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PHYSICOCHEMICAL PROPERTIES OF PUMPKIN SEEDS GROWN IN GHANA

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PHYSICOCHEMICAL PROPERTIES OF PUMPKIN SEED OIL GROWN IN GHANA

BY

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JULY, 2020

DECLARATION

STUDENT'S DECLARATION

I, Rita Boateng, declare that this Dissertation, with the exception of quotations and references contained in published works which have all been identified and duly acknowledge, is entirely my original work, and that to the best of my knowledge it does not contain any material which is formerly published or written by any other persons except where due reference is written.

STUDENT NAME: RITA BOATENG

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SUPERVISOR'S DECLARATION

I hereby declare that the preparation and presentation of this work was supervised in accordance with the guidelines for supervision of Dissertation as laid down by the University of Education, Winneba.

NAME OF SUPERVISOR: DR. GILBERT OWIAH SAMPSON

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DATE:…….

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DEDICATION

I dedicate this work to my sweet sister; Jewel Adu Dentaah

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ABSTRACT

The pumpkin seed oil has many health benefits and is considered as nutritional powerhouses, with a wide variety of nutrients ranging from magnesium and manganese to copper, protein, and zinc. Despite the enormous health benefits of pumpkins, they are grown on a low scale in Ghana because of lack of knowledge on its physiochemical properties, nutritional value, and market potential. Based on this, the study aimed at determining the physicochemical properties of pumpkin seed oil and to evaluate it nutritional composition. Several equipment and materials were used for this study. Some equipment and materials used for this study include; vacuum oven, stoppered conical flask, burette, retort stand. In this study free fatty acid, peroxide value, saponification value, and iodine value was determined and was then calculated (AOAC, 1990). The data obtained were processed and analyzed with the aid of the Statistical Package for Social Sciences (SPSS) version 23.0. The results were organized into simple Tables using Mean and standard deviation. The study showed that pumpkin seed had a free fatty acid value of 0.06%. The finding revealed that the acid value of pumpkin seed oil was 0.12 ± 0.00 mgKOH/g. The peroxide value of pumpkin seeds oil was 9.69± 0.12 meq o2/kg oil. Also, the saponification value of the oil was 44.19 ±0.80. The pumpkin seed oil had an iodine value was 10.62 ± 0.33 (g of I_2/100g oil), which indicated a low degree of unsaturation. It was concluded that the pumpkin seed oil can be considered as a new and valuable source of edible oil according to its physicochemical properties. The study recommended that more attention and care should be taken for pumpkin cultivation to produce seeds for oil production in Ghana. Also, the ministry of health should encourage the consumption of pumpkin seed oil as frequent consumption of pumpkin seed oil can increase better nutrition and health.

CHAPTER ONE

INTRODUCTION

1.1 Background to the Study

Pumpkin is cultivated throughout the world and traditionally used as medicine in China, Yugoslavia, Argentina, India, Mexico, Brazil, America and other African countries like Ghana (Song, Li, Hu, Ni, & Li, 2011). Pumpkin has gained extensive attention in current times due to the good nutritional composition and health shielding values of its seeds. Pumpkin seeds are typically flat and asymmetrically oval, have a white outer husk, and are light green in color after the husk is removed. Some cultivars are huskless, and are grown only for their edible seed (Song et al., 2011).

In Ghana, pumpkin is locally known as "ɛferɛ". In Ghana, one such less popular fruit is the pumpkin fruit (Kuada, 2005). Pumpkin plants are mainly grown on small scales by the households in Ghana for domestic consumption (Abbiw, 1990). Pumpkin belongs to the family Cucurbitaceae with different species and cultivated all over the world for multiple purposes ranging from commercial to agricultural intentions comprising with decorative uses (Patel, 2013). Crops belonging to this family are known as cucurbits and includes melon, water melon, pumpkin, squash and cucumbers. Pumpkin fruits vary in size, colour, shape and weight (Wang, Behera & Kole, 2012). Cultivars of pumpkin include *Cucurbita pepo, Cucurbita mixta, Cucurbita maxima and Cucurbita moschata* and they are classified according to the texture and shape of their stems (Rakcejeva, Galoburda, Cude & Stra, 2011).

Pumpkin seeds, generally considered agro-industrial waste, are an extraordinarily rich source of bioactive compounds with interesting nutraceutical properties (Rakcejeva, et al., 2011). In recent years, several studies Bardaa, Halima, Aloui, Mansour, Jabeur, Bouaziz & Sahnoun, 2016; Medjakovic, Hobiger, Ardjomand-

Woelkart, Bucar, Jungbauer, 2016; Wang, Lu, Zhang, Shen, Xu, Zhan, Jin, Zhang & Wang, 2018) have highlighted the health properties of pumpkin seed oil against many diseases, including hypertension, diabetes, and cancer. It also shows antibacterial, antioxidant, and anti-inflammatory properties (Yadav, Jain, Tomar, Prasad & Yadav, 2010; Gutierrez, 2016). Due to the presence of interesting natural bioactive compounds, such as carotenoids, tocopherols, and sterols, pumpkin-derived products have a wide spectrum of biological activity, proven by in vivo experiments (Montesano, Blasi, Simonetti, Santini & Cossignani, 2018).

