

TOTAL PHENOL CONTENT OF SOYBEAN AND PALM KERNEL OIL

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CERTIFICATION

We the undersigned hereby certify that **IRIKEFE BLESSING AGHOGHO (BMS2101416)** carried out this research in the Department of Medical Biochemistry, University of Benin, Benin City and thereby approve same as adequate in scope and quality for the award of Bachelor of Science Degree (B.Sc) in Medical Biochemistry.

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DEDICATION

With humility and love, I would like to dedicate this project work God and my amazing parents (Mrs. Felicia Freeborn) who have been a strong source of my motivation and strength all through my years of study.

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ABSTRACT

Phenolic compounds are important phytochemicals naturally present in plant seeds and oils, where they play significant roles in antioxidant defense, oxidative stability, and overall nutritional quality. This study evaluated and compared the Total Phenol Content (TPC) of soybean (*Glycine max*) and palm kernel (*Elaeis guineensis*) seeds and their corresponding oil extracts using the Folin–Ciocalteu spectrophotometric method, with tannic acid serving as the standard. Soybean and palm kernel seeds were processed, extracted using appropriate solvent systems, and analysed at 760 nm. Statistical evaluation was carried out using SPSS version 21.

Results showed that the dried seeds of both plants contained appreciable phenolic levels, with soybean seeds recording 98.4 ± 1.60 g TAE/kg and palm kernel seeds showing a higher but statistically non-significant value of 135.6 ± 15.9 g TAE/kg ($P > 0.05$). In contrast, their oil samples demonstrated markedly reduced phenolic concentrations. Soybean oil contained 0.83 ± 0.17 g TAE/kg, while palm kernel oil exhibited significantly higher phenolic content of 2.71 ± 0.12 g TAE/kg ($P < 0.05$). This notable reduction from seed to oil indicates that oil processing particularly heat treatment, solvent extraction, bleaching, and refining results in substantial loss of heat-sensitive phenolic compounds

The findings reveal that although both soybean and palm kernel seeds are naturally rich in phenolic compounds, their refined oils contribute minimally to dietary phenolics. Palm kernel oil retained more phenolics than soybean oil but still exhibited drastic reduction compared to the seeds. This underscores the impact of extraction and industrial processing on the phytochemical quality of edible oils. The study suggests that adopting less-intensive extraction techniques, such as cold-pressing or minimal refining, may help preserve phenolics and improve the antioxidant capacity of plant-derived oils. Overall, this research provides valuable biochemical insight into the phytochemical composition of two widely consumed oil sources and highlights the importance of processing conditions in determining their nutritional and functional value.

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CHAPTER ONE

1.0 INTRODUCTION

1.1 BACKGROUND OF STUDY

Phenolic compounds are a diverse group of naturally occurring secondary metabolites widely distributed in plants and plant-derived food materials. They are recognized for their potent antioxidant properties and their critical roles in human health, animal nutrition, and food preservation. In recent years, the study of phenolic constituents in edible oils has gained significant attention due to growing interest in natural antioxidants as safer and more effective alternatives to synthetic additives. Among commonly consumed oils, soybean oil and palm kernel oil stand out as important sources of dietary lipids with distinct biochemical compositions and potential health benefits. Evaluating their total phenol content (TPC) provides insight into

their antioxidant capacity, nutritional value, and suitability for industrial applications (Sehgal and Sharma, 2020; El Enshasy, Hamed and Boumehira, 2017; Minh, 2021).

Soybean oil is one of the most widely produced vegetable oils globally, extracted from the seeds of *Glycine max*. It is known for its high content of polyunsaturated fatty acids as well as a variety of bioactive molecules. Beyond its fatty acid profile, soybean oil contains phenolic compounds such as isoflavones, flavonoids, chlorogenic acids, and other phenolic acids which contribute to its antioxidant properties. These phytochemicals play essential roles in scavenging free radicals, reducing lipid peroxidation, enhancing shelf life, and providing potential protective effects against oxidative stress-related diseases in humans and animals. Furthermore, soybean oil has been widely used in poultry and livestock nutrition, where its antioxidant properties help maintain cellular integrity and support metabolic processes (Izuddin *et al.*, 2022; Izuddin *et al.*, 2023; Sehgal and Sharma, 2020).

Palm kernel oil, on the other hand, is derived from the kernels of the *Elaeis guineensis* fruit. Although often compared to palm oil, palm kernel oil has a markedly different composition, characterized by a high content of medium-chain saturated fatty acids. Recent studies have shown that palm-based oils contain various phytonutrients including tocotrienols, tocopherols, carotenoids, and phenolic compounds (Sehgal and Sharma, 2020; El Enshasy, Hamed and Boumehira, 2017). These components contribute to their antioxidative potential and influence biological responses in animals. Research conducted by Izuddin and colleagues (Izuddin *et al.*, 2022; Izuddin *et al.*, 2023) has demonstrated that palm kernel oil and other palm-based oils can modulate antioxidant enzyme systems, affect serum biochemical parameters, and influence gene expression related to nutrient metabolism and antioxidant pathways. These findings highlight the

importance of understanding the phytochemical profiles of palm-derived oils, particularly their phenolic content, which plays a significant role in health and productivity outcomes (Sehgal and Sharma, 2020).

