

NUTRIENT COMPOSITION OF *Jatropha curcas* SEED OIL

BY

**Divine Oghenekevwe MAGEGE
AGR1608559**

**DEPARTMENT OF FOREST RESOURCES
AND WILDLIFE MANAGEMENT,
FACULTY OF AGRICULTURE
UNIVERSITY OF BENIN
BENIN CITY, NIGERIA**

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**A PROJECT SUBMITTED TO THE DEPARTMENT OF FOREST
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CERTIFICATION

This is to certify that this project work was carried out by **Divine Oghenekevwe MAGEGE** in the Department of Forest Resources and Wildlife Management, Faculty of Agriculture, University of Benin, Benin City, Edo State, Nigeria.

DR. A.A ERAKHRUMEN,
(Project Supervisor)

PROF. (MRS.) E.G. OBOHO
(Head of Department)

Date

Date

DEDICATION

Special dedication goes to God almighty for sparing my life all through my years of study in the University of Benin.

ACKNOWLEDGMENT

My profound gratitude goes to God Almighty for his grace and favour towards the accomplishment of this project work. My sincere appreciation goes to my project supervisor Dr. Erakhrumen who supervised my project work to ensure that the project was appropriate and successful. My special thanks goes to my H. O.D Prof. Mrs Oboho and other lecturers Prof. C. P Kalu, Prof. O.T Aremu, Prof. G.U Emelue, Prof. D.N Izekor, Prof. E.M Isikhuemen, Dr. A.A Erakhrumen, Dr. O.S Ikponmwonba, Dr C.I Aisagbonhi, Dr. Mrs N. Osadolor, Dr. Z. Dododawa, Dr. C Okwu-Abolo, Mr. C.E Ameh, Mr. F.E Osayimwen, Mr Y.I Egonmwan and Miss. N. Aigbobo for their support. I also thank Mr. I. Omorodion, Mr. S. Okwa, Mr. K. Ihama, and other members and staff of the Department of Forest Resources and Wildlife Management.

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ABSTRACT

The study evaluate nutrient composition of *Jatropha curcas* seed oil. Fully ripened *Jatropha curcas* fruits were collected from *Jatropha* plantation closed to Wildlife Ecotourism proposed site, Faculty of Agriculture, University of Benin, Benin City. Samples were collected from three parts of the plantation. The seeds were extracted from the fruits and air-dried accordingly. The dried seeds were taken to Faculty of Agriculture, University of Benin, Benin City main laboratory. The seeds were dehusked and grinded using ceramic mortar and pestle. The oil was extracted using a 500ml soxhler extraction apparatus using N-Hexane. Excess N-Hexane were recovered after removing the extracted sample from the extractor and oil parameters determined according to AOAC official method. Data collected were subjected to inferential statistics using analysis of variance (ANOVA) at 5% level of significance. The results showed that moisture and crude protein content of the three samples of *Jatropha curcas* seed oils were significantly difference ($P < 0.05$). The moisture content ranged between 0.08 ± 0.001 to 0.10 ± 0.002 with an average of $0.09 \pm 0.01\%$. The crude protein content ranged between 17.50 ± 1.7 to 17.80 ± 1.5 with an average of 17.70 ± 1.5 . There was no significant difference ($P > 0.05$) in %fat among the samples. The %fat ranges between 39.4 ± 2.9 to 40.2 ± 2.2 , with an average of 39.8 ± 2.7 . The results revealed significant difference ($P < 0.05$) in acid values, saponification, and free fatty acid among the three samples of *J. curcas* seed oil, while the values of Iodine and peroxide among the three samples showed no significant difference ($P > 0.05$). The average acid values, Iodine, saponification, peroxide and free fatty acid of *Jatropha curcas* seed oil were 1.20 ± 0.3 , 82.4 ± 3.6 , 166.0 ± 4.9 , 1.93 ± 0.1 , and 0.40 ± 0.02 respectively. In conclusion, the proximate composition studied revealed that the *J. curcas* seed oil is a good source of protein and has high fat content. This makes the

seed a valuable dietary supplement which can help reduce the problem of protein energy malnutrition.

CHAPTER ONE

1.0

INTRODUCTION

1.1 Background of the Study

Jatropha curcas L. (Euphorbiaceae) is a multipurpose perennial shrub/small tree, native to Mexico and Subtropical America, now grows naturally in most tropical areas of the world. It is an underutilized plant of multiple values. Various parts of the *J. curcas* are globally used for healthcare management of plants, human being and domesticated animals. Besides ethnomedicinal usages, this species have much other ethnobotanical, economic and ecological importance. Present review deals with origin and distribution, taxonomic description, ethnobotany, pharmacological activities, phytochemical properties and future prospective of this species.

J. curcas has a high allelopathic effect in the germination and root growth of some cultivated species, essentially with the extract of leaf and pericarp (Saadaoui *et al.*, 2015b). The major attributes proclaimed for the wide scale adoption of *J. curcas* in developing countries were its biodiesel production potential, higher oil content than other biofuel crops, rapid growth, easy propagation, drought tolerant nature, ability to grow and reclaim various kinds of land, plants can grow without much irrigation and agricultural inputs, pest resistance, small gestation periods and suitable traits for easy harvesting (Edrisi *et al.*, 2015). *J. curcas* has medicinal interests; it has anti-malarial, insecticidal, anti-cancerous and anti-tumor properties.

Jatropha curcas L. or its common name, physic nut, is from the family Euphorbiaceae. It is drought-resistant plant which grows well in marginal land of low to high rainfall areas and can be used as a commercial crop. *Jatropha* is easy to propagate and can

easily survive with minimum care. It grows wild in many areas and even survives in infertile soil. The plant is easy to grow and produces seed for 50 years life span.

