

Addis Ababa University
College of Natural and Computational Sciences
Center for Food Science and Nutrition



Proximate Composition, Bioactive Compounds and acceptability of
Cauliflower (*Brassica oleracea var. botrytis*) Leaf as a Meal and Bread
Nutrient Enhancer

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A thesis submitted to the College of Natural and Computational Sciences in partial
fulfilment of the requirement for the Master of Science Degree in Food Sciences
and Nutrition

Advisor: - Zeweter Abebe (PhD)

Declaration

I, the undersigned, declare that this thesis titled '**Proximate Composition, Bioactive Compounds and acceptability of Cauliflower (*Brassica oleracea var. botrytis*) Leaf as a Meal and Bread Nutrient Enhancer**' has been carried out and written by me in the Center for Food Science and Nutrition, Addis Ababa University, Addis Ababa, under the supervision of **Dr. Zeweter Abebe**. The information derived from the literature has been duly acknowledged in the text and list of reference provided. No part of this thesis was previously presented for another degree or diploma at this or any other institution.

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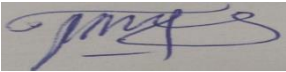
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List of Abbreviation

AACC.....	American Association of Clinical Chemists
AOAC.....	Association of Official Agricultural Chemists
GLS.....	Glucosinolate
GLV.....	Green Leafy Vegetables
ITC.....	Isothiocyanate
WHO.....	World Health Organisation

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Abstract

Cauliflower (*Brassica oleracea* var. *botrytis*) is one of the commonly known vegetables in Ethiopia and its leaves have been used as a by-product, the main objective of this research was to examine different ways of consuming and demonstrating nutritional value of cauliflower leaves. Different processing methods (boiling, blanching and fermentation) that are applicable at the household level were used to show its nutritional, antioxidant, vitamin C, mineral content and ways to reduce the anti-nutritional factors of the leaves. Moreover, the leaves' powder was incorporated with wheat flour to produce bread and the leaves were prepared as a meal and kimchi, and undergo a sensory evaluation. The result of the research showed that compared to the raw leaves, boiled leaves had statistically higher carbohydrate and energy contents and decreased crude protein, crude fat and crude fiber contents ($P < 0.05$). In terms of anti-nutritional factor all the processing methods showed a positive impact in decreasing the oxalate (2.74 to 0.78mg/100g) and phytate (1.15 to 0.07mg/100g) contents of cauliflower leaves. When talking about total antioxidant, flavonoid and vitamin C, the processing methods had a negative impact as they reduce all of them when compared with the raw except fermentation on total flavonoid showed a significant increase from 227.45 to 341.8 mg/100g ($P < 0.5$). In mineral analysis amount of zinc, calcium and iron showed an increment under boiling and fermentation and decrease under blanching. Bread that was formed in different formulation showed an increase in amount of protein (7.91 to 10.76g/100g), fat (0.75 to 2.25g/100g) and fiber (1.75 to 4.5g/100g) as amount of cauliflower leave powder in the flour increased. Sensory evaluation showed that the acceptability of the bread decreased as amount of cauliflower leave powder added increased. Leaves prepared as a meal were accepted by the panellists and Kimchi wasn't their favourite. Generally, according to this research it can be conclude that consuming cauliflower leave in different ways can be one way of reducing our low vegetable consumption habit, decrease nutrient deficiency in the country and reduce postharvest loss.

Keywords: Cauliflower leaves, proximate composition, Anti-nutritional factors, Enriched bread.

1. Introduction

1.1 Background

Fruits and vegetables could supply the body with nutrients, dietary fibre and other components necessary to the human body (FAO, 2015); therefore, adequate intake is necessary to ensure effective consumption of its components. Especially for people in developing countries, vegetables are the most available source of micronutrients (AVRDC, 1992). Low vegetable and fruit consumption contributes largely to morbidity and mortality and 31% of ischemic heart disease, 15% of cancers, and 11% of strokes are global disease burden attributable to low fruit and vegetable intake (Poirier *et al.*, 2019). Lack of vegetables alone causes 7.6% of deaths from heart disease, stroke, and type 2 diabetes (Cavanagh *et al.*, 2017).

The minimum recommended consumption of fruits and vegetables is 400g/day (FAO & WHO, 2003) or 146 kg per year; however, the consumption falls short of the global recommendation in most countries (Miller *et al.*, 2016; Micha *et al.*, 2015). For example in Ethiopia, the average Ethiopian household consumed only 42 kg of fruit and vegetables per adult equivalent annually in 2011 (Worku *et al.*, 2017), which only fulfilled around 29% of the World Health Organization's recommendation. Subsequently, nutrient deficiencies remain widespread in the country (CSA & ICF, 2016; Demissie *et al.*, 2010) due to heavy-dependence on cereals such as Tef, Maize, Wheat and others (Hunde, 2017). On the other hand, for better diet quality, efforts have been focused on improving nutrition knowledge through behavioural change communication and been successful in improving knowledge but consumption of nutrient rich foods remains poor (Kim *et al.*, 2016).

One of the reasons for the poor diet quality could be unaffordability of fruits and vegetables and other nutritionally rich foods (Warren & Frongillo, 2017). According to Hirvonen *et al.*, (2018), it requires respectively 11 and 27% of their income for the average and for the poorest households to meet the international guideline of fruit and vegetable consumption in Ethiopia. The reported high price could contribute for the products to be out of the reach of the poorest in the country. However, not only the fruits and the vegetables but also the by-products from them could be employed to mitigate health and nutrition problems (Tamasi *et al.*, 2019). Unfortunately, some vegetable have been underutilized by discarding its parts as by-products while they are actually nutritious. The improper utilization of the vegetables affects not only nutrition but also it makes the sustainable development goal target 12.3 that

aims to reduce global food waste by 50% by 2030 (United Nations, 2015) challenging to achieve.