The seeds of pumpkins are often eaten as a snack after roasting and salting in Arab countries (Al-Khalifa 1996). Xanthopoulou, Nomikos, Fragopoulou & Antonopoulou (2009) stated that pumpkin seeds are used as an additive in salads, flakes, pastries, etc. due to their beneficial phytochemical content. The addition of these seeds can be considered a good substitute for the nutritional enhancement of food products (Gorgonio, Pumar & Mothe, 2011). Pumpkin seeds are a rich natural source of protein with the range of 25 to 37% and oil with the range of 37 to 45% and are renowned as valuable oil seeds with protein for human consumption (Milovanoic & Vucelic-Radovic 2008). Pumpkin seeds contain unsaturated fatty acids especially omega 3 fatty acids (Murkovic, Hillebrand, Winkler, Leitner & Pfannhauser, 1996). These seeds are also rich in phytosterols (Phillips, Ruggio & Ashraf-Khorassani, 2005; Ryan, Galvin, O'Connor, Maguire & O'Brien, 2007), polyunsaturated fatty acids (Phillips, Ruggio & Ashraf-Khorassani, 2005).

1.2 Statement of the Problem

The pumpkin seed oil has many health benefits and is considered as nutritional powerhouses, with a wide variety of nutrients ranging from magnesium and manganese to copper, protein, and zinc. Despite the enormous health benefits of pumpkins, they are grown on a low scale in Ghana because of a lack of knowledge of its chemical properties, nutritional value, and market potential. Poor storage of the pumpkins also accounts for quick deterioration and spoilage of the fruits. It is very important to know its nutritive value to encourage the increase in its production, consumption, and usage for nutritional and technological applications.

Studies in Ghana on physiochemical and nutritional composition of pumpkin seed oil exist though is scanty. However, the nutritional or physiochemical properties of the pumpkin seed oil are limited. The seed oil can be exploited as biodiesel or used extensively in animal feed which at the moment the country spends a lot of foreign exchange to export. Also, the value Ghanaians place on the pumpkin is low, hence this present study analyzes the physiochemical and nutritional composition of locally available pumpkin seed (C. maxima Linn) in Ghana.

1.3 Main objective

The main objective of the study was to determine the physicochemical properties of pumpkin seed oil.

1.4 Specific Objectives

The specific objective of the study were to;

a) Determine the moisture contact of the pumpkin seed oil.
b) Determine the free falty acid in the pumpkin seed oil.
c) Determine the peroxide value in the pampking seed oil.
d) Determine the saponification value in the pumpkin seed oil.
e) Determine the iodine value in the pumpkin seed oil.

1.5 Significance of the Study

The impact of the study could be seen from different angles. First of all, the study would pinpoint the physical and chemical properties of pumpkin seeds that will encourage the increase in the production, consumption, and usage for nutritional and technological applications. The report can also be used as learning material especially within areas related to production and consumption of pumpkin seeds.

From the study, the government may use the findings in strengthening policies related to the growth of pumpkin seeds due to its market potential. The findings of the study may provide information to investors about the physical characteristics and nutritive value of local pumpkin seed produced in Ghana to decide whether to invest their assets.

Finally, the research will serve as an essential source of reference to future researchers who would be researching this problem and its related studies.

1.6 Scope of the study

The study was focused on pumpkin seeds produced in Ghana. The pumpkin seed was obtained from the Upper West Region of Ghana. The study specifically concentrates on the physicochemical properties of pumpkin seed samples from Upper West Region of Ghana.

1.7 Organization of the study

The study was organized into five chapters. Chapter one consisted of the background to the study, statement of the problem, main objective, specific objective, research questions, the significance of the study, the scope of the study, and the organization of the study. Chapter two consists of literature related to the topic. Chapter three discusses the methodology which includes; research design, population for the

study, data collection, data analysis plan. Chapter four dealt with the presentation and discussion of findings of results with the set of objectives. Chapter five was exclusively devoted to the summary of the major research findings, conclusions, and recommendations.

CHAPTER TWO

LITERATURE REVIEW

2.1 Description of pumpkin seeds

Pumpkin and their seeds are native to America and various species are found across the North, South and Central America. Pumpkin seeds - a renowned food among many inhabitant American tribes, who consume these seeds for their nutritional and medicinal properties (El-Adawy & Taha, 2001). From America, the pumpkin seeds got popularized and spread to the rest of the globe through trade and exploration over many centuries. India and other parts of Asia also included these seeds into a place of importance instead of discarding them. Today, China ranks first in the production of pumpkin and pumpkin seeds in the whole world. India, Russia, Ukraine, Mexico, and the U.S. are also major producers of pumpkin and pumpkin seeds (El-Adawy & Taha, 2001).

Pumpkin seeds also known as pepitas are flat, dark green seeds encased in a yellow-white husk. These seeds have a malleable, chewy texture and a subtly sweet, nutty flavour. They can be relished all over the year. Pumpkin seeds are available in the food stores in various forms like raw and shelled, raw and unshelled, roasted and shelled, roasted and unshelled. Like watermelon, cucumber, and squash seeds, pumpkin seeds belong to the *Cucurbitaceae* family. *Cucurbita pepo, Cucurbita maxima, Cucurbita moschata,* and *Cucurbita mixta* are the common varieties of pumpkin. Raw or roasted pumpkin seeds are used as snack food for human consumption in many cultures all over the world. The kernels of pumpkin seeds have been utilized as flavor enhancers in gravies and soups, and used in cooking, baking and ground meat formulations as a nutrient supplement and a functional agent (Tsaknis *et al*., 1997; El-Adawy & Taha, 2001).