The maximum oil content that has been reported for jatropha seeds has been close to 47%. However, the accepted average is 37-40%. Hence, it can be the best plant for future biodiesel production (Jongschaap *et al.*, 2007). Besides using as a biodiesel feedstock, jatropha can also be used for saponification of oil of making soap, cleaning, dye for clothes, medicinal purposes, and organic fertilizer and also as antidotes for snake bites. Jatropha oil can be used to soften leather and lubricate machinery. Each part of jatropha plant, its leaves, flowers, fruits, and seeds has its own uses which make it a multi-beneficial plant (Guitz, *et al.*, 1999 Felix and Clement, 2011). Jatropha oil is a vegetable oil which is produced from *Jatropha curcas* Linnaeus (*Jatropha curcas* L.) seed. Due to the toxicity of jatropha seeds, the plant is cultivated for the sake of extracting the oil as it cannot be ingested by humans. Jatropha oil can be used as biodiesel feedstock where biodiesel means diesel fuel that is extracted from oil and fats of plants (Omotoso, *et al.*, 2011). Biodiesel is a renewable form of energy which is similar to conventional diesel fuel. Besides, this energy is environmental friendly as jatropha oil has very low emission and it is also easily produced in rural areas.

An optimum extraction condition which is also cost effective is required to increase the yield of the oil. The research is also to determine the constituents of the oil obtained followed by the applicability and commercialization of the oil.

1.2 Statement of Problems

Recently, jatropha has been recognized as the major source of biodiesel production as it has the highest percentage of oil content which comes to an average of 37 to 40%. It has been a marked substitute in the fuel industries for automotive diesel of

locomotives (Omotoso, *et al.*, 2011). According to the current ratio, the price of jatropha seeds is much lower than the oil price. But as the demand of jatropha oil as biofuel increases, the cost will increase too with consideration of jatropha plantation and the product price. Besides, the cost also depends on the quality of the oil extracted from the seed. The increasing demand of jatropha oil also has opened up wide opportunities for global marketing which leads to the requirement of competitive products which comes in advantage in term of quality, cost and production time (Openshaw, 2000). Hence, it is best to identify a best extraction technique, as to extract higher yield of oil with higher quality at lower cost.

Other than that, this research is also conducted to improve existing extraction process toward more environmental friendly method. The existing process makes use of n-hexane, a solvent obtained from petrochemical sources which will be emitted during extraction and recovery process. Hexane is a volatile organic compound which can cause air pollution when it reacts with ozone in the atmosphere. But still, many existing processes are using n-hexane because it is lower in cost and gives good yield of oil recovery from the plant (Suzanna, *et al.*, 2003). Hence, it is necessary to come up with an alternative organic extraction solvent which gives approximately the same yield as n-hexane which is lower in cost and also environmental friendly.

Many researches have been conducted before using jatropha but still room for enhancement can be made.

1.3 Objective of the study

The main objective of this study is to chemically and nutritionally evaluate the value of seed oil from *Jatropha curcas* harvested from a plot of land in University of Benin, Benin City, Nigeria.

The specific objectives are to;

1. quantify some available chemicals in the *Jatropha curcas* seed oil.
2. evaluate selected nutrients available in the extracted Jatropha seed oil.
3. establish if variations or differences exists in the values obtained for chemical and nutritional assessments for *Jatropha curcas* oil.

1.4 Scope of Study

The scope of this research is to find the yield of jatropha oil extracted and also the importance of the oil. And to evaluate the selected nutrients available in the extracted jatropha seed oil and the oil extracted from jatropha seed oil be analyzed for its constituents.

1.5 Justification of the Study

The result of this research would signify the identification of the optimum extraction parameters for the production of jatropha oil from its seed to fulfill the high demand of jatropha oil as biofuel, feedstock and also for other purposes such as medicinal use, soap making, and as natural hedge. Besides, through this research an environmentally friendly method will be identified for the production of jatropha oil.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 An Introduction of *Jatropha curcas* Linnaeus

Jatropha is a genus of over 170 plants from the Euphorbiaceae family commonly found and utilized across most of the tropical and subtropical regions of the world. Among the different species of *Jatropha*, *Jatropha curcas* L. has a wide range of uses and promises various significant benefits to human and industry. It has a yield per hectare of more than four times that of soybean and ten times of corn (Sepidar, *et al.*, 2009, Nobrega & Sinha, 2007). Taxonomy of *Jatropha curcas* L. is given in Table 2.1

Table 2.1 Taxonomy of *Jatropha curcas* Linnaeus Taxonomy

TAXONOMY

Kingdom	Plantae
Division	Embryophyta
Class	Spermatopsida
Order	Malpighiales
Family	Euphorbiaceae
Genus	<i>Jatropha</i>
Species	<i>J. curcas</i>
Scientific name	<i>Jatropha curcas</i> Linnaeus
Common name	Physic nut

Jatropha curcas L. is a very adaptable, perennial plant, which lives for more than two years and can be easily grown on hard soil. *Jatropha* plant is regarded as a shrub or small tree as its height generally ranges from 3-5 meter. It can grow in arid condition, on any kind of ground and does not require irrigation. Therefore, it can be easily cultivated in marginal land (Felix & Clement, 2011).

Growth of jatropha plant occurs during the rainy season. Flowering usually occurs during rainfall and seed will be produced at the end of the rainy season, usually in the first or second year of growth (Brittania & Litaladio, 2010). The branches of the plant contain latex, a milky substance that hardens once out in the open air. The leaves are smooth and 10 to 15 cm in length and width. The seeds have thin shells in black colour and oblong shape. Picture of jatropha fruit and seed are given in Figure 2.1. The matured seeds are usually 2.5cm long and can be easily crushed to extract oil from them (Jongschaap, *et al.*, 2007).

Jatropha is believed to have been reach to countries in Africa and Asia by Portuguese seafarers from its centre of origin in Central America and Mexico via Cape Verde and Guinea Bissau. In Madagascar, Cape Verde and Benin, jatropha oil was used as mineral diesel substitute during the Second World War (Akbar, *et al.*, 2009, Agarwal, 2007).

The genus jatropha has 426 species and distributed throughout the world. *Jatropha curcas* L. was first described in 1753 by Carl Linnaeus, Swedish botanist. The name of this plant was derived from Greek where the first part of its name, ‘jatos’, meaning physician and the last part, ‘trophe’, meaning nutrition (Brittania & Litaladio, 2010).