Though, fruits and vegetables are rich in nutrients and other health promoting substances, some vegetables also contains anti-nutritional substances such as oxalates, saponins, tannins, phytic acid and alkaloids, which are known to bring gastrointestinal problems like bloating, constipation, nausea, gas, and inhibit bioavailability of nutrients such as calcium and iron (Sethia *et al.*, 2021). Also, anti-nutrients can become toxic when present above a certain level. Therefore identification of suitable processing procedures to reducing their concentration in foods should be encouraged before their consumption (Reddy and Pierson 1994) to prevent toxicity and associated health problems caused by these compounds (Gemedé and Ratta 2014). Boiling, Blanching and Fermentation processes are used in this research because those processing methods are easily applicable at the household level as one of the main objective of this research is to make sure our society uses the leaves for daily consumption it is critical to also suggest ways that can be applicable. Further, the acceptability of the products as a meal or as a food nutrient enhancer requires a thorough investigation.

One of the commonly known and expensive green leafy vegetable in Ethiopia is cauliflower (*Brassica oleracea var. botrytis*), which grows extensively in different parts of the world. Regrettably, it is among vegetables with significant waste rate (45-60%) (Castelão-Baptista *et al.*, 2021; Petkowicz & Williams, 2020). In many countries, including Ethiopia, only the head is eaten and its leaves are usually discarded as a non-edible waste material (Chakraborty & Datta, 2016). But some studies reported that consumption of cauliflower leaves might help reduce micronutrient deficiencies (Singh *et al.*, 2019) and it could also be used to enrich food products (Tukassar *et al.*, 2023). For example, the leaves can be used to prepare a healthy functional food such as Kimchi whose major ingredients are cruciferous vegetables; and other healthy functional foods like garlic, ginger, red pepper powder, and other sub ingredients. Therefore, the Kimchi prepared from the leaves could be taken as a vegetable probiotic with significant health benefits similar to yogurt (Ju *et al.*, 2018).

However, there are limited evidences on the nutritional and anti-nutritional characteristics of the leaves under different processing procedures, acceptability of the leaves as a vegetable probiotic, and the desirability of the leaf powder as a wheat bread enhancer. Therefore, this study aimed to investigate the nutritional and anti-nutritional composition of cauliflower

leaves under different processing methods and to examine the acceptability of the leaves as a meal and the powder as wheat bread nutrient enhancer.

1.2 Statement of the problem

The consumption of vegetables is low in Ethiopia. Some of the reasons that may contribute to our low vegetable consumption includes: First, vegetables and fruits are unaffordable, in a study by Hirvonen (2018), the average Ethiopian household would have to spend 11% of their income to meet the international recommendation of two servings of fruit and three servings of vegetables per person per day and this share is more than 2.5 times higher for poorest households. Second, they have short shelf life; most vegetables only last 2-7 days after being harvested, so buying those frequently to consume them fresh will not be cost effective. Third, their discarded part is much higher than the consumed part; the plant part that is consumed is much lower in amount when compared to the waste that is excreted. The later factor could be explained as: some vegetables' waste part may be toxic, so removing them may be the best option; on the other hand, the part we consider as a waste might be a very nutritious component, and people may remove it due to a lack of awareness on how to use it for consumption.

Cauliflower is a well-known vegetable in our country, but as previously stated, one of the factors that affects its wide usability is that the excreted portion is considerably higher than the ingested portion. The leaves represent nearly half of the plant's overall production (Pankar & Bornare, 2018), and it is considered as a waste product in despite the fact that it contains many nutrients and has an antioxidant impact. Thus, this study tried to address this gap by treating the leaves using different processing methods and investigated the possible usage of the leaves as part of the regular diet.

This research is conducted in light of the seriousness of nutritional problems and the large proportion of the population suffering from multiple forms of nutritional deficiencies (USAID, 2022), as well as the difficulties nutritionists face in convincing consumers to include green leafy vegetables in their diets to combat vitamin deficiencies.

1.3 Objectives

1.3.1 General Objective

To investigate the nutritional, anti-nutritional and bioactive compounds of cauliflower leave under different processing methods, and to evaluate its acceptability as a meal and bread nutrient enhancer

1.3.2 Specific Objectives

- ✓ To investigate the proximate composition and anti-nutritional factors in raw, blanched, boiled and fermented Cauliflower leaves.
- ✓ To determine the levels of flavonoid, total antioxidants and vitamin C in cauliflower leaves before and after processing.
- ✓ To evaluate the cauliflower leaves as a meal and determine the acceptability of the powder as a bread nutrient enhancer.

1.4 Significance of the study

Since, cauliflower is more expensive than other vegetables in our country (120 birr per piece), consuming only the flower and discarding the green portion creates a lot of waste and is inefficient economically. Studies show that cauliflower leaves can help in improving micronutrient consumption, and that they also contain several antioxidants that can help to minimize the risk of chronic diseases (Pankar & Bornare, 2018).

On the other hand, many bakeries are working to distinguish their products and add value to them; many potential nourishing foods have been proposed to increase the nutritional quality of bread. In addition to white bread, many so called 'health breads' are available which contain many added ingredients to white flour such as oat, bran, rye, barley, lupine multigrain and algae. The addition of such ingredients and others are attempting to create functional breads to attain additional health benefits (Alkurd *et al.*, 2020).

This study tried to primarily use different processing methods (boiling, blanching and fermentation) to determine and see the difference in the leaves' nutritional value and anti-nutritional factor; secondarily the leaves were used to make a meal like cabbage in order to evaluate the acceptability. Thirdly the leaves that undergo salt fermentation were used to make kimchi in order to recommend another way of consumption and finally the leaves' powder was used as an ingredient in making bread in different proportion and was tested if it improved the nutritional quality of the bread and if the bread can be acceptable by consumers. Because cauliflower leaves are treated as a waste in our country this study used part of a vegetable that is considered as a waste and tried to give ideas on how to consume it in processed form and powder form.

Thus, the research helps to improve our knowledge of cauliflower leaves and help to reduce waste from the plant. Finally, contribute to the improvement of the nutritional status in the country by providing suggestions on how to consume the leaves.

2. Literature Review

2.1 Vegetables

Vegetables are defined as the fresh parts of plants which either raw, cooked, canned or processed in some other way, provide sustainable human nutrition (Belitz *et al.*, 2008). They come in varied colors, forms, and tastes. Health specialists highly encouraged inclusion of vegetables in one's diet due to their inherent nutritional worth (Mohammed & Qoronfleh, 2020). A widely used definition of a vegetable is: a herbaceous plant or portion of a plant that is eaten whole or in part (Welbaum, 2015). Vegetables can be classified according to botanical classification, classification based on hardness or temperature, classification based on plant part used, classification based on culture and classification based on life cycle: they are classified as cole crops, leafy vegetables, cucurbit crops, solanaceous crops, pea and beans, bulb vegetables, perennial vegetables, tuber vegetables and okra based on culture.