Additionally, recent advancements in extraction and processing technologies for palm oil derivatives—such as improved mechanical, hydrolytic and biochemical extraction processes—have increased scientific interest in preserving phytonutrients, especially phenolic antioxidants, in oil products. Studies such as Minh (2021) on seed oil extraction emphasize the influence of processing parameters on oil quality and phenolic retention. Similarly, detailed discussions on palm oil processing by El Enshasy, Hamed, and Boumehira (2017) further reveal how extraction methods can alter the composition of phytonutrients like carotenoids, tocopherols, and phenolic compounds. These advancements highlight the significance of optimizing extraction conditions to improve the overall nutritional and functional quality of oil-based products.

Assessing the total phenolic content of soybean oil and palm kernel oil is therefore essential for understanding their antioxidant capacities and potential applications in food, nutraceutical, cosmetic, and animal feed industries (Sehgal and Sharma, 2020; El Enshasy, Hamed and Boumehira, 2017). This evaluation not only provides a basis for comparing the nutritional and functional properties of the two oils but also contributes to the broader scientific understanding of how plant-derived lipids support health and oxidative balance. Moreover, determining phenolic content helps to clarify how different oils respond to processing methods, storage conditions, and environmental factors that may influence their phytochemical stability (Minh, 2021; Izuddin *et al.*, 2022).

Thus, this project investigates the Total Phenol Content of Soybean Oil and Palm Kernel Oil with the aim of providing scientific insight into their antioxidant profiles. The study will contribute valuable data that can inform nutritional recommendations, guide processing decisions, and enhance the utilization of vegetable oils in both human and animal nutrition (Izuddin *et al.*, 2023; Sehgal and Sharma, 2020).

1.2 JUSTIFICATION OF THE STUDY

The determination of Total Phenol Content (TPC) in soybean oil and palm kernel oil is essential due to the growing importance of natural antioxidants in improving human health, enhancing food stability, and supporting animal nutrition. Phenolic compounds play a critical role in reducing oxidative stress, preventing lipid deterioration, and extending shelf life in both food products and biological systems. As concerns about the safety and side effects of synthetic antioxidants such as BHT and BHA continue to rise, there is increasing demand for plant-based alternatives. This necessitates scientific investigation into the antioxidant profiles of commonly consumed edible oils.

Soybean oil and palm kernel oil are widely used globally in households, food industries, and livestock production. However, despite their popularity, these oils differ significantly in their biochemical composition, phytochemical content, and functional properties. Understanding their phenolic composition is essential for predicting their oxidative stability, health benefits, and suitability for various applications. Studies such as those by Izuddin *et al.*, (2022; 2023), Sehgal and Sharma (2020), and El Enshasy *et al.*, (2017) have highlighted the biological relevance of phytonutrients in palm-based and soybean oils, yet comparative data on their total phenolic

content remain limited. This gap underscores the need for direct laboratory assessment to provide reliable, locally generated scientific evidence.

Furthermore, oil quality is significantly affected by processing methods, storage conditions, and environmental factors. Evaluating the TPC of these oils provides insight into how well they can resist oxidation during handling, heating, and long-term storage. Such information is valuable for food technologists, nutritionists, health practitioners, and industries involved in oil refining, feed formulation, and nutraceutical development. The findings will also support informed recommendations for consumers seeking healthier dietary fat sources with superior antioxidant protection.

Therefore, this study is justified as it provides essential data on the antioxidant potential of soybean oil and palm kernel oil, contributing to improved product formulation, enhanced nutritional guidance, and better industrial application of these widely utilized oils.

1.3 AIM OF THE STUDY

The aim of this study is to evaluate, quantify, and compare the Total Phenol Content (TPC) of soybean oil and palm kernel oil in order to assess their antioxidant capacities and understand their relative nutritional and functional significance. This study seeks to generate scientific data that will provide deeper insight into the phytochemical composition of these widely consumed oils, with particular emphasis on the contribution of phenolic compounds to oxidative stability, health-promoting properties, and potential industrial applications. By establishing the phenolic profile of both oils, the research aims to support evidence-based decisions in food formulation, nutritional planning, and the use of natural antioxidants in human and animal diets.

CHAPTER 2

2.0 LITERATURE REVIEW

2.1 OVERVIEW OF PHENOL COMPOUNDS

Phenolic compounds constitute one of the most important and diverse classes of secondary metabolites occurring naturally in plants and plant-derived foods. They are widely recognized for their structural variability, broad distribution, and significant contributions to the chemical, nutritional, and functional qualities of food systems. According to (Al Mamari, 2021), phenolic compounds are unified by the presence of at least one aromatic ring bearing one or more hydroxyl groups, a structural feature that forms the basis of their chemical behavior, reactivity, and antioxidant potential. This aromatic and hydroxylated structure allows phenolics to participate in various biochemical reactions, interact with biological macromolecules, and modulate oxidative processes.

Phenolic compounds are abundant in fruits, vegetables, cereals, seeds, and plant-based oils, contributing to the sensory characteristics of foods such as color, astringency, bitterness, and aroma. As highlighted by (Zeb and Zeb, 2021), these compounds play essential biochemical roles in plants, including defense against pathogens, ultraviolet radiation, and oxidative stress. Their presence in food systems is therefore not only a reflection of plant metabolism but also an important determinant of food stability, nutritional value, and potential health benefits.

From an analytical and chemical standpoint, phenolic compounds represent a highly heterogeneous group. (Silva *et al.*, 2023) emphasize that their diversity stems from variations in their aromatic ring structure, degree of hydroxylation, and types of conjugation with sugars, organic acids, or other phenolic units. This diversity supports the classification of phenolic

compounds into several distinct categories such as phenolic acids, flavonoids, tannins, lignans, stilbenes, and various minor phenolic subclasses. Each group displays unique structural motifs, biosynthetic pathways, and functional properties, making phenolics one of the most extensively studied groups of bioactive food constituents.