Description

Jatrophas are oil plants belonging to the Euphorbiaceae family. The *Jatropha* genus includes more than 175 species, among which *Jatropha curcas*L. and *Jatropha platyphylla* Müll. Arg. have drawn particular attention as potential biofuel and animal feed sources. *Jatropha curcas* has both toxic and non-toxic genotypes while *Jatropha platyphylla* is a non-toxic species (Makkar *et al.*, 2012).

The jatropha plant

Jatrophas are drought-resistant perennial and multipurpose shrubs or trees, similar to the cassava plant (Elbehri *et al.*, 2013). *Jatropha* is a deciduous tree, shedding its leaves during the dry season. It can grow to a height of 3-5 m, and remains productive for 30 to 50 years. *Jatropha* has a deep taproot and four shallow lateral roots. The taproot anchors the plant in the soil, stabilizing the soil against landslides, and the lateral roots prevent soil erosion. The trunk is covered with a smooth grey bark that exudes watery and sticky latex when cut. The leaves are smooth, 4-6 lobed, 10-15 cm long and wide, and are usually pale green in colour (Raheman, 2012). Inflorescences develop at the apex of the branches and bear approximately 10 or more ovoid fruits (pods) (Makkar *et al.*, 2012). Flowers and fruits develop during the rainy season or year-round in permanently humid regions. The pods contain many elliptic seeds and become yellow when they mature. Dry *Jatropha* fruits contain about 38% husks and 62% seeds. The seeds look like castor seeds in shape, and are black in colour (Raheman, 2012). They consist of 30-40% testa (shells) and 60-70% kernels. The kernels contain 44-62% oil (King *et al.*, 2009).

Most *Jatropha* species, including *Jatropha curcas*, contain numerous toxic components and the plant and its by-products, when not detoxified, are deleterious to humans and livestock. One species, *Jatropha platyphylla* is non-toxic and is eaten by indigenous people in the Sinaloa State of Mexico, who call it "sangregrado". This species has thick succulent branches and its leaves, borne on long petioles, are glabrous and larger (25-35 cm) than those of *Jatropha curcas*. Its seeds are almost circular and contain 50-60% edible oil (Makkar *et al.*, 2011).

Jatropha utilisation

Jatrophas are multipurpose plants. The name jatropha was derived from the Ancient Greek words *iatros*(doctor) and *trophos* (feed) because of its many potential medicinal applications (Elbehri *et al.*, 2013).

- **Jatropha seed oil** is used to make soap, lubricants, varnish, insecticides or medicines (Raheman, 2012). It has good feedstock qualities for biodiesel production. Jatropha biodiesel meets European Union (EN14214) and North American standards (ASTM D6751) (Makkar *et al.*, 2012).

Jatropha oil cake (oil kernel meal) results from the oil extraction of decorticated seeds. It can be used as fertilizer or as feedstock in biogas production. Jatropha kernel meal is a protein-rich product, but only kernel meal obtained from non-toxic genotypes of *Jatropha curcas*, from the non-toxic species *Jatropha platyphylla*, or detoxified kernel meal from toxic genotypes of *Jatropha curcas* can be safely used as feed resources (Makkar *et al.*, 2012). Jatropha seed meal is obtained after the extraction of non-dehulled seeds.

- **Jatropha plants** are used to make fences and shelter to protect from animals, wind or erosion (Elbehri *et al.*, 2013).
- **Jatropha roots** yield an oil that has anthelmintic properties.
- **Jatropha leaves** are used as feed in the rearing of silkworms, and in human nutrition as a vegetable for their antimicrobial and anti-inflammatory properties (Makkar *et al.*, 2009).
- **Whole seeds** of the non-toxic genotype are sometimes boiled or roasted and eaten as a snack by humans.
- **Jatropha latex** is used as dye or as a pesticide/molluscicide.
- **The fruit hulls** can be used as green manure or to produce biogas.

Distribution

Jatropha is thought to have originated from tropical America (Mexico). It could have been introduced to other regions from the Caribbean by Portuguese sailors travelling to Cape Verde and Guinea Bissau. It is now available in many arid, semiarid, tropical and subtropical regions of the world as it is able to thrive in many climatic zones with a wide range of rainfall (200 to 1200 mm annual rainfall) (Raheman, 2012). Jatropha is well adapted to arid and semiarid conditions. It can grow on marginal lands with limited water and agrochemical supply but will not yield enough seeds in such conditions (Elbehri *et al.*, 2013; Raheman, 2012; Brittain *et al.*, 2010). Jatropha does better in areas with 1000-1500 mm annual rainfall, temperatures of 20-28°C with no frost, and where the soils are free-draining sands and loams with no risk of waterlogging (Elbehri *et al.*, 2013; Raheman, 2012). However, it grows on almost any type of soil including gravelly, sandy, stony or rocky soils and in the crevices of rocks where soil depth and fertility are very low. It responds positively to fertilizer or manure with added calcium, magnesium and sulfur. For economic returns, a soil with moderate fertility is preferred (Raheman, 2012).

Processes

After harvesting, jatropha fruits are dehusked and the seeds are sun-dried and then shelled. The kernels are pressed for oil extraction, resulting in a protein-rich jatropha kernel meal.

Forage management

Jatropha is a fast-growing plant that can be propagated either by seedlings (generative method) or direct planting of cuttings (vegetative method) (Raheman, 2012).

Propagation

Jatropha curcas can easily be propagated by both seed or cuttings. Some people recommend propagation by seed for establishment of long-lived plantations.(Duong T.H. (2013)

When *jatropha* plants develop from cuttings, they produce many branches but yield fewer seeds and do not have enough time to develop their taproot, which makes them sensitive to wind erosion.Reubens *et al.*, 2011; Henning, 2009).

The seeds exhibit orthodox storage behaviour and under normal treatment and storage will maintain viability at high percentages for eight months to a year.(Heller, Joachim (1996), Duong T.H. (2013)

Propagation through seed (sexual propagation) leads to a lot of genetic variability in terms of growth, biomass, seed yield and oil content. Clonal techniques can help in overcoming these problems. Vegetative propagation has been achieved by stem cuttings, grafting, budding as well as by air layering techniques. Cuttings should be taken preferably from juvenile plants and treated with 200 micro gram per litre of IBA(rooting hormone) to ensure the highest level of rooting in stem cuttings. Gadekar Kumarsukhadeo Prakash. (2006 Cuttings strike root easily stuck in the ground without use of hormones.James A. Duke. 1983.