The oil content of pumpkin seed is 50% reported by (Belyaev, 1992). And also found in the range of 35-38% (Eckey and Lawrence, 1954). A study conducted by Kwiri, et al. (2014) in Zimbabwe on proximate composition of pumpkin seed revealed the oil content of pumpkin seed to be 43.460±0.098%. Achu et al. (2005) and Loukou et al. (2007) (mainly in Côte d'Ivoire) revealed similar results on the analysis of pumpkin seeds. For instance, their studies revealed oil content within the ranges of 28 - 40.49.

Ardabili et al. (2011) study on the chemical composition and physicochemical properties of pumpkin seeds indicated that the oil content of pumpkin seed was found to be 41.6 ± 2.7%. This value fell in the range reported for different species of *cucurbita* (9.8-52.1%) and different varieties of *C. pepo* (31.2-51.0%) (Stevenson *et al.*, 2007). It has been claimed that such differences in the oil content can be attributed to genetic diversity and climate conditions (Stevenson *et al.*, 2007). Also, the oil content of the pumpkin seed in the present study was found to exceed, or be comparable to, that of some common edible oils such as cottonseed (22-24%), safflower (30-35%), soybean (18- 22%), rapeseed (40-48%), and olive (12- 50%) (Nichols and Sanderson, 2003). Therefore, the pumpkin seed can be considered as a potential source of vegetable oil for domestic and industrial purposes.

2.2 Pumpkin seed oil

Fats and oils are produced throughout the world from both animals and plants. Vegetable oils are essential in meeting global nutritional demands and are utilized for many foods and other industrial purposes (Idouraine *et al*., 1996). Despite the broad range of sources for vegetable oils, the world consumption is dominated by soybean, palm, rapeseed, and sunflower oils with 31.6, 30.5, 15.5, and 8.6 million tons consumed per year, respectively (Stevenson *et al*., 2007). These conventional sources of vegetable

oil no longer meet the ever-increasing demands of domestic and industrial sectors (Idouraine *et al*., 1996).

Odoemelam (2005) reported that seeds with nutritive and calorific values are also good sources of edible oils and fats. The pumpkin seeds possess valuable dietary and medicinal qualities besides the source good- quality edible oils (Gohari *et al*., 2011). The oil of pumpkin seeds is being used as cooking oil in some countries in Africa and the Middle East, and as a salad in South Australia and adjacent regions in Solvenia (Wenzl et al, 2002). Pumpkin seed oil produced from roasted pumpkin seeds is thick, green-red color (Kerft & Kerft 2007). The pumpkin seed oil contains, vitamins, minerals, and dietary fiber and fatty acids, such as oleic acid and alpha-linolenic acid (Baves *et al*., 2007). The pumpkin seed oil has many health benefits derived from its consumption. It contains essential fatty acids that help maintain healthy blood vessels, nerves, and tissues (Levin, 2008).

In Ghana, the major sources of edible oils are groundnut oil (*Arachis hypogoea* L.); coconut oil (*Cocos Nucifera*); sesame seeds oil (*Sesamum indicum* L.). These oils are used mainly as cooking oils, beside the soap production. With increasing demand, which has led to the importation of cooking oils, so, there is a need to source for local oil-bearing seeds which can be used in oil production, both for consumption and industrial applications (Imad, 2003).

2.3 Physical characteristics of pumpkin seed oil

2.3.1 Density

The density of pumpkin seeds oil measured at 30 ºC was given 0.78 given by (Gohari *et al*., 2011). The Indian and American varieties to have specific gravities of 0.915 at 25/25 ºC and 0.919 at20/20 ºC, respectively (Butt, 1995). Devi, Prasad, and Palmei (2018) indicated that the whole pumpkin seeds had a lower bulk density and a

higher true density than the kernels. The mean value obtained for bulk density and true density is 398±0.80 kg/m3 and 1157±1.02 kg/m3 for whole pumpkin seed whereas for the kernel, it was found to be 475±0.95 kg/m3 and 1068±0.94 kg/m3. The study conducted by Altuntas, (2008) reported bulk density and true density which ranges from 381±0.80 kg/m3 and 1136±1.02 kg/m3. The 1000 seed weight, geometric mean diameter and porosity values for whole pumpkin seed (202.2±0.75g, 7.42±0.35mm, 65.60±0.47%) are greater than the dehulled pumpkin seed (148.1±0.87g, 5.54±0.29mm, 55.52±0.55%). Husk content of pumpkin seed was found to be 26.75±0.98%. The results obtained were found to be similar to the earlier findings reported by Yildiz et al., (2013).

2.3.2 Viscosity of pumpkin seed oil

Viscosity is an important parameter for the design of industrial processes. It can also be used to evaluate the quality of fats and oils used in frying (Nichols & Sanderson, 2003). The viscosity of pumpkin seeds oil is 48.9 reported by Imad (2003) and also the value of viscosity 47.3 reported by Oomah *et al*. (2000). The viscosity measured in the study conducted by Ardabili et al. (2011) was 93.659 ± 0.48 which is higher than those reported by Tsaknis *et al.* (1997) for *C. maxima* and *C. pepo* (72 cP) and that reported by Alfawaz (2004) for *C. maxima* (48.09 cP). Oomah *et al.* (2000) reported a value of 26.0, 47.3, and 49.4 cP for raspberry, safflower, and grape seed oils, respectively. Viscosity is an important parameter for the design of industrial processes. It can also be used to evaluate the quality of fats and oils used in frying (Nichols & Sanderson, 2003).