2.1.1 Green leafy vegetables

'Leafy greens' or 'greens' are broad terms used for a number of vegetable crops with edible leaves. Leafy greens, also called potherbs greens, vegetable greens or salad greens, are plant leaves eaten as a vegetable, sometimes accompanied by tender petioles and shoots. Plants in this group belong to several unrelated taxonomic plant families that include *Brassicaceae*, *Chenopodiaceae*, and *Asteraceae*. Although they come from a very wide variety of plants, most share a great deal with other leafy vegetables in nutrition and cooking method.

There are numerous underutilized greens in nature with promising nutritional content that can feed the world's expanding population. Many of them are tolerant of harsh climatic conditions, hardy, and adaptive. Despite the fact that they may be raised on marginal lands with lower management costs, they are still underutilized due to a lack of awareness and the lack of popularization of utilization technology. Underutilized foods are becoming more important these days as a way to enhance the amount of food available per capita (Sheela *et al.*, 2004).

2.1.2 Nutritional value

Because of their abundance in vitamins, minerals, amino acids, dietary fiber, and numerous vital bioactive substances, vegetables are frequently referred as "protective foods" in the human diet (Natesh *et al.*, 2017). They play important role in food and nutritional security. Researchers have estimated that every serving increase in fruit and vegetables consumption reduces the risk of cancer 15%, cardiovascular disease by 30% and mortality by any cause by 20% (Gupta & Prakash, 2009). Particularly, green leafy vegetables are considered as exceptional source for vitamins, minerals and phenolic compounds (Natesh *et al.*, 2017).

Green leafy vegetables (GLV) are recognized for their characteristics color, flavour and therapeutic value (Gupta *et al.*, 2005). They are rich sources of many nutrients and form a major category of vegetable groups that have been designated as 'nature's anti-aging wonders'. Gupta & Prakash, (2009), have reported that several GLV are rich sources of antioxidants, vitamins and they are the richest and cheapest sources of proteins. This is because of their ability to synthesize and accumulate amino acids with the help of abundant source of sunlight, water, oxygen and nitrogen, which is readily available in the atmosphere (Natesh *et al.*, 2017). They have been traditionally recognized as good sources of dietary fiber. It was more significant in resolving the problem of constipation, diabetes, diverticulosis and obesity (Pankar & Bornare, 2018).

2.2 Cauliflower

2.2.1 Nomenclature

Family: *Brassicaceae*

Genus: *Brassica*

Species: *oleracea*

Common name: Cauliflower

Scientific name: *Brassica oleracea var. botrytis*

2.2.2 Historical Background

The evolution of cauliflower and broccoli would seem to have taken place in the Mediterranean basin, in particular in its east coast (Gray, 1982). In ancient Rome, the close trading ties that existed between a large numbers of Mediterranean nations helped to facilitate the spread and trade of genetic material among various areas. With the cultivation and selection of genotypes with interesting agronomical and qualitative qualities, as well as the discovery of many varieties

and shapes of cauliflower and broccoli, the evolution process likely led to adaptation to various soil-climatic conditions (Branca, 2008).

Due to the challenges of generating seed in environmental conditions that were distinct from those of their area of origin, cauliflower crops may have been introduced in western Europe after a much longer time than they were on the east side of the Mediterranean coast (Branca, 2008).

Cauliflower seems to originate along the east coast of the Mediterranean basin from wild forms of *B. oleracea* which were widespread in Italy, where the entree of the broccoli gene pool probably took place (Branca, 2008). On the other hand some authors put forward the hypothesis that cauliflowers originate in the central Mediterranean basin directly from broccoli and, only after that, spread to the other areas (Gray, 1982).

China is said to be the main producer of Brassica vegetable with almost 45% of the total production, in 2020 their estimated production was 100 million tons, cultivated in 4 million hectares, where cabbages and other Brassicas represent $\frac{3}{4}$ of the production and cauliflower and broccoli represent $\frac{1}{4}$ (Silva Dias, 2022).

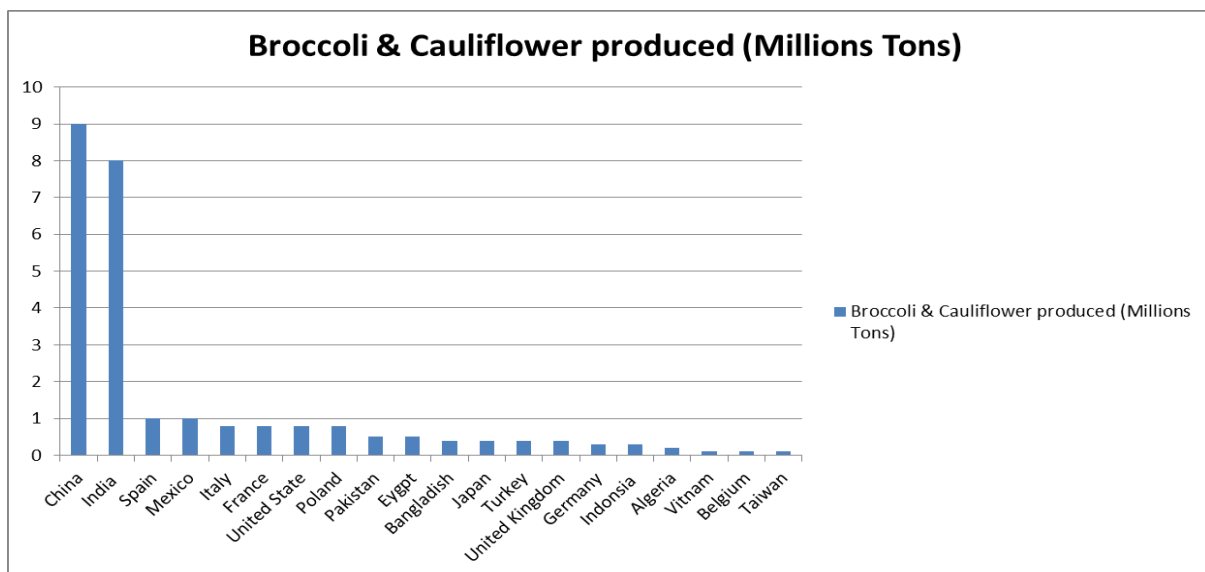


Figure 1: Production of Broccoli and Cauliflower worldwide (Gupta *et al.*, 2019)

When we talk about production in Ethiopia, production of cauliflower specifically is not available but in a study that was conducted at Bahir Dar University by Demoz Kidane G/meskel (2016), generally the production of cabbage and other brassicae are cultivated in mid altitude and highland areas of Ethiopia, which include Oromia region, Amhara region, S.N.N.P.R and Benishangul-Gumuz respectively with production level (Gebremeskel, 2016).