Cultivation

Jatropha cuttings can be planted directly in the field, in nursery beds, or in polyethylene bags, during the rainy season, at 10-20 cm depth in the soil. When *jatropha* plants develop from cuttings, they produce many branches but yield fewer seeds and have not enough time to develop their taproot, which makes them

sensitive to wind erosion. Presoaked seeds (24 h in cold water), directly sown in wet soil or in seed beds under nursery conditions at the beginning of the rainy season are advisable for seed production. *Jatropha* can be easily intercropped with shade-tolerant plants, legume seeds such as black gram (*Vigna mungo*), and vegetables, such as tomato, bitter melon (*Momordica charantia*), pumpkin, and cucumber can be profitably grown under *jatropha* plants for the first two years. Irrigation and fertilizers are beneficial during the establishment period. After establishment, irrigation is not necessary but fertilizer should be applied every year during the productive period. Pruning should be done to promote branching and subsequent fruit yield (Raheman, 2012).

Harvest and yields

Depending on climatic conditions, *jatropha* may be harvested twice in a year under semiarid conditions, or continuously in more humid areas. Fruits have to be handpicked and harvest is time-consuming. *Jatropha* seed yields are variable, depending on growing conditions: in degraded land with low inputs, yields were reported to range from 1 to 2.5 t/ha/year; in fertile soils with high inputs *jatropha* could produce 2 to 5 t/ha/year (Elbehri *et al.*, 2013). It has been noted that even on adequately fertile soils, *jatropha* is no match for sunflower and that the hugely variable yields have contributed to its decline in the early years of the present century (Promode Kant *et al.*, 2011).

Environmental impact

Soil erosion control and wind shelter

Jatrophas, because of their taproot and lateral roots, can play two important roles in soil preservation. Lateral roots protect soil from heavy rains and provide soil

cohesion, decreasing soil erodibility (Reubens *et al.*, 2011; Henning, 2009). The deep taproot extracts subsurface soil moisture and enhances vegetative cover or allows intercropping, even in very dry environments. Planting jatropha in lines to form contour bunds improves rainwater infiltration. Jatropha hedges reduce wind erosion by reducing wind velocity and by binding the soil with their surface lateral roots (Henning, 2009).

Live fence

Jatropha is used as a living fence in many tropical and subtropical countries. Jatropha leaves are not eaten by livestock: goats would die of starvation rather than browsing jatrophas. When jatropha is intended for live fences, jatropha cuttings are planted as a hedge to protect homes, gardens and fields from wandering animals. Hedges planted very close together (5 cm) form a barrier that is impenetrable even by chickens (Henning, 2009).

Application of *Jatropha curcas* Linnaeus Oil

Jatropha oil has been found for using in many different fields for various applications around the world. There are some chemical compounds in the seed of jatropha which possesses poisonous and purgative elements and renders it from being edible to human. Therefore, jatropha oil has been commercialized in other field for non edible purposes. Some of the applications of jatropha oil which have been discussed by Felix & Clement (2011) and Parajuli (2009) have been listed below.

• Source of Biodiesel

The extracted oil of jatropha is used as transesterified oil or biodiesel.

Biodiesel is a clean and renewable energy replacing the diesel fuel. It has some advantages: it is non-flammable, non-explosive, biodegradable, and has significantly lower toxicity.

- **Hedge**

Jatropha plant was commonly used as hedges or living fences to protect valuable plants from eaten by animals as it produces latex and is toxic. Besides, they also provide shade for other plants while the dropped leaves will decompose and become fertilizer.

- **Fertilizer and insecticide**

The residue from the oil extraction, pressed seed cake, is rich in nitrogen, phosphorous, potassium and more fertilizing nutrients. Besides this, they also have insecticidal properties which can reduce amount of nematodes in soil.

- **Medicinal Uses**

Tannins and latex extracted from the bark has anti microbial properties and astringent properties each. Extracts from *Jatropha curcas* L. have been shown to have anti-tumor activity, the leaves as a remedy for malaria and high fever, the seeds for treatment of constipation and the sap in accelerating wound healing procedure.

- **Manufacturing**

Jatropha oil gives a good foaming; white soap with positive effect on the skin is due to the glycerin content of the soap. It is also useful for manufacture of candles and cosmetic industry. The saponification of *jatropha* oil indicates the amount of sodium hydroxide necessary to make a solid soap. In China, it is used to make varnish while

in England; it is used for wool spinning. The protein content of jatropha oil can be used as raw material for plastics and synthetic fibers.

2.3 Extraction of Jatropha Oil from *Jatropha curcas* Linnaeus seed

The extraction process can be classified based on combination of phases (solid, liquid, gas, supercritical fluid). For solid – liquid, this extraction is useful for the isolation and purification of naturally occurring sources while liquid – liquid is a more common method depending on solubility properties of components.

Various solvent are used for extraction such as organic solvents and inorganic solvents where, organic solvents are less dense than water while inorganic solvents are denser than water. Commonly used organic solvents are diethyl ether, toluene, hexane, ethyl acetate, ethanol, and inorganic solvents are dichloromethane, chloroform and carbon tetrachloride.

Other uses

Jatropha oil is also used to soften leather and lubricate machinery (e.g. chain saws). If seed cake is available in large quantities, it can also be used as a fuel for steam turbines to generate electricity. Apart from the bio-diesel application, the oil finds the application in cosmetic industries, for the manufacture of candles and soap. The extraction of the biodiesel after transesterification of the seed oil, leads to two main by-products that is glycerol and press or oil cake. Glycerol has many useful industrial applications as a raw material for the synthesis of 1, 3 propane-diol and other polymeric materials (Sharma, 2008). The bark of jatropha contains tannin. It can also have the honey production potential as the flowers can attract bees.