2.3.3 Refractive index of pumpkin seed oil

The refractive index of common pumpkin seeds oil is 1,466 reported by Gohari *et al*. (2011). The European varieties have a refractive index within the range of 1.406-

1.469 reported by Imad (2003). The Refractive index is used by most processors to measure the change in unsaturation as the fat or oil is hydrogenated.

The refractive index of oils depends on their molecular weight, fatty acid chain length, degree of unsaturation, and degree of conjugation (Nichols & Sanderson, 2003). The pumpkin seed oil showed a refractive index of 1.4662 ± 0.0001, which was similar to those reported by Lazos (1986) for the pumpkin (1.4616) and melon (1.4662) seed oils (Ardabili et al., 2011). This value that fell in the range reported for the pumpkin seed oils (1.466-1.474) was lower than the range reported for sunflower and olive oils; higher than that for palm, palm kernel, and coconut oils; and within the range reported for canola, rapeseed and corn oils (Nichols and Sanderson, 2003). Pure oils have marked ranges of refractive index and density; thus, the degree of variation of a typical oil from its true values may indicate its relative purity.

2.3.4 Color of pumpkin seed oil

Pumpkin seeds oil was greenish-brown in color with nut-like taste (Gohari *et al*, 2011). The pumpkin seed oil has a pale yellow color described by Butt (1995). The characteristic dark color of the pumpkin seed oil was due to the carotenoids and chlorophyll (Untersuchungen, 1978). Most of the oils of Cucurbitaceae family seeds were nearly colorless (Gohari *et al*, 2011).

2.4 Chemical characteristics of pumpkin seed oil

2.4.1 Acid value of pumpkin seed oil

Commission expressed the permitted maximum acid values of 10 and 4 mg KOH/g oil for virgin palm and coconut oils, respectively (Alfawaz, 2004). It has been shown that oils become rancid when the peroxide value ranges from 20.0 to 40.0 meq O2/kg oil (Ajayi *et al.,* 2006). High acid value (4-12) for common pumpkin seeds oil

and high acid value for Indian and American varieties where they gave values 10.1 and 12.40 respectively (Gohari *et al*., 2011).

2.4.2 Free fatty acid content of pumpkin seed oil

The fatty acid composition of common pumpkin seed oil was within 24-41% as oleic, and the ranges of 7-12% Palmitic,6-7%stearic, 57%linoleic acids (Eckey & Lawrence, 1954). Yugoslavian pumpkin seeds oil contained: Palmitic 11.2% Stearic 5.0-5.3%, oleic 27, 1- 30.4 and linoleic 53.4-56.7% (Markovic & Bastic, 1976). Ardabili et al. (2011) considered the content of free fatty acids (0.39 ± 0.01 % as oleic acid), acid value (0.78 ± 0.02 mg KOH/g oil) and peroxide value (10.85 ± 0.62 meq O2/kg oil), the extracted pumpkin seed oil had an acceptable initial quality. The Codex Alimentarius Commission expressed the permitted maximum acid values of 10 and 4mg KOH/g oil for virgin palm and coconut oils, respectively (Alfawaz, 2004). It has been shown that oils become rancid when the peroxide value ranges from 20.0 to 40.0 meq O2/kg oil (Ajayi *et al*., 2006). On the other hand, according to the Codex Alimentarius Commission, the peroxide value for unrefined olive oil may be a maximum of 20 meq/kg oil (Markovic & Bastic, 1975). Therefore, considering that the oil studied was unrefined and its initial quality indicators were within the reported limits, the pumpkin seed oil can be regarded as an edible oil with good quality.

2.4.3 Peroxide value of pumpkin seed oil

Detection of peroxide value gave the initial evidence of rancidity in unsaturated fats and oils (Marco *et al* 2015). Peroxide value is expressed in units of milliequivalents per kg. Bialek *et al* (2016) determined the peroxide value of pumpkin seed oil which was 2.89 meq/kg. Vujasinovic *et al* (2010) found that the peroxide value of naked pumpkin seed and pumpkin seed with the hull as 2.95 and 5.04 mmol/kg. Srbinoska *et*

al (2012) studied the peroxide value of pumpkin seed whole and pumpkin seed kernel of two different varieties (*Cucurbita maxima* and *Cucurbita pepo*). Results showed that the peroxide value of whole seed and kernel of *Cucurbita maxima* was 4.93 and 4.26 meq/kg extract whereas, in *Cucurbita pepo*, it was 6.06 meq/kg extract for whole seed and 5.70 meq/kg extract for seed kernel.

Tsaknis *et al* (1997) analyzed the characteristics of crude and purified pumpkin seed oil in which peroxide value of crude and pumpkin seed oil was obtained as 9.20 and 9.04meq/kg. Adeel *et al* (2014) found that the peroxide value of pumpkin seed oil was 6.74meq 2/kg oil. More peroxide value is harmful to the shelf life of the oil. Exposure of peroxide provides initial evidence of rancidity and deterioration of pumpkin seed oil caused by unsaturated fats and oils. The peroxide value of pumpkin seeds oil in rang 10.85-11.47(Gohari *et al*, 2011). And peroxide value in the range 3.9-9.0 reported by Imad (2003).

2.4.4 Saponification value of pumpkin seed oil

Saponification is an indicator of the average molecular weight and, hence, chain length. It is inversely proportional to the molecular weight of the lipid. The Saponification of the examined oil was 190.7 ± 1.4 mg KOH/g oil (Alfawaz, 2004). Saponification value of pumpkin seeds oil is a range of 185-195 as reported by (Nichols & Sanderson, 2003) in rang 174-197. This value indicated that the pumpkin seed oil had fatty acids with a higher number of carbon atoms in comparison with coconut (248–265) and palm kernel (230–254) oils (Nichols & Sanderson, 2003).