2.2.3 Agronomic characteristics

Cauliflower is a seed-reproducing annual plant. Usually, the stalk and surrounding dense, green leaves of aborted floral meristems are discarded and just the head (the white curd) is consumed. Prior to the lengthening of flower stalks, it begins hypertrophy, making the primordial reproductive buds that are present at this stage of development infertile (Branca, 2008).

The flower branches represent the sink organ where the reserve substances accumulate, resulting in their abnormal development which determines a continuing branching and the amplitude of curd angle (Branca, 2008).

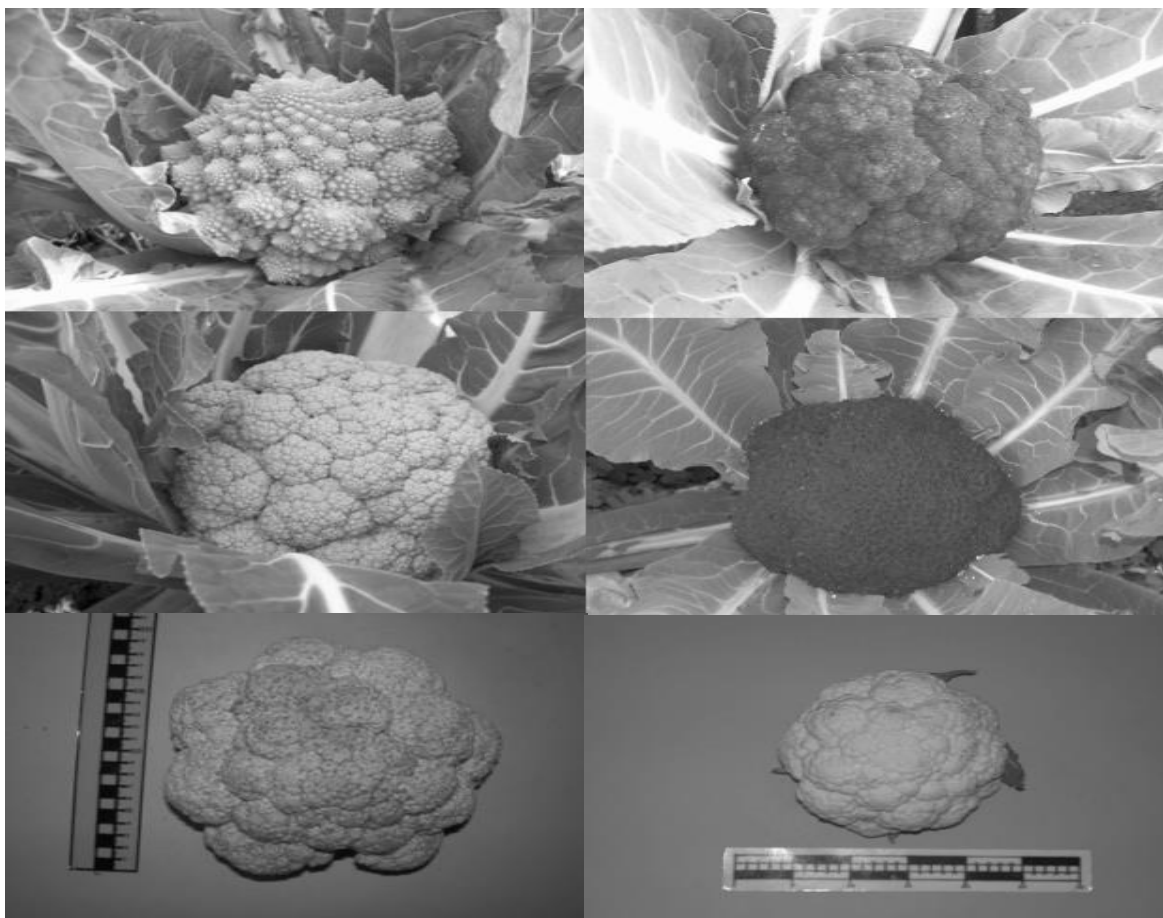


Figure 2: Different types of cauliflower curd (Branca, 2008)

Surrounding the curd are ribbed, coarse green leaves that protect the flower, which are attached to a central stalk, from sunlight, impeding the development of chlorophyll. Cauliflower leaves are medium to large in size and long and rounded in shape.



Figure 3: Cauliflower

The leaves range in hue from light forest green to dark emerald green, although each cauliflower plant has slightly different color and shape variations. The leaves have a thick, fibrous central rib that is bright green. Harvesting cauliflower leaves while they are young and tender is optimal because they are crunchy and have a flavor that is light, mild, and fresh.

Cauliflower can be cultivated in various types of weather such as in cool, warm and hot weather conditions. The field that receives warm sunshine, fertile soil, facility of irrigation and drainage, and not cultivated cauliflower for two years is better for cauliflower cultivation.

2.2.4 Nutritional value of Cauliflower

The *Brassicaceae* (Cruciferae) family is composed of 350 genera and about 3500 species. *Brassica* vegetables (for example, broccoli, cabbage, kale, mustard greens, Brussels sprouts, cauliflower, etc.) represent a rich source of health-promoting phytochemicals such as vitamins, carotenoids, fiber, soluble sugars, minerals and phenolic compounds (Ahmed & Ali, 2013), their beneficial effects have been attributed to the anticancer properties of glucosinolates (GLSs) and their isothiocyanate (ITC) derivatives (Larocca *et al.*, 2017).