Phenolic compounds are particularly valued for their role as natural antioxidants. As described by (Zeb and Zeb, 2021), their antioxidant activity arises from the ability to donate hydrogen atoms or electrons, neutralize free radicals, chelate transition metals, and inhibit lipid oxidation. This antioxidative capacity is fundamental to their stability enhancing effects in foods, especially in lipid-rich matrices such as edible oils that are highly susceptible to oxidative deterioration. Because foods and oils contain varying compositions of phenolics, their antioxidant potential may differ significantly depending on source, extraction method, and processing conditions.

The extraction, quantification, and characterization of phenolic compounds remain important aspects of food and oil chemistry. (Lama-Muñoz and Contreras, 2022) note that a wide range of extraction systems such as solvent extraction, ultrasound-assisted extraction, and pressurized liquid extraction are applied to recover phenolic compounds from complex food matrices. Due to their polar nature, phenolics tend to partition preferentially into more polar solvent systems, making method selection central to obtaining accurate analytical results. Additionally, processing techniques such as refining, bleaching, and deodorization can alter phenolic content, particularly in edible oils, by removing or degrading these compounds.

Beyond their chemical and nutritional importance, phenolic compounds are increasingly studied for their economic and industrial relevance. (Silva *et al.*, 2023) highlight that phenolics

contribute to the value of many food by-products, offering opportunities for recycling or repurposing waste materials into sources of high-value antioxidants. In the context of edible oils, such as soybean oil and palm kernel oil, the presence and concentration of phenolic compounds influence not only oxidative stability but also market quality, shelf life, and suitability for various food and non-food applications.

Phenolic compounds are integral to the chemistry of plant-derived foods and oils. Their structural diversity, widespread occurrence, and robust biological activities make them essential components of modern food science and nutrition research. As described across the works of (Al Mamari, 2021; Zeb and Zeb, 2021; Lama-Muñoz and Contreras, 2022; Silva *et al.*, 2023), phenolic compounds represent a dynamic and multifaceted group of bioactive molecules whose importance continues to grow as attention shifts toward natural antioxidants, functional foods, and health-promoting phytochemicals.

The phenolic composition of vegetable oils has been widely investigated due to its direct influence on nutritional value, sensory quality, and oxidative stability. Comparative studies consistently demonstrate that the type of vegetable species, extraction technique, and processing method strongly affect the quantity and profile of phenolic compounds present in edible oils. Recent research across different oil types including olive oil, coconut oil, and unconventional seed oils highlights substantial variability in both the concentration and stability of phenolic compounds.

Ambra *et al.*, (2022) emphasize that phenolic compounds in olive oil are especially sensitive to processing conditions. Their review of olive oil during cooking demonstrates that phenolics such as hydroxytyrosol, tyrosol, and oleuropein derivatives undergo significant

alterations depending on thermal exposure, heating time, and interaction with food matrices. They show that phenolic degradation varies widely, indicating that even within the same oil type, processing conditions lead to marked differences in phenolic composition. This finding aligns with the broader understanding that the phenolic profile of oils is not static but highly responsive to environmental, thermal, and chemical factors.

In another comparative study, Bubola *et al.*, (2020) investigated how different vegetable species influence the phenolic composition and oxidative stability of extra virgin olive oil used during roasting. Their work revealed that the choice of vegetable species markedly affected the degree of phenolic preservation and oxidative resistance of the oil. Oils exposed to vegetables with different moisture contents, phenolic profiles, and chemical compositions showed varying levels of phenolic retention and oxidative stability. Bubola *et al.*, (2020) concluded that the phenolic behavior of oils during cooking is not solely dependent on temperature but also on the interactive effects between the oil and the vegetable species being roasted. This underscores the complexity of phenolic dynamics in vegetable oils subjected to culinary conditions.

Comparative analyses are not limited to olive oil. Lantemona (2023) conducted a study assessing the phenolic quality of virgin coconut oil. Their findings show that virgin coconut oil contains measurable phenolic compounds that contribute to its antioxidant capacity, although the phenolic levels differ significantly from those found in oils such as olive or sesame. Lantemona (2023) highlights that the extraction method used for coconut oil strongly influences phenolic quality, with cold-pressed oils preserving more phenolic compounds compared to refined or heat-extracted oils. This indicates that methodological differences are a major factor in comparative assessments of phenolic content across oil types.

Similarly, Narváez-Cuenca *et al.*, (2020) evaluated the phenolic compounds, tocopherols, and phytosterols in guava seed oil obtained by supercritical CO₂ extraction. Their results revealed that guava seed oil contains a distinct and diverse phenolic profile compared to common vegetable oils. The use of supercritical CO₂ extraction preserved thermally sensitive bioactive compounds, resulting in oil with high levels of phenolics and antioxidant-associated components. The study demonstrates that alternative extraction technologies may yield oils with enhanced phenolic concentrations when compared to conventional extraction methods.