Markovic and Bastic (1975) study found the saponification value of pumpkin seeds oil is a range of 185.5-195.3, however, it was lower than 200-218 range reported by Al-Khalifa (1996), 206 of El-Adawy and Taha (2001) and 201 of Tsaknis *et al.* (1997) and was higher than 132.3 reported by Younis *et al.* (2000) for *Cucurbita*

species. Furthermore, it fell in the range reported for olive, canola, corn, and sunflower oils (Nichols & Sanderson, 2003).

2.4.5 Iodine value of pumpkin seed oil

The iodine value of pumpkin seeds oil was 105.1 reported by Alfawaz (2004), and Tsaknis et al. (1997) found it 107.0, and the iodine value was 123.0 reported by Younis *et al*. (2000). According to Ardabili et al (2011), the pumpkin seed oil had an iodine value of 104.4 ± 0.0, indicating a high degree of unsaturation. This value was close to 103.2, 107.0, and 105.1 reported by, respectively, Lazos (1986), Tsaknis *et al.* (1997), and Alfawaz (2004), but higher than 80.0 that was indicated by Esuoso *et al.* (1998), and lower than 123.0 of Younis *et al.* (2000) and 116.0-133.4 of Markovic and Bastic (1975) for *Cucurbita* species. It also lied within the range reported for cottonseed, canola, rapeseed, and corn oils (Nichols & Sanderson, 2003).

2.5 Pumpkin seed oil and health

1. **Cardioprotective and Anti-hypertensive effect:** Pumpkin seeds play a crucial role in soothing vessels and lowering blood pressure. El-Mosallamy *et al* (2012) examined the effects of pumpkin seed oil treatment on chemically induced hypertension in rats. 40-100 mg/kg pumpkin seed oil was given once daily for six weeks. It was observed that the consumption of the oil considerably reduced the increased blood pressure caused by the chemical. The higher magnesium content in pumpkin seeds is credited to lower the risk of a heart attack. Abuelgassim and Al-showayman (2012) stated that rats induced with atherosclerosis were fed with pumpkin seeds for 37 days. HDL cholesterol was significantly increased in rats but also a 48% decrease in total cholesterol and a 79% reduction in LDL cholesterol was observed.

2. **Bone protection:** Pumpkin seed oil is a good source of minerals i.e. magnesium and phosphorous which optimize bone health and avert osteoporosis. Ryder *et al* (2005) assessed the relationship between magnesium intake and bone mineral density, a major factor in the development of osteoporosis, in over 2000 elderly men and women aged 70-79 yrs. After taking into account confounding factors of age, calcium intake, osteoporosis status, Body Mass Index, and physical activity, they concluded that higher intakes of magnesium were correlated with greater bone mineral density, particularly for Caucasian (white people of Europe) individuals. They believed that magnesium promotes an alkaline environment inside the bones, which had shown to be conducive to boost the bone mineral density.
3. **Easing arthritis:** Pumpkin seed oil has influential antioxidant properties that relieve inflammation related to arthritic symptoms. Fahim *et al* (1995) conducted an experiment in which rats were induced with arthritis showed significantly increased levels of inflammation which were reduced on the supplementation of rats with pumpkin seed oil; results that compared favourably to when the rats received the non-steroidal anti-inflammatory drug indomethacin. Furthermore, the indomethacin-supplemented rats experienced high levels of lipid peroxidation in the liver i.e. an indicator of liver injury, whereas the pumpkin seed oil supplemented group of rats experienced no side effects.
4. **Anxiety relief:** Hudson *et al* (2007) conducted a study that revealed that tryptophan was abundant in pumpkin seeds which can help to lessen anxiety. Tryptophan is converted into serotonin which is a hormone that enhances mood and promotes the healthiness of the brain. They investigated whether consuming

tryptophan-rich food could boost serotonin levels and reduce anxiety symptoms. They discovered that subjects with an anxiety disorder who consumed tryptophan-rich pumpkin seeds before an anxiety test showed better improvements in subjective and objective measures on the Endler Multidimensional Anxiety Scale compared with those who didn't consume pumpkin seeds.

5. **Hypolipidaemic effect**: Makni *et al* (2008) evaluated the effect of intake of a mixture of flax and pumpkin seeds in rats fed with a 1% cholesterol diet. A significant increase in monounsaturated and polyunsaturated fatty acids was observed in rats fed with pumpkin seed. Improved efficiency of the antioxidant defense system indicated the anti-atherogenic perspective of the seed mixture. Gossell-Williams *et al* (2008) examined the effect of pumpkin seed oil supplementation on cholesterol and blood pressure in rats. Both non-ovariectomized and ovariectomized rats were supplemented with corn oil or pumpkin seed oil for five days per week for twelve weeks (40mg/kg given orally). Blood analysis showed satisfactory lipid profile in the group of rats supplemented with pumpkin seed oil. Barakat and Mahmoud (2011) examined the potential of pumpkin seeds used alongside with flax seed or purslane seed on hyperlipidemia in rats fed with high cholesterol diet. An administration of two percent cholesterol diet significantly increased the total cholesterol, total lipids, and triacylglycerol in both the liver and serum of rats. The consumption of flax and pumpkin or purslane and pumpkin seed mixtures notably decreased the lipid parameters suggesting the hypolipidaemic perspective of the seed mixture.

