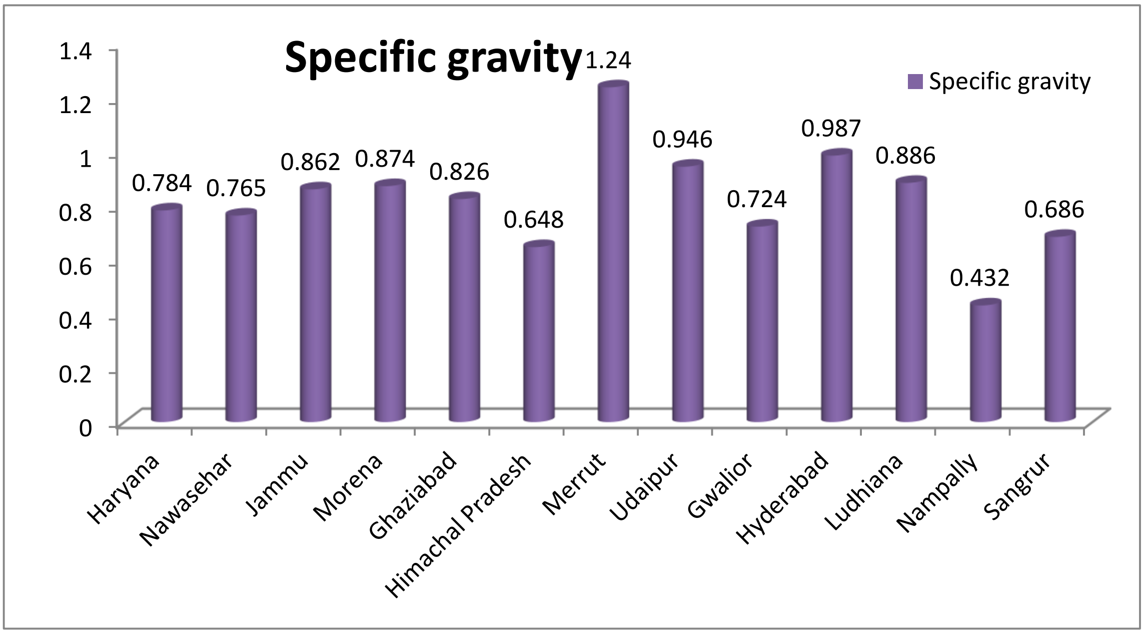

Fig 27: Showing the observed result of specific gravity and geographical region of seed sample

The result obtained by the above figure (27) I. e specific gravity was observed in which Meerut sample has the highest specific gravity 1.240 gm and the lowest value was observed in 0.432gm.The similar result was reported by **Nangbes *et al.* (2013).**

The moderate specific gravity was found in the Udaipur castor oil (0.946gm) the present result of specific gravity was same as observed by **Okechukwu *et al*. (2015) and Abdelaziz *et al*.(2014)** value was the ASTM follows the range 0.957-0.968.

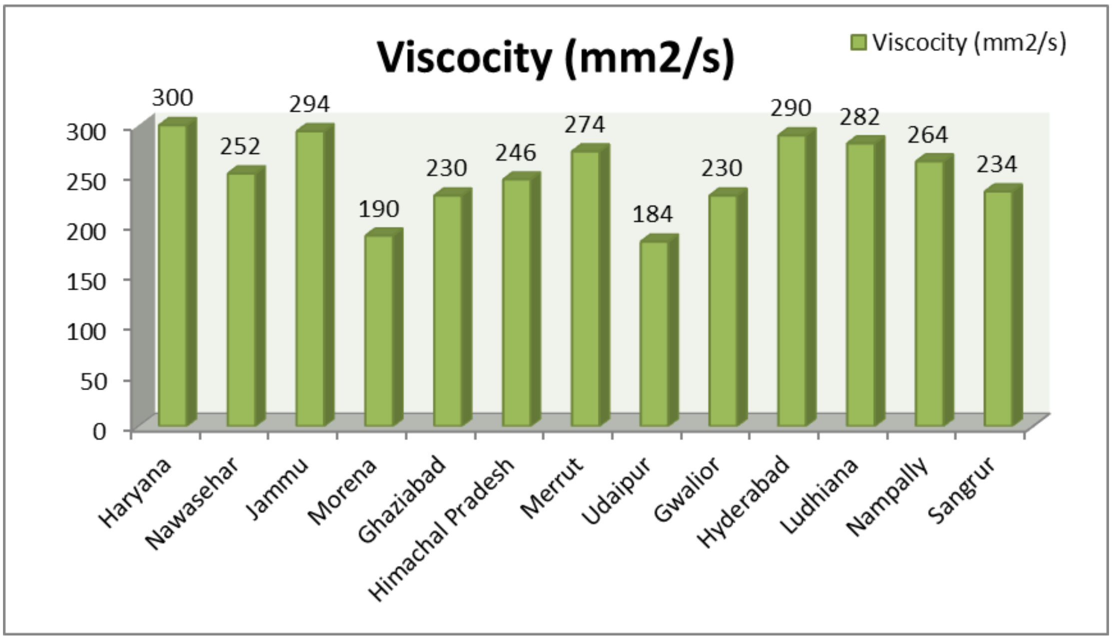

Fig 28: Showing viscosity of different samples taken from different regions.