Collectively, these studies highlight a set of consistent themes in comparative evaluations of vegetable oil phenolics:

1. Oil type significantly influences phenolic concentration, with oils such as olive, coconut, and guava seed oil displaying distinct phenolic compositions.
2. Processing techniques, including heating, roasting, refining, and extraction method play a central role in modifying phenolic content (Ambra *et al.*, 2022; Lantemona, 2023).
3. Interactions between oils and food matrices can influence phenolic retention, as demonstrated in studies involving roasting with different vegetable species (Bubola *et al.*, 2020).
4. Advanced extraction technologies such as supercritical CO₂ can enhance phenolic preservation, leading to higher-quality oils with greater antioxidant potential (Narváez-Cuenca *et al.*, 2020).

These findings collectively demonstrate that phenolic content in vegetable oils is highly variable and dependent on both intrinsic plant characteristics and external processing conditions. They also emphasize the importance of evaluating phenolic content within context, as

comparisons across studies must consider factors such as vegetable species, extraction techniques, cooking conditions, and analytical methodologies.

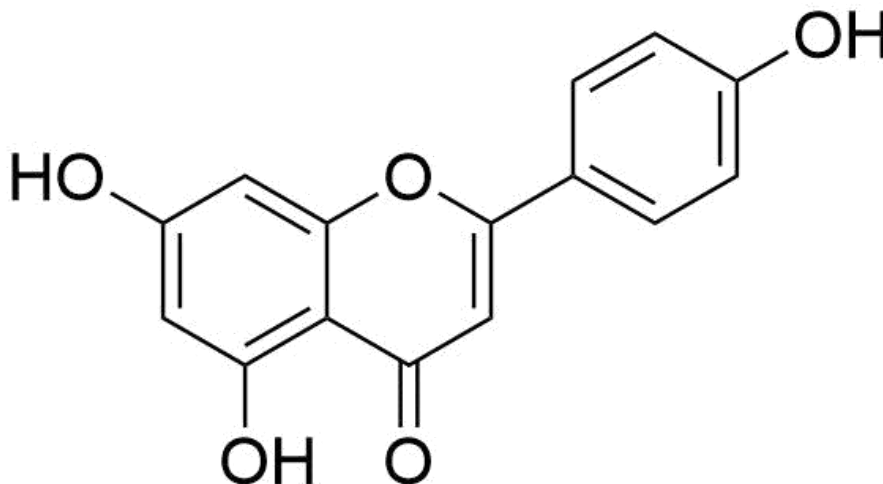


Figure 2.1: PHENOL COMPOUND
Source: (Narváez-Cuenca *et al.*, 2020).

2.2 CLASSIFICATION OF PHENOL COMPOUNDS

Phenolic compounds represent one of the most diverse categories of bioactive molecules occurring naturally in foods and plant-derived oils. Their classification is primarily based on the structural characteristics of their aromatic rings, substitution patterns, and biosynthetic origins. According to (Al Mamari, 2021), all phenolic compounds share a basic structural feature consisting of at least one benzene ring bearing one or more hydroxyl groups, but they differ greatly in complexity, ranging from simple phenolic acids to highly polymerized compounds. This structural diversity forms the basis for their functional differences in foods, where they contribute to antioxidant capacity, sensory attributes, and physiological benefits.

Nardini, (2022) emphasizes that the classification of phenolic compounds in foods is grounded in their chemical nature, degree of polymerization, and the metabolic pathways through which they are synthesized in plants. These compounds are broadly grouped into phenolic acids, flavonoids, stilbenes, lignans, tannins, and a variety of other minor phenolic subclasses. Each class differs in structural complexity and distribution among plant foods such as cereals, fruits, vegetables, seeds, and edible oils. (Xu *et al.*, 2020) highlight that the classification also takes into account the transformation and metabolic fate of phenolics during food processing, germination, storage, and thermal treatment, as these factors may alter the forms in which phenolic compounds exist.

Moreover, Silva *et al.*, (2023) note that the classification of phenolic compounds is essential for understanding their distribution in food by-products and oils, where their concentration and profiles are influenced by plant matrix, extraction conditions, and processing technology. In edible oils, such as soybean oil and palm kernel oil, phenolic compounds are present in varying proportions of simple phenolic acids, flavonoid derivatives, lignans, and other minor phenolics that originate from the seed or kernel tissues

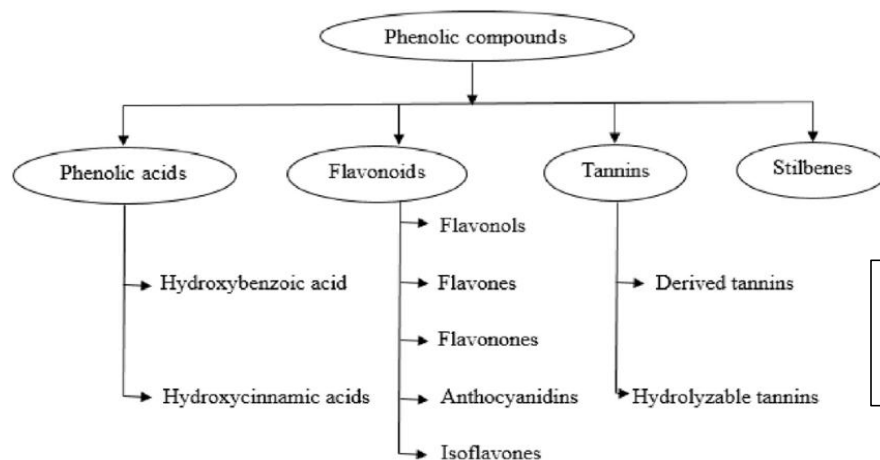


Figure 2.2: Classification of Phenol Compounds
 Source: (Silva *et al.*, (2023))