Table 1: Major nutritional components of commonly used cruciferous vegetables

Cruciferous vegetables	g/100g fresh weight			
	Protein	Fiber	Fat	Carbohydrate
Arugula	2.58	1.60	0.66	3.65
Bok choy	1.42	1.42	0	2.85
Broccoli	2.82	30.40 ^a	0.37	6.64
Brussels sprouts	2.55	26.94 ^a	0.51	8.67
Cabbage	1.53 (green) 0.97 (red)	23.24 ^a	0	6.00
Cauliflower	29.9^a	26.70^a	0.45^b	4.11^b
Chinese Cabbage	1.50	1.00	0.20	2.20
Collard greens	3.00	4.60	0.40	7.10

Note- Values are converted from content per serving or fresh average vegetable weight.

a- Dry weight

b- Boiled (Manchali *et al.*, 2012)

In addition to being a popular crop grown all over the world, cauliflower also has a wide range of uses as an ingredient in dishes like salads and soups. It and broccoli are considered to be the same variety of the cruciferous family because of their close resemblance. In addition to their wonderful phytochemicals, they also contain nutrients such as vitamin A, Thiamine, Riboflavin, Niacin, vitamin C, calcium, iron, phosphorous, and fat to aid in disease prevention (Ahmed & Ali, 2013).

Table 2: Macro-minerals composition of commonly consumed cruciferous vegetables

Cruciferous vegetables	mg/g of fresh weight				
	Phosphorus	Potassium	Magnesium	Sodium	Calcium
Arugula	0.50	3.70	0.45	0.25	1.60
Bok choy	0.37	2.50	0.19	0.92	1.06
Broccoli	0.66	3.17	0.21	0.33	0.47
Brussels sprouts	0.69	3.89	0.23	0.25	0.42
Cabbage (domestic)	0.23	2.46	0.15	0.18	0.47
Cauliflower	0.44	3.03	0.15	0.30	0.22
Chinese Cabbage	0.37	2.51	0.19	0.65	1.05

Note- Values are converted from content per serving or fresh average vegetables weight (Manchali *et al.*, 2012)

Table 3: Micro-minerals composition of commonly consumed cruciferous vegetables

Cruciferous Vegetables	µg/g of fresh weight				
	Iron	Selenium	Copper	Manganese	Zinc
Arugula	0	0	0	0	0
Bok choy	8.00	-	-	1.42	1.00
Broccoli	7.70	0.025	-	2.20	4.40
Brussels sprouts	14.00	0.016	1.10	3.40	4.50
Cabbage (domestic)	5.60	0.009	0.22	1.50	1.70
Cauliflower	4.00	0.006	-	2.00	0.30
Chinese Cabbage	8.50	0.004	-	1.40	1.40

Note- '-' not reported, '0' not detected. Values are converted from content per serving or fresh average vegetables weight (Manchali *et al.*, 2012)

2.2.5 Nutritional value of Cauliflower leaf

Cauliflower leaf is considered as one of the green leafy vegetables, which means it consists all the characteristics and benefits of GLVs, for instance it is a good source for both fiber and vitamin C. It also contains vitamin A, folate, calcium, and potassium as well as selenium, which works with Vitamin C to boost the immune system.

Table 4: Major nutritional composition of cauliflower leaf

Parameter	Result
Energy (Kcal)	66
Protein (g/100g)	5.9
Carbohydrate (g/100g)	7.6
Fat (g/100g)	1.3
Crude Fiber (g/100g)	2
Moisture (g/100g)	80

(Pankar & Bornare, 2018)

Table 5: Mineral content (mg/100g) of cauliflower leaf

Parameter	Result
Calcium	626
Copper	1.55
Iron	60.38
Zinc	5.10

(Pankar & Bornare, 2018)

2.3 Anti-nutritional factors

Anti-nutrients are compounds found in plants that serve as a defence mechanism for the plant as well as assisting in other biological activities. They impair the capacity of nutrients like minerals, vitamins, and even proteins to function within plant material. As a result, the nutritional content of the plants will be affected. Anti-nutrients comprise of amino acids to proteins, simple amines to alkaloids, glycosides and phenolic compounds. Anti-nutrients are taken when a plant food is consumed as a nutritional source, posing a health danger to the user (Essack *et al.*, 2017). Some of the anti-nutrients commonly found in leafy vegetables are nitrates, oxalates, tannins, phytates

and cyanogenic glycosides. The presence of anti-nutrients indicates that the evaluation of anti-nutritional factor is therefore necessary.

2.3.1 Oxalate

Kidney stones are one of the most painful disorders to afflict human and they are one of the most common disorders of the urinary tract. A large proportion of these kidney stones are made up of oxalate derived either from normal metabolism in the body or from dietary sources (Savage, 2002). Depending on species, oxalate can occur as a soluble salt of potassium and sodium salts and as insoluble salts of calcium, magnesium and iron or as a combination of the two forms. Insoluble oxalate is excreted in the faeces while the soluble oxalate is absorbed by the body. Soluble oxalate forms strong chelates with dietary calcium, rendering it unavailable for absorption and assimilation (Uusiku *et al.*, 2010).

The absorption of oxalates from food appears to make an important contribution to urinary oxalate excretion. People who suffer from kidney stones are often advised to try and limit their intake of known high oxalate containing foods (Savage, 2002). Oxalic acid accumulates in plants especially during dry conditions. Reports of some tropical leafy vegetables revealed that dry vegetables had higher oxalate concentration than fresh vegetables (Savage, 2002).

Oxalates affect calcium and magnesium metabolism and react with proteins to form complexes which have an inhibitory effect in peptic digestion (Veer *et al.*, 2021). The ingestion of 4-5g of oxalate is the minimum dose capable of causing death in adults, but reports have shown that 10-15g is the usual amount required to cause fatalities, its ingestion results in corrosion of the mouth and gastrointestinal tract, gastric haemorrhage, renal failure and haematuria (Bsc & Bsc, 1999). Accurate and reliable information on the oxalate content of common foods is not readily available due to difficulty in measuring it (Bsc & Bsc, 1999). Some commonly used data on the oxalate content of foods has been determined on raw foods and no account has been taken of the effect of cooking and processing.