CHAPTER THREE

MATERIALS AND METHOD

3.1 Study Area

The study were carried out in the Department of Forest Resources and Wildlife Management; *Jatropha* plot, Ecotourism unit, Faculty of Agriculture, University of Benin, Benin City. The plot is situated between latitude 6.40N° and longitude 5.62E°. University of Benin is located in the rain forest zone of southern Nigeria. The mean annual rainfall ranges from 1500mm to 2000mm; mean relative humidity is about 75% at noon and about 95% at 6am while minimum and maximum temperature are between 27°C and 32°C respectively (UNIBEN master plan, 1993). The altitude is 74.5m above sea level, the northern part of the campus is drained by Ikpoba River (UNIBEN master plan, 1993).

3.2 Experimental Procedures

3.2.1 Seed collection

Fully ripened *Jatropha curcas* fruits were collected from *Jatropha* plot close to Wildlife Ecotourism proposed site, Faculty of Agriculture, University of Benin, Benin City. The plot was measured to be 1008sqm and was divided into three (3) portions measured 336sqm each, the fruits were randomly picked from stand within the three portions. The seeds were extracted from the fruits and air-dried accordingly. The dried seeds were taken to Faculty of Agriculture, University of Benin, Benin City main laboratory. The seeds were dehusked and grinded using ceramic mortar and pestle.

3.2.2 Oil Extraction

The oil was extracted using a 500ml soxhlet extraction apparatus using N-Hexane. N-Hexane were recovered after removing the extracted sample from the extractor and oil parameters determined are;

3.2.2.1 Oil Moisture Content (m/c %) determination

Oil moisture content (m/c %): 2mls of oil was measured into some beaker of known weight. It was placed in drying oven for 3 hours at 103°C and dried to constant weight.

Moisture content was calculated as follows: -

$$MC = \frac{\text{initial weight} - \text{final weight after drying}}{\text{weight of sample (2mls of the oil)}} \times 100$$

3.2.2.2 Oil Percentage

The oil percentage was determined as follows;

$$\text{Oil \%} = \frac{\text{weight of oil} + \text{weight of empty flask}}{\text{weight of extracted grinded seeds}} \times 100$$

3.2.2.3 Iodine Value Determination

Mass of iodine in grams that is consumed by 100g of a chemical substance. Iodine numbers are often used to determine the degree or number of unsaturation in fatty acids. (Hanus method was used: a method for determining the iodine number of an oil or fat that consists in adding a mixture of iodine and bromine in glacial acetic acid and estimating the excess of unused halogen by titration with sodium thiosulfate.) with iodine bromide (AOAC official method 1993)

3.2.2.4 Acid Value Determination

This is the weight of KOH in mg needed to neutralize the organic acids present in 1g of fat or oil. Procedure: - 5ml of neutral ethyl alcohol was heated with 2g of oil in 50ml beaker until the mixture boils. It was allowed to cool and was titrated with 0.1N KOH solution using 2 drops of phenolphthalein as indicator while shaking consistently. A permanent pink and point was obtained. Acid value = 0.56 x Amt. of 0.1N KOH used.

3.2.2.5 Free Fatty Acid Determination

$$\text{FFA \%} = \frac{\text{Acid value (\%)}}{2}$$

Formula to determine free fatty acid

3.2.2.6 Peroxide value:

Procedures; Weigh out 1g of oil or fat into a clean dry boiling tube and while still liquid add 1g powdered potassium iodide and 20ml of solvent mixture (2 vol glacial acetic acid + 1 vol chloroform). Place the tube in boiling water so that the liquid boils within 30 seconds and allow to boil vigorously for not more than 30 seconds. Pour the contents quickly into a flask containing 20ml of potassium iodide solution (5%), wash out the tube twice with 25ml water and titrate with 0.002M sodium thiosulphate solution using starch. Perform a blank at the same time. The peroxide value is often reported as the number of ml of 0.002 N (M) sodium thiosulphate per g of sample. If the value so obtained is multiplied by 2, the figure then equals milliequivalents of peroxide oxygen per kg of sample (ml M per kg), which has greater international recognition.

3.2.2.6 Crude protein:

Procedures: Weigh 1 to 5 g of the food or feed material into a kjedahl flask and add 25ml of sulphuric acid. Add about 25.5g of potassium sulphate, a pinch of copper sulphate and a pinch of selenium (or a drop of mercury), all of this to act as catalysts. Some pellets containing all this catalytic chemical or variants of them are also available commercially for use. Place the material in a furnace or heater (usually a heating mantle) and heat (digest) until the mixture is clear, when all the nitrogen (except that in nitrate and nitrite) present in the material would have been converted to ammonia. This usually correspond with the time when the digest is clear, having changed from dark brown to light green or golden yellow color. Cool the flask, add some quantity of water to reduce the acidity of the digest. The quantity of water should be at least five times the quantity of digest available for distillation.

Then add some quantity (in excess) of 40% sodium hydroxide to the digest. Place the flask back on the heater and distill off into a flat bottom flask containing 5ml of boric acid/ methyl red indicator solution and continue to distill until about 50-150 ml distillate have been collected. The ammonia is liberated by adding the sodium hydroxide. The distillate obtained is usually green or golden yellow in colour. Titrate the distillate so obtain with HCL to the end point. The end point is achieved when the green distillate turns to the original pink color of the boric acid/ indicator. This step is called the titration stage. Calculate the nitrogen(N) content of the material. The crude protein content can't be obtained by multiplying the nitrogen content by a factor, 6.25. The factor 6.25 is used because of the assumption that the average protein contains 16% nitrogen ($100/16=6.25$). The factor 6.25 is not true for all materials.

$$\% \text{ crude protein} = \frac{N_a \times V_a \times 1.4 \times 6.25}{\text{wt of sample}}$$

Where:

N_a = normality of acid

V_a = volume of acid obtained on titration

1.4 = atomic weight of N

W_t = weight

3.2.2.7 Saponification Value

Procedures; Weigh 2g of the oil or fat into a conical flask and add exactly 25ml of the alcoholic potassium hydroxide solution. Attach a reflux condenser and heat the flask in boiling water for 1hour, shaking frequently. Add 1ml of phenolphthalein(1%) solution and titrate hot the excess alkali with 0.5M hydrochloric acid (titration = aml).

3.3 Data Analysis

Data collected were subjected to inferential statistics using analysis of variance (ANOVA) at 5% level of significance. Significant variables were separated using Duncan New Multiple Range Test at 5% level of significance.