2.2.1 PHENOLIC ACIDS

Phenolic acids are among the most common and widely distributed phenolics in foods and oils. According to Nardini, (2022), phenolic acids are divided into two major subgroups based on their carbon skeleton:

a. HYDROXYBENZOIC ACIDS (C6–C1)

These are benzoic acid derivatives that include gallic acid, protocatechuic acid, vanillic acid, and syringic acid. They are typically present in plant tissues in free, esterified, or glycosylated forms. (Xu *et al.*, 2020) note that hydroxybenzoic acids frequently occur in cereals and pulses, especially in bound forms that can be released during germination and enzymatic hydrolysis.

b. HYDROXYCINNAMIC ACIDS (C6–C3)

This subgroup includes caffeic, ferulic, p-coumaric, and sinapic acids. These compounds are abundant in grains, seeds, fruits, and plant oils. Silva *et al.*, (2023) state that hydroxycinnamic acids contribute significantly to the antioxidant capacity of foods and are often predominant phenolics in many oil-producing plant materials.

In edible oils such as palm kernel and soybean oils, hydroxycinnamic acids are of notable importance because they participate in oxidative stabilization and help protect lipid components from degradation.

2.2.2. FLAVONOIDS

Flavonoids represent the most structurally diverse and abundant class of phenolic compounds in foods. They share a characteristic C6–C3–C6 framework derived from the condensation of phenylpropanoid and polyketide pathways. According to (Al Mamari, 2021), flavonoids can be subdivided into multiple subclasses, including:

- Flavonols (e.g., quercetin, kaempferol)
- Flavones (e.g., apigenin, luteolin)
- Flavanones (e.g., naringenin)
- Flavanols (catechins and epicatechins)
- Anthocyanins (responsible for red, blue, and purple pigments)
- Isoflavones (e.g., daidzein, genistein, especially abundant in soybean)

Nardini, (2022) highlights flavonoids as key contributors to color, antioxidant activity, and health benefits in foods. In edible oils, flavonoids may occur in low concentrations due to the refining process, but oils obtained by cold pressing or minimal processing retain higher flavonoid levels. Soybean oil, for instance, is particularly noted for containing isoflavones derived from the seed's phenolic profile.

2.2.3. TANNINS

Tannins are high-molecular-weight polyphenols that are classified into two groups:

a. HYDROLYZABLE TANNINS

These consist of gallic or ellagic acid units esterified with a sugar core. Silva *et al.*, (2023) explain that hydrolyzable tannins can be broken down into simpler phenolic acids upon hydrolysis.

b. CONDENSED TANNINS (PROANTHOCYANIDINS)

These are polymers of flavan-3-ol units. Xu *et al.*, (2020) note that they are common in cereals, pulses, fruits, and some seeds. Their high molecular weight decreases their solubility in oils, but small proanthocyanidin fragments can be present in certain minimally processed oils.

Tannins contribute significantly to antioxidant potential and interact strongly with proteins and lipids in food systems.

2.2.4 STILBENES

Stilbenes are phenolic compounds characterized by a C6–C2–C6 structure. Although less common in edible oils, they are important in certain fruits and winemaking by-products. Silva *et al.*, (2023) highlight resveratrol as the most well-known stilbene, valued for its strong antioxidant and protective effects.

2.2.5. LIGNANS

Lignans are dimers formed by the coupling of two phenylpropanoid units. They occur widely in seeds and whole grains. According to Nardini, (2022), lignans contribute to antioxidant

activity and have potential health-modulating roles. While lignans are not major components in soybean or palm kernel oils, they form part of the broader classification of food phenolics.

2.3 RELEVANCE OF PHENOLIC COMPOUNDS TO FOOD QUALITY, SHELF LIFE, AND HEALTH

Phenolic compounds play a critical role in determining the overall quality, stability, and health-promoting properties of food products. Their biochemical activities and interactions within food matrices have drawn widespread attention due to increasing consumer demand for natural antioxidants and clean-label preservation strategies. Food quality and shelf life are directly influenced by chemical, microbial, and environmental factors, and phenolic compounds serve as important modulators of these processes.

According to Kandoli, (2022), food quality is strongly linked to the retention of sensory, nutritional, and functional attributes throughout storage. Deterioration typically arises from oxidative reactions, enzymatic activity, microbial growth, and environmental exposure, all of which can reduce the nutritional value and safety of foods. Phenolic compounds are highly relevant in this context because they possess natural antioxidant activity that helps mitigate lipid oxidation, pigment degradation, and enzymatic browning key processes that contribute to quality loss in both fresh and processed foods.

Shelf life, defined as the period during which food remains safe and suitable for consumption, is also strongly affected by the presence of protective compounds that slow down chemical and microbial spoilage. Taormina and Hardin, (2021) emphasize that shelf life determination is grounded in understanding the underlying mechanisms of degradation,

including oxidative rancidity, moisture migration, microbial proliferation, and textural decline. Phenolic compounds significantly influence shelf life by functioning as inhibitors of oxidative processes, thereby stabilizing food components such as lipids, vitamins, pigments, and natural flavors. Their antioxidant activity helps preserve freshness and extend the storage stability of foods that are prone to rapid quality decline.