The viscosity is a very important parameter for biodiesel. The value of viscosity in Haryana sample (300mm^2/s) the sample from Morena 294mm^2/s and the lowest viscosity was found in the Udaipur sample 184 mm^2/s. The viscosity value of the present work value lying between the range reported by **Okullo *et al*.(2012).** The viscosity was observed similar to the result obtained by **Sreenivas *et al*., 2011.**

The result of higher viscosity was due to presence of Hydroxyl bonding that form an H- bond in the castor fatty acid molecule **(Okullo *et al* ,2012).** The value which exceed from the standard stage in viscosity may be the reason of having suspended particles present in the crude oil sample of castor, extraction procedure with impurities (**Aldelaziz *et al.*, 2014).** The viscosity observed higher the values ASTM, due to the few impurities **(Nangbes *et al*., 2013).**

CHAPTER- 7

CONCLUSION

CONCLUSION

The castor oil was highly enriched with high oil content. This oil content % differs from region to region. Here, the sample of *Ricinus communis* seeds was taken to extract the biodiesel by using the soxhlet apparatus .To separate out the n- hexane and crude oil sample distillation was performed. Then the crude oil extract after distillation was transesterified by using the chemicals such as, sodium metal and methanol to form biodiesel.

The sample from Morena have flash point (160^0C) and fire point (190^0C). It also contains low viscosity 190 mm^2/s and specific gravity 0.74 mm^2/s but this sample contains the higher concentration of acid value and fatty acid value (3.520 and 1.760 mg KOH/g) higher flash point in this make the sample safe ,combustible and storage.

The oil sample from Meerut was considered best for the four parameters namely, flash point (180^0C), fire point (160^0C), oil yield (57.6%), and FFA i.e. free fatty acid (0.774 mg KOH/g). The utilization of non-edible crops prevents the nation from the environmental degradation, pollution and avoids the food shortage. The biodiesel obtained from castor is renewable cheap, availability of raw material for castor was locally available and it allows making a substitution for diesel fuel. Hence, can be used in cosmetics, transportation and pharmaceutical industries. More the biodiesel production from castor it enhances the growth of the country in the economic and industrialization sector.

The highest Saponification value was found in Nawasehar sample (274.89 mg KOH/g), this castor sample is excellent raw material for soap and cosmetic industries. The sample from Udaipur has highest iodine value (97.79 mg KOH/g) for this it is used in the industries for coating, hydraulic brake fluids and also in the production of lubricants. The highest free fatty acid value sample was used in making of oil on quantity of free fatty acid value.

CHAPTER- 8

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CHAPTER- 9

APPENDIX

APPENDIX

Chemical preparations

1) Starch preparation

In 10 ml of distilled water, 1 gm of starch was added and mixed well and then it was poured into 100 ml of boiling water and stirred and boiled for 1 min and leave to cool down.

2) 0.5 N alcoholic KOH

Firstly prepared 95% ethanol –

95 ml ethanol was added in 5 ml of distilled water. Then for preparing alc. KOH 2.8055 gm of KOH was mixed in 95% ethanol.

3) 0.5 N HCL

1.824 ml in 100 ml distilled water

4) 1.5 gm in 100 ml distilled water.

Abbreviations

GRASE - Generally recognized as safe as effective.

FAME - Fatty acid of methyl esters.

CLSER - Central laboratory of science, environmental and soil research

FDA - Food and Drug Administration

ASTM - American society for testing and material – provisional specification for biodiesel

CFPP - Cold filter plugging point

KOH - Potassium hydroxide

NaOH - Sodium hydroxide

H_2SO_4 - Sulphuric acid

FFA - Free fatty acid

I.V - Iodine value

S.V - Saponification value

A.V - Acid value

B20 - 20% biodiesel + 80% high speed diesel

B100 - 100 % biodiesel

B10 - 10% biodiesel + 90% high speed diesel

CaO - Calcium oxide

Ca $(OH)_2$ - Calcium oxide

NO_2 - Nitrogen oxide

SO_2- Sulphur oxide

CO-Carbon oxide

HCL- Hydrochloric acid

AOCS- American Oil Chemist's Society