Plate 1: student in *Jatropha curcas* plot



Plate 2: *Jatropha curcas*



Plate 3: *Jatropha curcas* dried seed

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Results

The results showed that moisture and crude protein content of the three samples of *Jatropha curcas* seed oils were significantly difference ($P < 0.05$). The moisture content ranged between 0.08 ± 0.001 to 0.10 ± 0.002 with an average of $0.09 \pm 0.01\%$. The crude protein content ranged between 17.50 ± 1.7 to 17.80 ± 1.5 with an average of 17.70 ± 1.5 . There was no significant difference ($P > 0.05$) in %fat among the samples. The %fat ranges between 39.4 ± 2.9 to 40.2 ± 2.2 , with an average of 39.8 ± 2.7 (Table 4.1).

Table 4.1: Nutrients composition of the samples of *Jatropha curcas* seed oil

Composition	Sample I	Sample II	Sample III	Mean
%Moisture Content	0.09 ± 0.01^a	0.08 ± 0.01^a	0.10 ± 0.02^a	0.09 ± 0.01
%Crude Protein	17.7 ± 1.4^b	17.5 ± 1.7^{ab}	17.8 ± 1.2^a	17.7 ± 1.5
%Fat	39.8 ± 3.1^a	39.4 ± 2.9^a	40.2 ± 2.2^a	39.8 ± 2.7

Means with same letter across the Table are not significant at 5% level

The results revealed significant difference ($P < 0.05$) in acid values, saponification, and free fatty acid among the three samples of *J. curcas* seed oil, while the values of Iodine and peroxide among the three samples showed no significant difference ($P > 0.05$). The average acid values, Iodine, saponification, peroxide and free fatty acid of *Jatropha curcas* seed oil were 1.20 ± 0.3 , 82.4 ± 3.6 , 166.0 ± 4.9 , 1.93 ± 0.1 , and 0.40 ± 0.02 respectively (Table 4.2).

Table 4.2: Chemical composition of the samples of *Jatropha curcas* seed oil

Composition	Sample I	Sample II	Sample III	Mean
Acid value (mgNaOH/g)	1.01±0.2 ^b	1.20±0.4 ^a	1.25±0.1 ^a	1.20±0.3
Iodine (mgI ₂ /g)	82.3±2.5 ^a	81.3±1.3 ^a	83.7±1.9 ^a	82.4±3.6
Saponification (mgNaOH/g)	153.2±2.8 ^b	152.6±3.1 ^b	192.1±2.5 ^a	166.0±4.9
Peroxide values (mEqO ₂ /kg)	1.92±0.1 ^a	1.90±0.3 ^a	1.97±0.1 ^a	1.93±0.1
Free Fatty Acid	0.33±0.0 ^b	0.50±0.2 ^a	0.38±0.2 ^b	0.40±0.02

Means with same letter across the Table are not significant at 5% level

Table 4.3 showed the fatty acid composition of the samples of *Jatropha curcas* seed oil. The results revealed no significant difference ($P>0.05$) in fatty acid composition among the samples of *J. curcas* seed oil used for this study. The average palmitic, stearic, oleic, linoleic, arachidic, arachidoleic, and behenic composition of *J. curcas* seed oil were 11.5±0.2, 17.9±1.1, 52.8±2.4, 49.2±1.8, 4.5±0.1, 1.6±0.2 and 0.50±0.0 respectively.

Table 4.3: Fatty Acid composition of the samples of *Jatropha curcas* seed oil

Fatty Acid	Sample I	Sample II	Sample III	Mean
Palmitic	11.6±0.2 ^a	11.0±0.4 ^a	11.8±0.1 ^a	11.5±0.2
Stearic	18.1±2.5 ^a	17.4±1.3 ^a	18.3±1.9 ^a	17.9±1.1
Oleic	52.8±2.8 ^a	52.2±3.1 ^a	53.4±2.5 ^a	52.8±2.4
Linoleic	48.5±0.1 ^a	49.2±0.3 ^a	49.8±0.1 ^a	49.2±1.8
Arachidic	4.4±0.0 ^a	4.5±0.2 ^a	4.7±0.2 ^a	4.5±0.1
Arachidoleic	1.6±0.1 ^a	1.4±0.3 ^a	1.7±0.1 ^a	1.6±0.2
Behenic	0.50±0.0 ^a	0.45±0.0 ^a	0.55±0.0 ^a	0.50±0.0

Means with same letter across the Table are not significant at 5% level

The chemical composition of *Jatropha curcas* seed oil as compared with those from commercial available plant oil are given in Table 4.4. The results showed that the acid values of the plant oils ranged from 0.95±0.01-25.62±2.26 mg/NaOH/g, the highest (25.62±2.26 mg/NaOH/g) was observed in palm kernel oil, while the lowest (0.95±0.01mgNaOH/g) was observed in sunflower oil. The iodine values ranged from 54.14±1.19-127.03±3.60 mgI₂/g, the highest (127.03±3.60 mgI₂/g) was observed in palm kernel oil, while the lowest (54.14±1.19 mgI₂/g) was observed in shea nut oil. The saponification values ranged from 112.54±0.03-258.78±4.25 mgNaOH/g, the highest (258.78±4.25 mgNaOH/g) was observed in palm kernel oil, while the lowest (112.54±0.03 mgNaOH/g) was observed in *Cucumis melodrama* oil. The peroxide values ranged from 0.39±0.01-6.50±0.18 mEqO₂/kg, the highest (6.50±0.18 mEqO₂/kg) was observed in shea nut oil, while the lowest (0.39±0.01 mEqO₂/kg) was observed in palm kernel oil.

Table 4.4: Chemical composition of *Jatropha curcas* seed oil as compared with those from commercial available plant oil.