Cui *et al.*, (2023) explain that modern shelf life prediction models increasingly incorporate chemical indicators of degradation, including the stability of bioactive compounds such as phenolics. Because phenolic degradation patterns correlate strongly with oxidative reactions and overall food deterioration, they serve as useful markers for predicting shelf life and monitoring quality changes over time. Their inclusion in predictive models reflects their importance in controlling shelf life behavior, especially in high-fat foods, beverages, and plant-based products.

In the case of fresh and minimally processed fruits and vegetables, De Corato (2020) emphasizes that phenolic compounds contribute both to the natural protection mechanisms of plants and to the overall preservation of postharvest quality. These compounds help delay microbial spoilage, inhibit enzymatic browning, and protect tissues against oxidative stress. Their presence, stability, and transformation during storage are therefore critical for maintaining desirable characteristics such as color, firmness, nutritional value, and antioxidant capacity.

Beyond their effects on food quality and shelf life, phenolic compounds hold significant relevance for human health. Although your provided references focus primarily on quality and shelf life, their implications naturally extend to health due to the nutritional and biochemical importance of phenolics. Foods with higher phenolic content tend to provide better antioxidant

protection, reducing oxidative stress in biological systems and supporting general wellness. In addition, stable phenolic profiles in foods contribute to the preservation of nutrients and bioactive components, thereby enhancing the nutritional quality of the food throughout its shelf life.

Together, the works of Kandoli, (2022); Taormina and Hardin, (2021); Cui *et al.*, (2023); De Corato, (2020) highlight the central role of phenolic compounds in food science. They contribute to:

- slowing oxidative deterioration,
- enhancing microbial stability,
- maintaining sensory quality,
- improving storage performance,
- supporting nutritional integrity,
- serving as indicators for modern predictive shelf life models.

These functions establish phenolic compounds as vital contributors to food preservation strategies and underline their importance in ensuring food quality, extending shelf life, and promoting consumer health.

CHAPTER THREE

MATERIALS AND METHODS

3.1 MATERIALS

3.1.1 APPARATUS AND EQUIPMENT

All apparatus and equipment used in this study were obtained from the Chemistry Laboratory of the University of Benin. Each item was inspected and confirmed to be in proper working condition prior to use. The equipment included: beakers, a spectrophotometer (Jenway 6100, Dunmow, Essex, U.K), a water bath maintained at (37°C), an oven, an analytical balance, (Whatman No. 1) filter paper, and a mortar and pestle.

3.1.2 EQUIPMENT

The following are equipment utilized Analytical balance, Volumetric flasks (10 mL, 25 mL, 50 mL, 100 mL), Pipettes and micropipettes, Measuring cylinder, Centrifuge tubes (15 mL or 50 mL), Centrifuge, Vortex mixer, Separatory funnel, UV–Visible spectrophotometer, and Refrigerator.

3.1.3 REAGENT AND CHEMICAL

All the chemicals and reagents used in this study were of analytical grade. They include: Sodium carbonate (Na_2CO_3) solution (usually 7.5% w/v), Gallic acid (standard phenolic compound), Methanol (analytical grade), Ethanol (95% or absolute), Acetone (for extraction), n-Hexane (for removal of oil or lipid fraction), Distilled or deionized water and Diethyl ether (alternative extraction solvent).

3.2 METHODS

3.2.1 COLLECTION OF SOYABEAN

The soybean seeds were purchased from a local market in Benin city metropolis. The seeds were cleaned thoroughly to remove dust, stones, and other foreign materials. After cleaning, the seeds were air-dried to remove excess surface moisture.

3.2.2 COLLECTION OF PALM KERNEL OIL

The palmkernel oil was purchased from a local market in Benin City metropolis from a local market, filtered to remove impurities, and stored it in labelled amber bottle at 4°C until needed.

3.3. EXTRACTION OF SOYABEAN

The extraction of soybean oil was carried out using the solvent extraction method with hexane. The process began with the cleaning of the soybeans to remove dust, stones, sticks, and other foreign materials. After cleaning, the outer husk of the soybeans was removed.

The cleaned and dehusked soybeans were then crushed and rolled into thin flakes to increase the surface area for efficient solvent contact. These flakes were heated to a moderate temperature in a process called conditioning to soften them for oil extraction.

The conditioned flakes were then treated with a solvent. A mixture of acetone and hexane was used for the extraction. The hexane acted as the primary solvent to dissolve and separate the oil from the solid soybean matrix.

After the extraction was complete, the solvent was removed from the oil by heating. The evaporated solvent was condensed and recovered for reuse. The resulting product was crude soybean oil, which contained impurities such as gums, free fatty acids, and color pigments

3.4 DETERMINATION OF TOTAL PHENOL CONTENTS

The amount of total phenolics in the extract was determined with Folin–Ciocalteu reagent according to the method of Singleton and Rossi(1965) with slight modification using tannic acid as a standard.

Briefly, 1.0ml of extract solution (250 µg/ml) was added in a test tube. Then, 1.0 mL of Folin–Ciocalteu reagent was added, and the contents of the flask were mixed thoroughly. After 5 min, 1.5 ml Na₂CO₃ (20 %) was added and allowed to stand for 2 hours. The absorbance was measured at 760 nm using a UV-Vis spectrophotometer (Jenway 6100, Dunmow, Essex, U.K.). The total phenolic content was determined as µg of tannic acid equivalent (TAE) using an equation obtained from the standard tannic acid calibration graph.