6. **Anti-diabetic effect:** Makni *et al* (2011) observed that consumption of a mixture of pumpkin seeds and flax seeds reduced the increased levels of the plasma enzymes produced by the initiation of diabetes and caused a consequent revival towards normalization as compared to the control group animals. Its use in food on a daily basis may be efficient in the prevention of diabetes and its side effects. Teugwa *et al* (2013) evaluated the anti-diabetic effect of proteins obtained from several species of Cucurbitaceae, including *C. moschata*. The result showed that globulin is the most abundant protein found in pumpkin seeds which measured 295.11mg/g dry matter of extracted proteins and able to lower the blood sugar levels in rats with high blood sugar levels.
7. **Cancer management:** Consumption of pumpkin seeds has revealed extensive benefits in benign prostate hyperplasia i.e. enlargement of the prostate gland in men. Gossell-Williams *et al* (2006) studied the efficacy of pumpkin seed oil on rats with testosterone-induced prostate hyperplasia. During hyperplasia induction, pumpkin seed oil and corn oil (vehicle) were orally administered for 20 days. On the 21st day, rats were killed and their prostate was weighed. The induced increase in prostate size was repressed in rats fed with pumpkin seed oil i.e. 2mg/100g. The protective effect of pumpkin seed oil was considerable at the higher dose. The results showed that the utilization of pumpkin seed oil can help manage benign prostatic hyperplasia. Zaineddin *et al* (2012) given a food-frequency questionnaire to a vulnerable group of women. It was found that the eating of sunflower and pumpkin seeds was associated with an extensively reduced postmenopausal breast cancer threat.
8. **Bladder stone alleviation:** Lim (2012) reported that supplementation of pumpkin seeds in diet could be helpful to decrease the risk of bladder stones in

children and adolescents in Thailand. The results revealed that the longer supplementation period of pumpkin seeds showed better effects on bladder stone mitigation. Pumpkin seeds lowered oxalcrystalluria i.e. occurrence of calcium-oxalate crystals and calcium level but increased phosphorous, glycosaminoglycans, pyrophosphate, and potassium levels in urine as compared to orthophosphate supplementation. Pumpkin seeds provided high phosphorous levels and increased levels of inhibitors of crystal formation or aggregation which would consequently reduce the formation of bladder stones.

9. **Gynecological effect:** Phytoestrogens are plant metabolites similar to 17 beta-estradiol in structural and functional properties. They are renowned to lessen the risk of osteoporosis, heart disease, menopausal problems, and breast cancer (Zaineddin *et al* 2012). The pumpkin seed oil has been discovered to have a higher content of phytoestrogens as other plant sources like soybean, flaxseed, sesame, sunflower seed, etc. Gossell-Williams *et al* (2011) evaluated the credible beneficial effects of pumpkin seed oil on postmenopausal women. The randomized, double-blinded, and placebo-controlled study was conducted on thirty-five women who had undergone menopause naturally or due to surgery. The women consuming pumpkin seed oil showed a noteworthy increase in high-density lipoprotein and a considerable decrease in diastolic blood pressure. A decrease in the severity of hot flushes, frequent headaches, and joint pains was reported in the women utilizing pumpkin seed oil. The placebo group administered with wheat germ oil complained of more depression and emotional anxiety. The positive reaction of pumpkin seed oil administration implies further studies to find out menopause curative properties of pumpkin seeds.

CHAPTER THREE

METHODOLOGY

3.1 Sample Collection

The dried pumpkin seeds (*C. pepo* subsp. *pepo* var. Styriaca) were obtained from Upper West Region of Ghana. They were stored in a sealed vessel wrapped with a polyethylene bag at 4°C until analysis and oil extraction.

3.2 Equipment and materials

Several equipment and materials were used for this study. Some equipment and materials used for this study include; vacuum oven, stoppered conical flask, burette, retort stand.

3.3 Sample Analysis

3.3.1 Physico-chemical Analysis

3.3.1.1 Moisture Content determination

The moisture content of the sample was determined using the air oven method of AOAC (1990). Five grams (5 g) of pumpkin seed oil sample was put into a washed and dried moisture dish and placed into a vacuum oven and dried at 700^0C at a constant pressure of 70mm Hg for 5 hours. The samples were cooled in a desiccator and weighed (AOAC, 1990). The weight loss was obtained as the moisture content and is calculated as:

$$\% \text{ Moisture content} = \frac{W2-W3}{W2-W1} \times 100$$

Where; W1 = initial weight of empty moisture cans; W2 = weight of moisture cans + sample before drying; W3 = final weight of moisture cans + sample after drying.

3.3.1.2 Free Fatty Acid Determination

Five grams (5 g) of the well-mixed oil sample was weighed. A specified amount of 95% Ethanol/Ether (1:1) and phenolphthalein indicator was added to the pumpkin seed oil sample. A titration with 0.1N NaOH was done against the oil samples shaking constantly until pink color persists for 30 seconds (AOAC, 1990).

$$\%\text{FFA (as oleic)} = \frac{Vol.of\ NaOH\ (ml) \times Normality\ of\ NaOH \times 28.2mg}{\text{sample weight}}$$

3.3.1.3 Peroxide Value Determination

Five grams (5 g) of the oil samples were weighed into a dry 250ml stoppered conical flask. 10ml of Chloroform was added to the oil samples to dissolve the oil by swirling. 15ml of glacial acetic acid and 1ml fresh saturated aqueous solution of KI was added, stoppered the flask, and shake for 1in and place the flask for exactly one further minute in the dark. 75ml water was added and mixed and titrated with 0.01M Sodium thiosulphate solution using a soluble starch solution (1%) as an indicator. The flask was shaken vigorously during the titration to transfer the liberated iodine from the chloroform layer to the aqueous layer. The procedure was repeated on the blank sample and the peroxide value was then calculated (AOAC, 1990).