Plant Oil	Acid value (mgNaOH/g)	Iodine (mgI ₂ /g)	Saponification (mgNaOH/g)	Peroxide values (mEqO ₂ /kg)
<i>Jatropha curcas</i> (Present study)	1.20±0.3	82.4±3.6	166.0±4.9	1.93±0.1
<i>Sesamum indicum</i> (Ouattara, 2015)	1.16±0.06	108.28±3.11	191.8±2.33	3.38±0.20
<i>Cocubita pepo</i> (Ouattara, 2015)	1.29±0.05	95.55±3.6	117.13±2.37	1.33±0.15
<i>Cucumis melodrama</i> (Ouattara, 2015)	2.51±0.03	76.34±2.6	112.54±0.03	1.45±0.02
Shea nut (Ouattara, 2015)	11.17±1.62	54.14±1.19	197.40±0.07	6.50±0.18
<i>Trichilia emetic</i> (Bizuneh, 2014)	8.13±1.35	60.15±1.89	180.09±1.44	0.56±0.03
Palm kernel (Bizuneh, 2014)	25.62±2.26	127.03±3.60	258.78±4.25	0.39±0.01
<i>Colocynthis citrullus</i> (Bizuneh, 2014)	6.17±0.82	111.46±2.42	201.03±3.74	2.72±0.35
Soya bean (Bizuneh, 2014)	4.30±0.45	97.29±3.02	192.45±1.48	0.37±0.02
Sunflower (Bizuneh, 2014)	0.95±0.01	119.92±2.69	182.23±2.15	6.32±0.68

4.2 Discussion

The findings showed no significant difference ($P>0.05$) in % moisture content, % crude protein and % fat among the three samples of *Jatropha curcas* seed oils used for this study (Table 4.1). However, the mean % moisture contents of *J. curcas* seed oils was 0.09 ± 0.01 . The findings was in the same range reported for *Arachis hypogaea* oil (0.089%) by Ouattara *et al.*, (2015). High moisture content creates problem in trans esterification. High moisture content of oil makes the oil more susceptible to change in the physical appearance and mold growth. The mean % crude protein (17.7 ± 1.5) was higher than the value reported for shea nut (12.93 ± 2.60) and slightly lower than the values reported for *Sesamum indicum* (19.96 ± 0.73), *Cucurbita pepo* (18.87 ± 1.50) and *Cucumis melodrama* (18.60 ± 0.02) by Ouattara *et al.*, (2015). The high % crude protein observed in this study indicates that *J. curcas* seed oil could be used for food enrichment for human consumption and also as source of raw material for livestock feed. The fat content can also be compared with 37.1% for mustard and 39% for niger seeds (Gopalan *et al.*, 2007), hence are regarded as oil seeds. The average % crude fat content of *J. curcas* seed oil observed in this study was $39.8\pm 2.7\%$, which exceed that of some common edible oils such as cotton seed oil (22 to 24%), safflower (30 to 35%), soy bean oil (18 to 22%), olive oil (24 to 40%) (Nichols and Sanderson, 2003) and unconventional oilseeds such as *Canariumschwenfurthii* fruits oil (38.1%) (Nzikou *et al.*, 2006). Thus, *J. curcas* seeds oil observed in this study is rich source of fats. Therefore, *J. curcas* can be considered as a potential source of vegetable oil for domestic and industrial purposes after detoxification.

The findings revealed the chemical composition of *J. curcas* compared favourably with those commercial available oils as shown in Table 4.2 and 4.4. The average acid

value of *J. curcas* seed oil (1.20 ± 0.3 mgNaOH/g) compared favourably with 1.20 ± 0.3 mgNaOH/g reported for *S. indicum* reported by Ouattara, (2015) and 0.95 ± 0.01 mgNaOH/g reported by Bizuneh, (2014). The value is slightly lower than 1.29 ± 0.05 mgNaOH/g and 2.51 ± 0.03 mgNaOH/g reported for *C. pepo* and *C. melodrama* respectively by Ouattara, (2015). However, the acid value of *J. curcas* seed oil recorded is lower than that reported for shea nut (11.17 ± 1.62 mgNaOH/g) by Ouattara, (2015), *T. emetic* (8.13 ± 1.35 mgNaOH/g), palm kernel (25.62 ± 2.26 mgNaOH/g), *C. citrullus* (6.17 ± 0.82 mgNaOH/g) and soya bean (4.30 ± 0.45 mgNaOH/g) reported by Bizuneh, (2014). The acid values observed in this study disagree with the acid values of *J. curcas* seed oil (9.48 ± 0.22) reported by Ugbogu *et al.*, (2014). The different acid values could be attributed to the region the samples were collected as well as variety of the *J. curcas* used for the studies. Acid value is a direct measure of the percentage content of free fatty acids in a given amount of oil. It is a measure of the extent to which the triglycerides in the oil have been decomposed by lipase action into free fatty acids; acid value depends on the degree of rancidity which is used as an index of freshness (Ochigbo and Paiko, 2011). It is common knowledge that these parameters are a measure of the level of spoilage of oil. It is used as indicator for edibility of oil and suitability for use in paint industry (Bizuneh, 2014). Acid value of the oil suitable for edible purpose should not exceed 4 mg NaOH/g (Oladele and Oshodi, 2008).

The iodine value observed in this study was 82.4 ± 3.6 mgI₂/g. This result compared positively with the acceptable range of iodine value of oil samples which falls within the range of 30 to 100 recommended for vegetable oils (Kagwachie and Anozie, 1995). Most non-conventional seed oils have higher iodine values. Iodine value of a vegetable oil of any given species of plant may be influenced by its varietal genetic

and the environment in which the oil is investigated (Encyclopedia Britannica, 2015). The iodine value of *J. curcas* seed oil observed in this study is lower than many commercial oils (Table 4.4) such as palm kernel oil (127.03 ± 3.6 mgI₂/g), soy bean oil (97.29 ± 3.02 mgI₂/g) and sunflower (119.92 ± 2.69 mgI₂/g) reported by Bizuneh, (2014). The relative low iodine value of *J. curcas* seed oil may be indicator of the presence of a few unsaturated bonds and low susceptibility to oxidative rancidity (Eka, 1980).

The saponification value of *J. curcas* seed oil observed in this study was 166.0 ± 4.9 (Table 4.2). The value is compared favourably with some conventional oils. According to Ezeagu *et al.*, (1998) a saponification value of 200 mg mgNaOH/g indicates high proportion of fatty acids of low molecular weight. The high saponification value of *J. curcas* seed oil could make it useful as sources of essential fatty acids required in the body (Akanni *et al.*, 2005). However, saponification values obtained are within the range for edible oils reported by Eromosele *et al.*, (1994).