3.5 STATISTICAL ANALYSIS

Statistical analysis using SPSS version 21 revealed no significant difference in phenolic content between the dried seed extracts of soybean and palm kernel. However, a significant difference was observed in their oil extracts, with palm kernel oil containing higher phenolic levels. This indicates that oil processing methods, particularly heat and chemical refining, substantially reduce phenolic compounds, resulting in lower antioxidant potential in refined oils compared to unprocessed seeds.

CHAPTER FOUR

4.0 RESULT

The results obtained from this study are presented in the tables below:

Table 4.1 and Figure 4.1 shows the total phenolic content of dried seed extracts of soya beans and palmkernel.

Table 4.1: The Result of the Total phenolic contents of dried seed extracts of Soybean and Palmkernel.

Parameters	Soybeans	Palmkernel
Total phenolic content (g TAE/kg)	98.4±1.60 ^a	135.6±15.9 ^a

Values are expressed as Mean ± Standard Error of triplicate experiments. Mean values with similar superscript within rows are not significantly different from each other (P>0.05). Mean values with different superscript within rows are significantly different from each other (P<0.05).

Phenolic content Soya beans sample was 98.4±1.60 while Phenolic content of Palmkernel was 135.6±15.9. Phenolic content of both soya bean and palmkernel were not significantly different (P>0.05)

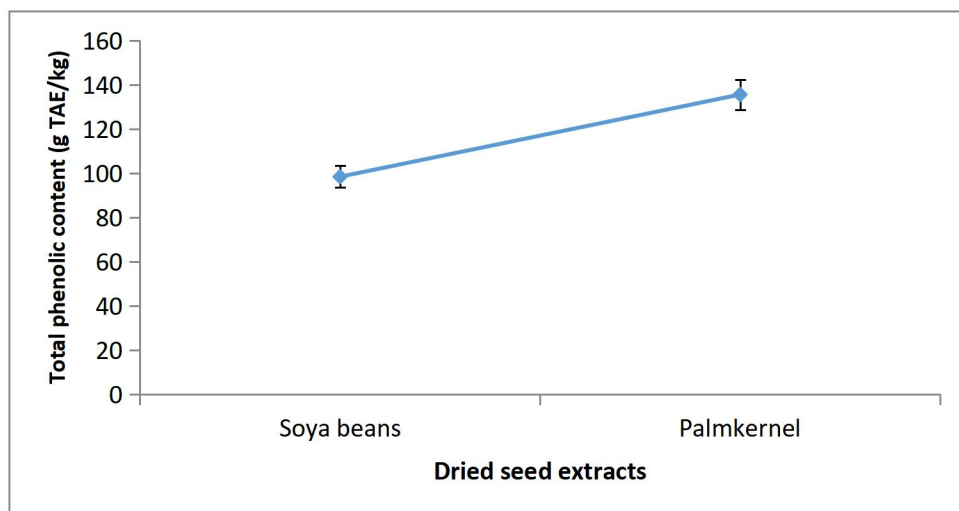


Figure 4.1: The total phenolic contents of dried seed extracts of Soybean and Palmkernel

The total phenolic content of dried oil extracts of soybeans and palmkernel is shown in Table 4.2 and Figure 4.2.

Table 4.2: The Result of the total phenolic contents of oil samples of Soybean and Palmkernel.

PARAMETERS	Soybeans	Palmkernel
Total phenolic content (g TAE/kg)	0.83±0.17 ^a	2.71±0.12 ^b

Values are expressed as Mean ± Standard Error of triplicate experiments. Mean values with similar superscript within rows are not significantly different from each other ($P>0.05$). Mean values with different superscript within rows are significantly different from each other ($P<0.05$).

Phenolic content Soya bean sample was 0.83 ± 0.1 while that of Palmkernel was 2.71 ± 0.12 . There was a significant difference between the total phenolic content of soya bean and palmkernel ($P<0.05$)