2.3.2 Phytate

Phytic acid is a natural substance that acts as a major storage of phosphorus in all leafy vegetables (Natesh *et al.*, 2017). Phytic acid has been shown to decrease mineral absorption, diminish the bioavailability of metal ions such as zinc and iron, and affects protein and starch digestion. A phytic acid consumption of 4-9 mg/100g reduces iron absorption in humans by 4-5 folds, and a phytate-rich diet is linked to nutritional disorders in children and adults such as rickets and osteomalacia (Essack *et al.*, 2017).

Processing techniques such as boiling and cooking have no effect in reducing the level of phytic acid as the phytate is relatively heat stable. However, evidence showed that although food preparation techniques help in reducing the phytic acid level, the most effective methods are lactic acid fermentation and soaking in acid medium (Natesh *et al.*, 2017).

2.4 Bread

The code of Food, Beverage and shared objects defines ‘Bread’ as, the product prepared by baking in special furnaces and under defined conditions of mass consisting of wheat flour, water, dough and a small amount of salt (Kourkouta *et al.*, 2017). Man learned the art of bread making more than 4000 years ago. Though not always in the same form or as we know it today, bread has been a popular staple food for ages (Dewettinck *et al.*, 2008).

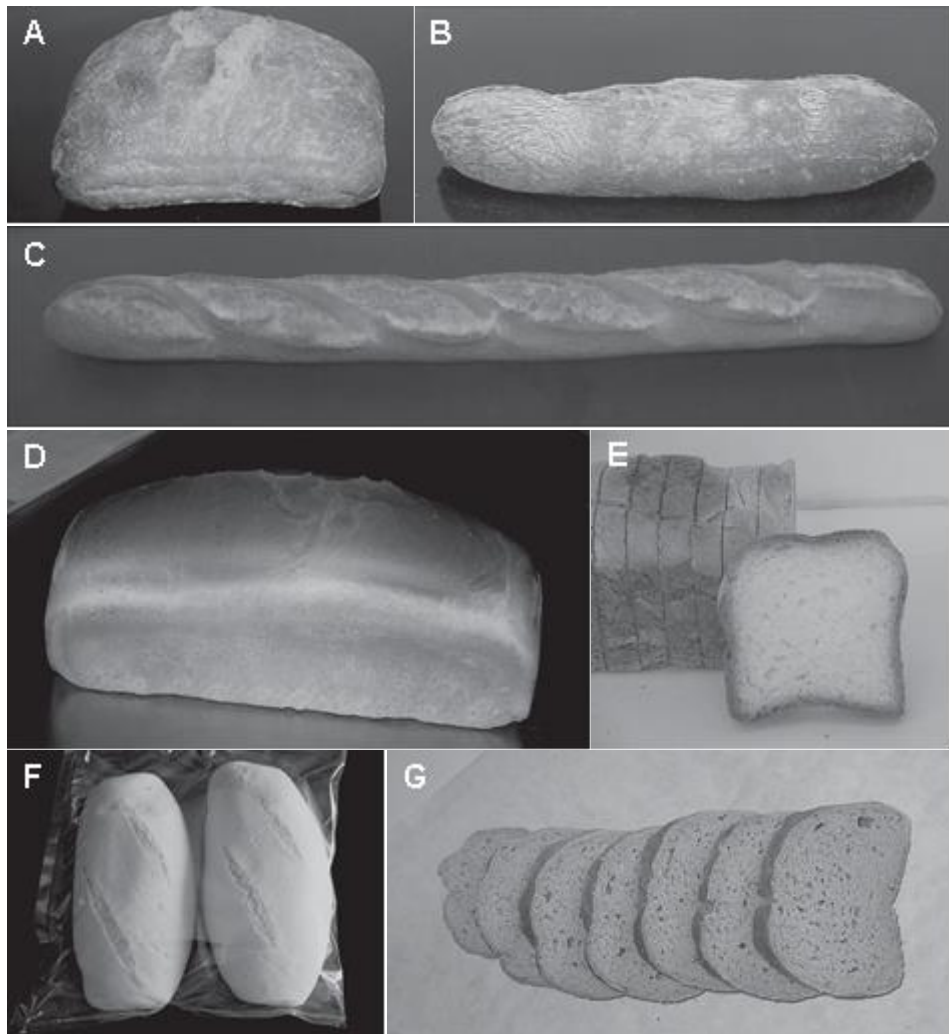


Figure 4: Different types of breads. (A and B) crusty bread named Ciabatta, (C) Baguette, (D and E) Pan bread, (F) partially baked bread and (G) fiber enriched bread (Rosell, 2011)

2.4.1 Nutritional Value of bread

Bread is an excellent source of complex carbohydrates that provide the body with the energy needed; wheat is the only cereal whose flour has the ability to form dough when contacted with water. As a result, flour has the exclusive property of shaping the structure and appearance of bakery products (Kourkouta *et al.*, 2017). Flour consists of proteins, starch and other carbohydrate, lipids, water and ash as well as low levels of vitamins, minerals and enzymes (Kourkouta *et al.*, 2017).

The amount of protein contained in the flour determines the amount of gluten to be formed; this in turn determines the strength, shape and structure of the dough. In addition to the protein that constitute 10-12% of the flour, most of it consists of starch (70-75%), while water is about 14%, it is also composed of non-starch polysaccharides (2-3%) (Kourkouta *et al.*, 2017). Most types of bread contain low-value, low-fat vegetable protein. They also contain B vitamins, vitamin E and trace elements such as iron and selenium. Interactions between bread and companion foods have effects on the nutritional quality (Dewettinck *et al.*, 2008).

2.4.2 Production in Ethiopia

Bread wheat is a widely adapted crop that can be grown in many different environments, though its productivity is higher in suitable environments (Alemu *et al.*, 2021). Ethiopia is the largest wheat producer country in sub-Saharan Africa with a potential expansion area more than 1.4 million ha. Wheat is 4th and 3rd most important food crop in terms of production and productivity respectively, wheat covered 1.61 million hectares and ranks 4th after Tef, Maize and Sorghum, Bale and Arsi highlands of south eastern Ethiopia is known by high bread wheat producing areas in the country (Tsegaye, 2012).