$$\text{Peroxide value} = \frac{(V - V_0)}{M} \text{ x } 10^3 mEq\ kg$$

V = Titre value of sample

Vo = Titre value of blank

T = Motality of thiosulphate solution

M = Weight of sample

3.3.1.4 Saponification Value Determination of Pumpkin Seed Oil

Two grams (2 g) of the oil sample were weighed into a flash and 25ml of 0.5N alcoholic KOH were added. The blank sample was prepared by putting 25ml of the alcoholic KOH in a similar flask. Both flasks were fit to condensers and contents were boiled for one hour, swirling the flask from time to time. The flasks were allowed to cool and then the condensers were washed with distilled water. The excess KOH was titrated with 0.5N HCl using phenolphthalein indicator and the saponification value was then calculated (AOAC, 1990).

$$\text{Saponification value} = \frac{28.05 \times (blank - sample\ titration)}{\text{weight of sample}}$$

3.3.1.5 Iodine Value Determination of Pumpkin Seed Oil

Two grams (2 g) of the oil sample was weighed into a dry flask and 20ml of carbon tetrachloride and 25ml of Wijs solution from a pipette were used to dissolve the oil. A blank sample was prepared alongside the sampling procedure. The flasks were stoppered and swirled to mix well the contents and were allowed to stand in a dark cupboard for 30mins at normal room temperature. After 30 mins the flask was removed and to each of the flasks 20ml of 15%, KI followed by 100ml of distilled water was added. The liberated iodine was slowly titrated with a 0.1N thiosulphate solution until the yellow color just disappears. At this stage, 2ml of the starch solution was added and a blue color that appears was discharged by the further slow titration of thiosulphate. Calculate the iodine value by the formula;

$$\text{Iodine value} = \frac{126.9 \times N \times (B - S)}{weight\ of\ sample}$$

Where B = Blank titration

S = Sample titration

N = the normality of Sodium thiosulphate solution

126.9 = Atomic weight of the iodine

3.4 Data analysis of Pumpkin Seed Oil

Data obtained were processed and analysed with the aid of Statistical Package for Social Sciences (SPSS) version 23.0. The results were organized into simple Tables using Mean and standard deviation to make the presentation of the information easy for understanding.

CHAPTER FOUR

RESULTS AND DISCUSSION

4.1 Physicochemical Characterization of Pumpkin Seed Oil

The results obtained for various physicochemical characteristics such as free fatty acid, acid value, peroxide value saponification value, and iodine value of pumpkin seed oil are shown in Table 4.1.

Table 4. 1: Quality characteristics of pumpkin seed oil

Parameter	Mean	Standard deviation
Free fatty Acid (% as oleic acid)	0.06	0.00
Acid Value (mg KOH/g of oil)	0.12	0.00
Peroxide Value (ml eq O_2/kg oil)	9.69	0.12
Saponification Value (mg KOH/g of oil)	44.19	0.80
Iodine Value (g of I_2/100 g oil)	10.62	0.33

4.1.1 Free fatty acid content of Pumpkin Seed oil

As presented in Table 4.1, the pumpkin seed had a free fatty acid value of 0.06%. This value was lower than that value (0.39% as oleic acid) reported by Gohari *et al*. (2011) and lower than the value as 0.04% as oleic acid reported by Tsaknis *et al*. (1997). On the other hand, Ardabili et al. (2011) considered the content of free fatty acids (0.39 ± 0.01 % as oleic acid). Azeman et al., (2014) stated that the high content of free fatty acids in oil affects the quality of the oil and leads to various health and environmental issues. The maximum free fatty acids content set by the Oil Refiners Association of Malaysia is 5% and < 0.1 % in refined bleached deodorized oil. This is an indication that the pumpkin seed oil is of good quality and better health effects on the consumer. Free fatty acids are products of hydrolysis and therefore it is expected

that the higher the lower the free fatty acid content of pumpkin seed oil the lower the moisture content of the oil (Azeman et al., 2014).

4.1.2 Acid value

The acid value of pumpkin seeds oil was shown in Table 4.1. It was revealed that the acid value of pumpkin seed was 0.12 ± 0.00 mgKOH/g. This value is lower than the value 0.86 reported by Imad (2003) and 0.78 reported by Gohari *et al*. (2011). Commission expressed the permitted maximum acid values of 10 and 4 mg KOH/g oil for virgin palm and coconut oils, respectively (Alfawaz, 2004). It has been shown that oils become rancid when the peroxide value ranges from 20.0 to 40.0 meq O2/kg oil (Ajayi *et al.,* 2006). High acid value (4-12) for common pumpkin seeds oil and high acid value for Indian and American varieties where they gave values 10.1 and 12.40 respectively (Gohari *et al*., 2011). Therefore, considering that the oil studied was unrefined and its initial quality indicators were within the reported limits, the pumpkin seed oil can be regarded as an edible oil with good quality.