The average peroxide value 1.93 ± 0.1 mEqO₂/kg as shown in Table 4.2 is lower than the maximum acceptable value of 10 mEqO₂/kg set by the Codex Alimentarius Commission for seed oils (Abayeh *et al.*, 1998). Peroxide value is one of the most widely used chemical tests for the determination of the quality of oils. It measures the rancidity during the initial stages of lipid oxidation because the value increases to a maximum and then decreases as storage time increases (Akinoso *et al.*, 2010). Peroxide values are used as indicator of deterioration of oils. Fresh oils have peroxide values less than 10mEqO₂/kg (Codex Alimentarius Commission, 1982). While values above 20 indicates rancid taste and disagreeable odour (Pearson, 1976). The low peroxide value of *J. curcas* seed oil indicates low level of oxidative rancidity of the oil and also suggests the presence of high levels of antioxidants.

The average free fatty acid (FFA) values of *J. curcas* seed oil obtained in this study was 0.4 ± 0.02 (Table 4.2). Free fatty acid decreases as oil is subjected to refining processes. Free fatty acid is the most commonly used parameter for determining the quality of oil. Acid value is a measure of free fatty acid in the oil as a result of deterioration. These deteriorations result in hydrolysis of triglycerides (oils) to yield free fatty acids. The low percentage FFA content of *J. curcas* (0.4 ± 0.02 %) seed oil observed in this study indicates that the oil can be stored for a long time without spoilage through oxidative rancidity.

The fatty acid composition of *J. curcas* seeds oil observed in this study revealed Oleic acid (52.8 ± 2.4 %) and Linoleic acid (49.2 ± 1.8 %) (Table 4.3) which are unsaturated fatty acids as the most highest. The findings is in agreement with the work of Pradhan *et al.*, (2011), Ugbogu *et al.*, (2014) and Michael *et al.*, (2019) who reported high values of Oleic acid and Linoleic acid in *J. curcas* seed. The findings is also in agreement with those of (Augustus *et al.*, 2002; Akintayo, 2004). The prevalence of the unsaturated fatty acids and high values of the iodine index indicate that the *J. curcas* oil is of the unsaturated type (Nzikou *et al.*, 2009). This high level of polyunsaturated fatty acid in the seed oil can be harnessed in the management of cardiovascular diseases (Nzikou *et al.*, 2009; Chinyere *et al.*, 2009).

CHAPTER FIVE

5.0 CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

The nutritional and chemical compositions of *J. curcas* seed oil was investigated, findings from this study showed that *J. curcas* seed oil has great potentials for human nutritional purposes and being used in the formulation of new foods and feeds which will foster economic utility, but due to the presence of some toxic substances this cannot be achieved at the moment. The proximate composition studied revealed that the *J. curcas* seed oil is a good source of protein and has high fat content. This makes the seed a valuable dietary supplement which can help reduce the problem of protein energy malnutrition. The Acid value and percentage free fatty acid are used as indicator of the edibility of oil. These two parameters determine the use of oil for either edible or industrial utility. The chemical composition of *J. curcas* seed oil studied revealed that the acid, iodine, saponification, peroxide and free fatty acid values, compared favourably with the specifications set by Codex Alimentarius Commission (CAC) for some named edible oil. This attributes promotes *J. curcas* seed oil as a relevant source of vegetable oil which could be explored commercially in order to reduce the pressure on frequently used vegetable oils in the country.

5.2 Recommendation

Based on the findings of this study, the following recommendations were made

1. Further study on detoxification of *J. curcas* seed oil should be carried out.

2. Further studies on how *J. curcas* seed oil can be more useful in human nutrition in form of dietary supplements and possibly for livestock feed production should be carried.

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APPENDIX

ANALYSIS OF VARIANCES FOR MOISTURE CONTENT OF *J. curcas*

Anova: Single Factor

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Su</i> <i>m</i>	<i>Average</i>	<i>Variance</i>
Column 1	3	0.2 8	0.09333 3	0.00013 3
Column 2	3	0.2 4	0.08	0.0001
Column 3	3	0.2 9	0.09666 7	0.00023 3

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	0.00046	7	0.00023	1.5	0.29629	5.14325
Within Groups	0.00093	3	0.00015		6	3
Total	0.0014	8	6			

ANALYSIS OF VARIANCES FOR IODINE VALUE OF *J. curcas*

Anova: Single Factor

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
Column 1	3	246.95	82.31667	1.755433
Column 2	3	243.8	81.26667	0.841233
Column 3	3	251.05	83.68333	3.227233

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	8.810556	2	4.405278	2.269241	0.184552	5.143253
Within Groups	11.6478	6	1.9413			
Total	20.45836	8				

ANALYSIS OF VARIANCES FOR ACID OF *J. curcas*

Anova: Single Factor

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Su m</i>	<i>Average</i>	<i>Variance</i>
Column 1	3	3.2	1.06666	0.00173
Column 2	3	3.6	1.2	0.0001
Column 3	3	3.7	1.26	0.0013

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	0.05875	6	0.02937	28.1276	0.00089	5.14325
Within Groups	0.00626	7	0.00104	6	5	3
Total	0.06502	2	8			

ANALYSIS OF VARIANCES FOR FREE FATTY ACID CONTENT OF *J. curcas*

Anova: Single Factor

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Su m</i>	<i>Average</i>	<i>Variance</i>
Column 1	3	1.0	0.33666	3.33E-05
Column 2	3	1.5	0.5	0.0007
Column 3	3	1.1	0.38	0.0001

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	0.04295	6	0.02147	77.32	5.21E-05	5.14325
Within Groups	0.00166	7	0.00027	8	8	3

	0.04462		
Total	2	8	

ANALYSIS OF VARIANCES FOR SAPONIFICATION VALUE OF *J. curcas*

Anova: Single Factor

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
		459.6	153.226	6.84463
Column 1	3	8	7	3
Column 2	3	457.8	152.6	12.73
		576.2	192.073	5.44973
Column 3	3	2	3	3

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	3067.6	2	1533.8	183.876	4.14E-06	5.14325
Within Groups	50.0487	6	8.34145			
Total	3117.64	9				