In Ethiopia, a number of improved bread wheat varieties have been released by different breeding institutes and these have contributed to increased productivity and production for the last decade (Alemu *et al.*, 2021). As bread is becoming the second most eaten food in the country the health consequence of white wheat bread is also increasing, researches that include ways to enhance quality of bread by incorporating other ingredients needs to be addressed highly.

2.5 Processing Methods of Cauliflower

There is a need to preserve the nature's storehouse of nutrients through convenient processing techniques. Different studies suggest different methods in order to attain a better meal. In a study

by Gupta & Prakash (2011), dehydration seems to be the simplest technology for preserving greens especially when they are abundantly available.

Factors such as storage, cooking and drying influences micronutrient, antioxidant and anti-nutritional factor of green leafy vegetables (Uusiku *et al.*, 2010), for instance high oxalate foods should be boiled to reduce the oxalate content. Soaking raw foods will also reduce the oxalate content, but soaking foods to remove soluble oxalate will also result in other useful nutrients such as vitamin C being lost at the same time (Savage, 2002).

Food processing techniques are often utilized in order to reduce anti-nutrients in food. Cooking is a common form of processing of plants that are consumed as a food source at household level. Some studies have suggested that different cooking methods may have different effects on different nutrients and anti-nutrients (Schönfeldt & Pretorius, 2011). Thus, the micronutrients and anti-nutrients are affected differently by processing, depending on the type of processing, as well as the type of vegetable species. Cooking causes changes in the phyto-chemistry of the leafy vegetable affecting its nutrients bioavailability and health benefit properties. The degree of these changes depends largely on the cooking methods as well as the type of the vegetable (Odhav *et al.*, 2007).

2.5.1 Boiling

The process of evaporation associated with vapour bubbles in a liquid is called boiling, it is cooking food by just immersing them in water at 96-98°C and maintaining the water at that temperature till the food is tender (5 minutes), when foods are cooked by boiling, the food should be brought to a vigorous boil first and the heat is then turned down, as violent boiling throughout tends to break the food. The temperature of the water cannot be increased any further after it begins to boil. Continued vigorous boiling results in excessive evaporation of water and waste of fuel and foods are likely to get burnt at the bottom and form a dry crust at the top (Babalola *et al.*, 2010).

2.5.2 Blanching

Blanching is defined as the enzyme (heat resistant) deactivation phenomena, which helps in retaining color, reduction in initial microbial growth, cleansing the product, product preheating prior to processing, and gas exhausting from plant tissue (Shaheen *et al.*, 2012). Blanching is done by dipping of the vegetable into boiled water (96-98°C) for a very short time (1 minute) (Babalola *et al.*, 2010).

2.5.3 Fermentation

Fermentation is one of the oldest methods of food preservation technology in the world. The process relies on the biological activity of microorganisms for production of a range of metabolites which can suppress the growth and survival of undesirable micro flora in foodstuff (Zagorec *et al.*, 2008), as raw vegetables have a high microbial load and cannot be pasteurized without compromising product quality, most vegetable fermentations occur as a consequence of providing growth conditions such as added salt.

In a study by Zagorec *et al.*, (2008) steps in salt fermentation of vegetables include:

1. The vegetable will be washed and drained and placed in fermenting container.
2. Salt will be sprinkled over all and then another layer will be added, followed by more salt.
3. The salt will extract the juice from the vegetable tissues, thus forming the brine
4. A cloth will be placed above the vegetable and a weight will be added to compress the vegetables and facilitate brine formation and expulsion of air
5. Fermentation starts right after the formation of the brine, the process will last between one to four weeks, depending on the ambient temperature.

3. Materials and Methods

3.1 Study Area

The study area of this study was in Addis Ababa, Ethiopia

3.2 Sample Collection and Sample Preparation

The cauliflower leave samples were collected early morning from randomly selected vegetable markets established by Addis Ababa city administration. The vegetable markets in the city were identified from Addis Ababa city trade and industry bureau registry. So it is believed each sub-city has one big vegetable markets as Addis Ababa have 12 sub-cities one third of the vegetable shops (4 vegetable shops) within the selected markets were identified for sample collection using a lottery method, those are Kirkos sub-city (Agoza), Nifas Silk Lafto sub-city (Garment), Yeka sub-city (zero 2) and Adis Ketema sub-city (Merkato). Vegetable shops that were selling the vegetables only under shade were identified. Cauliflowers were bought 3 times 20 pieces from each market, that were fresh looking and free from bruises were transported to the laboratory after covering with plastic bags followed by immediate separation of the white curd from the leaf and cleaning under running water.

3.3 Treatment Combination, Experimental Design, Experimental framework and Processing Method

3.3.1 Treatment Combination

3.3.1.1 Processing and Leaf Powder preparation

The cleaned leaves were randomly assigned to salt fermentation, boiling, and blanching processes, then the remaining leaves were allocated to the control group without undergoing any processing procedure. The fermentation was done for 8 days in a plastic jar by adding 30g of salt into 1000g of cauliflower leaves (Round, 1917) under room temperature. Similarly, to boil the leaves, they were left in boiled water for 5 minutes until the texture changes. Blanching was effected by dipping the leaves in water boiled for two minutes followed by immediate immersion in cold water (Livingston *et al.*, 1972). Then leaves were dried in an oven (Genlab. model OV/125/SS/F/DIG/A) with the heat uniformly distribution and maintained 450C. It took 16 hours for the boiled, blanched, and fermented leaves to dry but it took only 14 hours for raw leaves to get dried. The dried leaves were grinded in to fine powder using Xian Siway Scientific instrument high speed sample miller model ZN-08.

3.3.1.2 Preparation of a meal using cauliflower leaves

Cauliflower leaves that is separated from the head was washed and blanched then the leaf, 1 piece garlic and 1 piece onion were chopped, while doing that a pan was placed on an oven with 10ml of sunflower oil in it. After the oil got warm the chopped leaf was added with the onion and garlic. It was stirred and cooked for half an hour and 1 teaspoon salt was added and finally it was placed on a different plate. A controlled dish was prepared by the same method by using Ethiopian spinach and sensory evaluation was done for both.