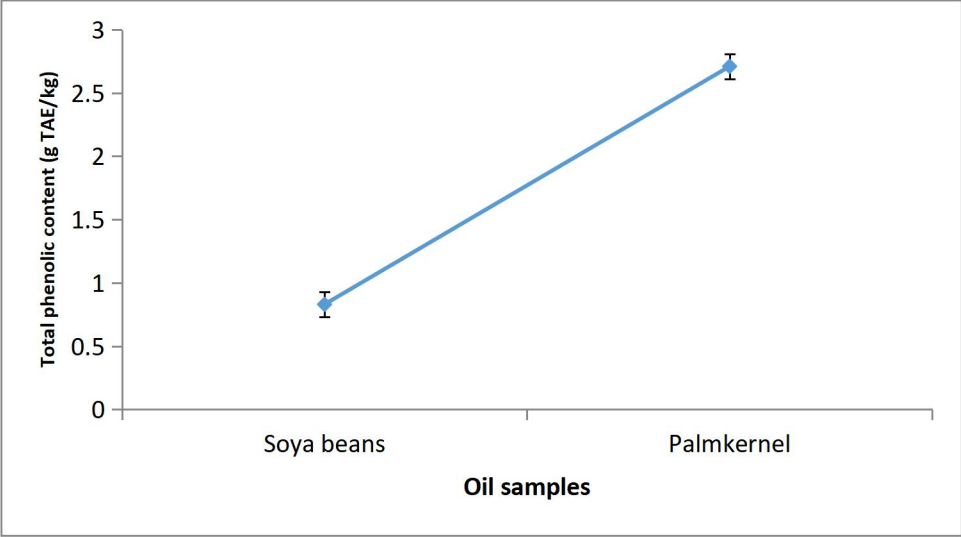


Figure 4.2: Total phenolic contents of oil samples of Soybean and Palmkernel

CHAPTER FIVE

5.0 DISCUSSION AND CONCLUSION

5.1 DISCUSSION

Phytochemical is a broad term for the hundreds of naturally occurring bioactive compounds found in plants. These compounds are not considered essential nutrients for human life, unlike vitamins or minerals, but they play crucial roles in the plant's defense mechanisms against pests, diseases, and environmental stressors (Liu, 2004). The total phenol contents of soybean and palmkernel was investigated in this study. The result revealed that high phenolic content. The presence of phenol in soybeans observed in this study agrees with the findings of Lee *et al.*, 2020 who reported that phenol was one of the highest phytochemical constituents of dried soybeans samples. The total phenolic content of soybeans *Glycine max* is a key indicator of their antioxidant potential and overall phytochemical richness. This findings also agrees with Chaiyaso *et al.*, 2018 who demonstrated that there was high presence of phenolic contents in dried soybean sampled. Phenolic contents observed in this study has been reported by Nguyen *et al.*, 2022 to be linked to greater antioxidant activity and potential health benefits. Phenolic-rich soybean varieties have demonstrated strong DPPH radical-scavenging and ferric-reducing antioxidant capacities (Lee *et al.*, 2020).

This study also revealed that dried palmkernel seed contain high proportion of phenols. This findings agrees with the study of Ezema *et al.*, 2022 who reported that dried palmkernel seed contain adequate level of phenols. The presence of phenols in palmkernel has been reported by Nwabueze 2019 to be influenced by several factors such the genetic variety of the oil palm, environmental conditions, and, critically, the method of processing or drying the seeds. The primary significance of these phenolic compounds lies in their potent antioxidant capacity.

Studies have shown that extracts from palm kernel seeds exhibit strong free radical scavenging activity, which is directly correlated with their total phenolic content (Ezema *et al.*, 2022). This antioxidant potential makes palm kernel seeds a promising source of natural preservatives for the food industry and functional food ingredients aimed at mitigating oxidative stress, which is linked to chronic diseases like cancer, cardiovascular disorders, and neurodegeneration.

Result of the total phenol contents in Soya beans oil shows that it contains low level of phenol. This may be as a result of processing oil soybeans oil which involves the application of heat. According to Nwabueze 2019 thermal processing can sometimes degrade heat-sensitive phenols, but it can also release bound phenolic compounds, making them more extractable and measurable. Studies have identified low levels of phenols in soybean oil and highlighted their role in oxidative stability (Shi, 2022; Najafi, 2019). The low levels are due in part to phenolics being primarily associated with the seed matrix rather than migrating into the lipid phase during oil extraction and refining. The practical relevance lies more in their influence on oil stability and potential enrichment rather than in high nutritional phenolic load (Najafi, 2019).

Palmkernel oil is show low level of phenol compared to the dried seed. This agrees with the findings of Nde and Foncha, 2020 who reported that the phenolic profile and concentration in palmkernel seed are significantly different and generally lower than those found in palmkernel oil. Ezema *et al.*, 2022 reported that the concentration of these compounds is heavily influenced by the extraction method. Conventional industrial refining processes, which involve high temperatures, bleaching, and deodorization, lead to a substantial loss of these heat-sensitive and polar phenolic compounds, resulting in a highly refined palmkernel oil with minimal phenolic content (Nde and Foncha, 2020).

5.2 CONCLUSION

This study confirms that soybean and palm kernel seeds contain high levels of phenolic compounds, which contribute to their antioxidant and health-promoting properties. In contrast, the extracted oils showed much lower phenolic content, indicating significant loss of these compounds during industrial processing, especially due to heat and refining. This suggests that while the seeds are valuable sources of natural antioxidants, their refined oils contribute minimally to dietary phenolics. The findings also highlight the need for gentler extraction methods, such as cold-pressing, to produce oils with better phytochemical retention. Overall, the study emphasizes the influence of processing on the nutritional and functional value of plant-derived products.

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APPENDIX

ANOVA TABLES

TOTAL PHENOLIC CONTENTS OF DRIED SEED EXTRACTS

DESCRIPTIVES

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean
					Lower Bound
Soya beans	3	98.4233	2.77544	1.60240	91.5288
Palmkernel	3	135.5863	27.49053	15.87167	67.2961
Total	6	117.0048	26.82722	10.95217	88.8514

DESCRIPTIVES

	95% Confidence Interval for Mean	Minimum	Maximum
	Upper Bound		
Soya beans	105.3179	96.80	101.63
Palmkernel	203.8766	103.84	151.46
Total	145.1583	96.80	151.46

ANOVA

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	2071.633	1	2071.633	5.427	.080
Within Groups	1526.865	4	381.716		
Total	3598.498	5			

PHENOLIC CONTENTS OF OIL SAMPLES.

DESCRIPTIVES

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean	
					Lower Bound	Upper Bound
Soya beans	3	.8307	.28601	.16513	.1202	1.5412
Palmkernel	3	2.7087	.20232	.11681	2.2061	3.2113
Total	6	1.7697	1.05222	.42957	.6654	2.8739

DESCRIPTIVES

	Minimum	Maximum

	1.00	.59	1.15
Soya Beans	2.00	2.48	2.87
	Total	.59	2.87

ANOVA

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	5.290	1	5.290	86.207	.001
Soya Beans Within Groups	.245	4	.061		
Total	5.536	5			