4.1.3 Peroxide value

The peroxide value of pumpkin seeds oil was 9.69± 0.12 meq o2/kg oil. Detection of peroxide value gave the initial evidence of rancidity in unsaturated fats and oils (Marco *et al* 2015). Peroxide value is expressed in units of milliequivalents per kg. The extracted pumpkin seed oil had an acceptable initial quality. This value was higher than the value of 8.5 reported by Mohammed (2004). Bialek *et al* (2016) determined the peroxide value of pumpkin seed oil which was 2.89 meq/kg. Vujasinovic *et al* (2010) found that the peroxide value of naked pumpkin seed and pumpkin seed with the hull as 2.95 and 5.04 mmol/kg. Srbinoska *et al* (2012) studied the peroxide value of pumpkin seed whole and pumpkin seed kernel of two different

varieties (*Cucurbita maxima* and *Cucurbita pepo*). Results showed that the peroxide value of whole seed and kernel of *Cucurbita maxima* was 4.93 and 4.26 meq/kg extract whereas, in *Cucurbita pepo*, it was 6.06 meq/kg extract for whole seed and 5.70 meq/kg extract for seed kernel.

Tsaknis *et al* (1997) analyzed the characteristics of crude and purified pumpkin seed oil in which peroxide value of crude and pumpkin seed oil was obtained as 9.20 and 9.04meq/kg. Adeel *et al* (2014) found that the peroxide value of pumpkin seed oil was 6.74meq 2/kg oil. More peroxide value is harmful to the shelf life of the oil. Exposure of peroxide provides initial evidence of rancidity and deterioration of pumpkin seed oil caused by unsaturated fats and oils. The peroxide value of pumpkin seeds oil in range 10.85-11.47(Gohari *et al*, 2011). The Codex Alimentations Commission expressed the permitted maximum acid values of 10 and 4 mg KOH/g oil for virgin palm and coconut oils, respectively (Alfawaz, 2004). It has been shown that oils become rancid when the peroxide value ranges from 20.0 to 40.0 meq O_2/kg oil (Ajayi *et al*., 2006). On the other hand, according to the Codex Alimentary Commission, the peroxide value for unrefined olive oil may maximum of 20meq O_2/kg oil (Markovic & Bastic, 1975). Therefore, considering that the oil studied was unrefined and its initial quality indicators were within the reported limits, the pumpkin seed oil can be regarded as an edible oil with good quality.

4.1.4 Saponification value

The saponification value of the oil was 44.19 ±0.80 as shown in Table 4.1. This value is lower than the range 174-197 reported for the pumpkin seeds oil by (Nichols & Sanderson, 2003). And also below in range 185.5-195.3 reported by Markovic and Bastic (1975), however, it was lower than 200-218 range reported by Al-Khalifa (1996), 206 of El-Adawy and Taha (2001) and 201 of Tsaknis *et al*. (1997) and was higher than

132.3 reported by Younis *et al*. (2000). Moreover, it is lower than the value of 199.2 reported by Butt (1995).

4.1.5 Iodine value

The pumpkin seed oil had an iodine value was 10.62 ± 0.33 (g of I_2/100g oil) shown in Table 4.3, indicating a low degree of unsaturation. This value falls below 103.2 and 105.1 reported by, respectively, Lazos (1986) and Alfawaz (2004). The value was still lower than 80.0 that was indicated by Esuoso *et al*. (1998), and lower than 123.0 of Younis *et al*. (2000). Tsaknis et al. (1997) found 107.0 iodine value, and the iodine value was 123.0 reported by Younis *et al*. (2000). According to Ardabili et al (2011), the pumpkin seed oil had an iodine value of 104.4 ± 0.0, indicating a high degree of unsaturation. Markovic and Bastic (1975) attained an iodine value of 116.0-133.4 for *Cucurbita* species. It also lied within the range reported for cottonseed, canola, rapeseed, and corn oils (Nichols & Sanderson, 2003).

CHAPTER FIVE

CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

The pumpkin seed oil has many health benefits derived from its consumption. The physicochemical properties analysed for the pumpkin seed oil samples included; free fatty acid, acid value, peroxide value saponification value, and iodine value. The study revealed that pumpkin seed had a free fatty acid value of 0.06% that helps maintain healthy blood vessels, nerves, and tissues. The acid value of pumpkin seeds oil was 0.12 ± 0.00 mgKOH/g. Considering that the oil studied was unrefined and its initial quality indicators were within the reported limits, the pumpkin seed oil can be regarded as an edible oil with good quality.

The peroxide value of pumpkin seeds oil was 9.69± 0.12 meq o2/kg oil. The saponification value of the oil was 44.19 ±0.80. Also, the pumpkin seed oil had an iodine value was 10.62 ± 0.33 (g of I_2/100g oil). It can be concluded that pumpkin seed has a high nutritional value and can provide good quality oil. These seeds may also serve as constituents of minerals to humans through the development of various value-added products. Considering the yield of oil and its free fatty acid profile of seeds similar to sesame, sunflower, peanuts, and soyabean oils, the pumpkin seed oil can be considered as a new and valuable source of edible oil according to its physicochemical properties.

5.2 Recommendation

1. More attention and care should be taken for pumpkin cultivation to produce seeds for oil production in Ghana.

2. The ministry of health should encourage the consumption of pumpkin seed oil as frequent consumption of pumpkin seed oil can increase better nutrition and health.
3. Imparting education to women from different villages in the Upper West Region of Ghana regarding the importance of pumpkin seeds which are commonly discarded as waste and encourage them to use these seeds in various means.

5.3 Suggestion for Further Studies

Similar studies can be conducted on pumpkin seed grown in different part of the country since this study was conducted in the Upper West Region of Ghana. Further research is required to study the complete potential of pumpkin seed flour. In addition study is required on the microbial properties of the pumpkin seed.

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