3.3.1.3 Preparation of Kimchi

Sliced leaves of cauliflower were placed in a bowl and 1 teaspoon of salt was added and set aside for an hour, after setting that aside the paste was prepared by blending 1 piece of garlic, ginger, chilli sauce, 1 teaspoon sugar, and 5ml rice vinegar together in small bowl. After 1 hour, the leaf was rinsed under cold running water, drained and dried thoroughly. The paste and the leaf were mixed in a large bowl along with the 200g radishes, 1 piece carrot and spring onions. Then finally the prepared kimchi was packed into a jar, sealed and was kept in the fridge for a week, a controlled dish was prepared by the same method by using Napa cabbage and served for sensory evaluation.

3.3.1.4 Preparation of Bread

The proportion of cauliflower leave powder to be added to the wheat flour was determined based on previous studies that made bread by mixing wheat flour with green leafy vegetable powder (Famuwagun *et al.*, 2016, Odunlade *et al.*, 2017). Based on the result of the studies, the minimum and the maximum cauliflower leaf powder to be added in to the wheat flour was set to be 0% and 9% respectively. The raw leaf powder was mixed with the wheat flour (*Var. Triticum aestivum*) , which is the dominant wheat type covering currently more than 90% of wheat production in Ethiopia (Assefa *et al.*, 2022), using flour mixer to ensure proper mix-up.

Table 6: Different formulation of wheat flour and cauliflower leaves powder mix used to make bread

Sample code	Cauliflower leaf powder (%)	Wheat flour (%)
F1	9	91
F2	7	93
F3	5	95
F4	3	97
F5	1	99
F6	0	100

Note: F- Formulation

Then, the different formulations of flour were mixed well with yeast (4gyeast/100g of flour) and the dough was left to rise for 4 hours. Finally, bread was made using an oven (Vigor, model 3 decks) at 200°C until the loaf turns golden brown and sounds hollow, which took 35 minutes.

3.3.2 Experimental Design

Completely randomized design (CRD) was used. The experiment was done in duplicate for each laboratory analysis. The experiment was conducted at Addis Ababa University, Center for Food Sciences and Nutrition laboratories, and breads with different formulations were beaked at Entoto Polytechnic College.

3.3.3 Experimental Framework

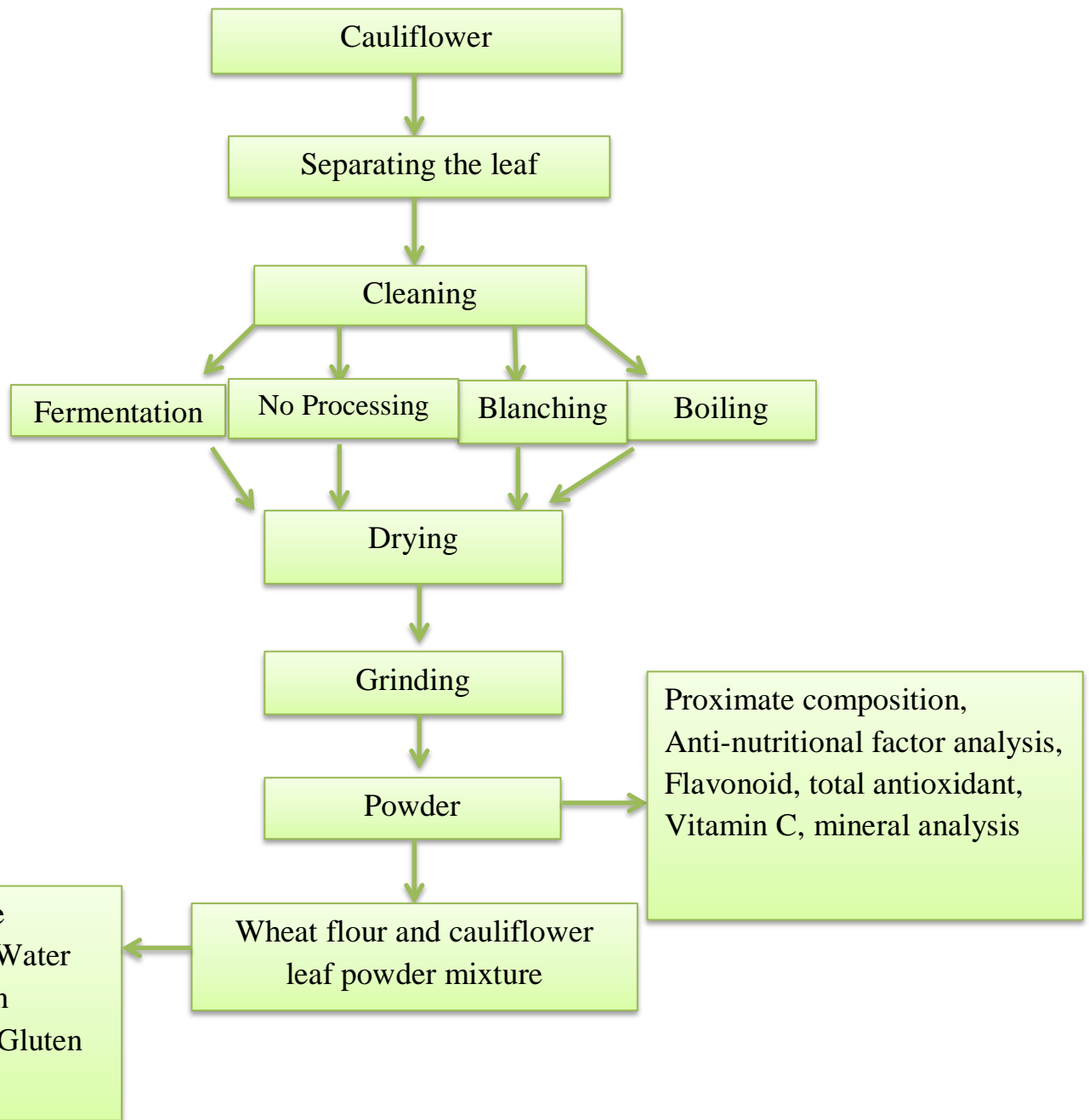


Figure 5: Diagrammatic representation of sample preparation for laboratory analysis.

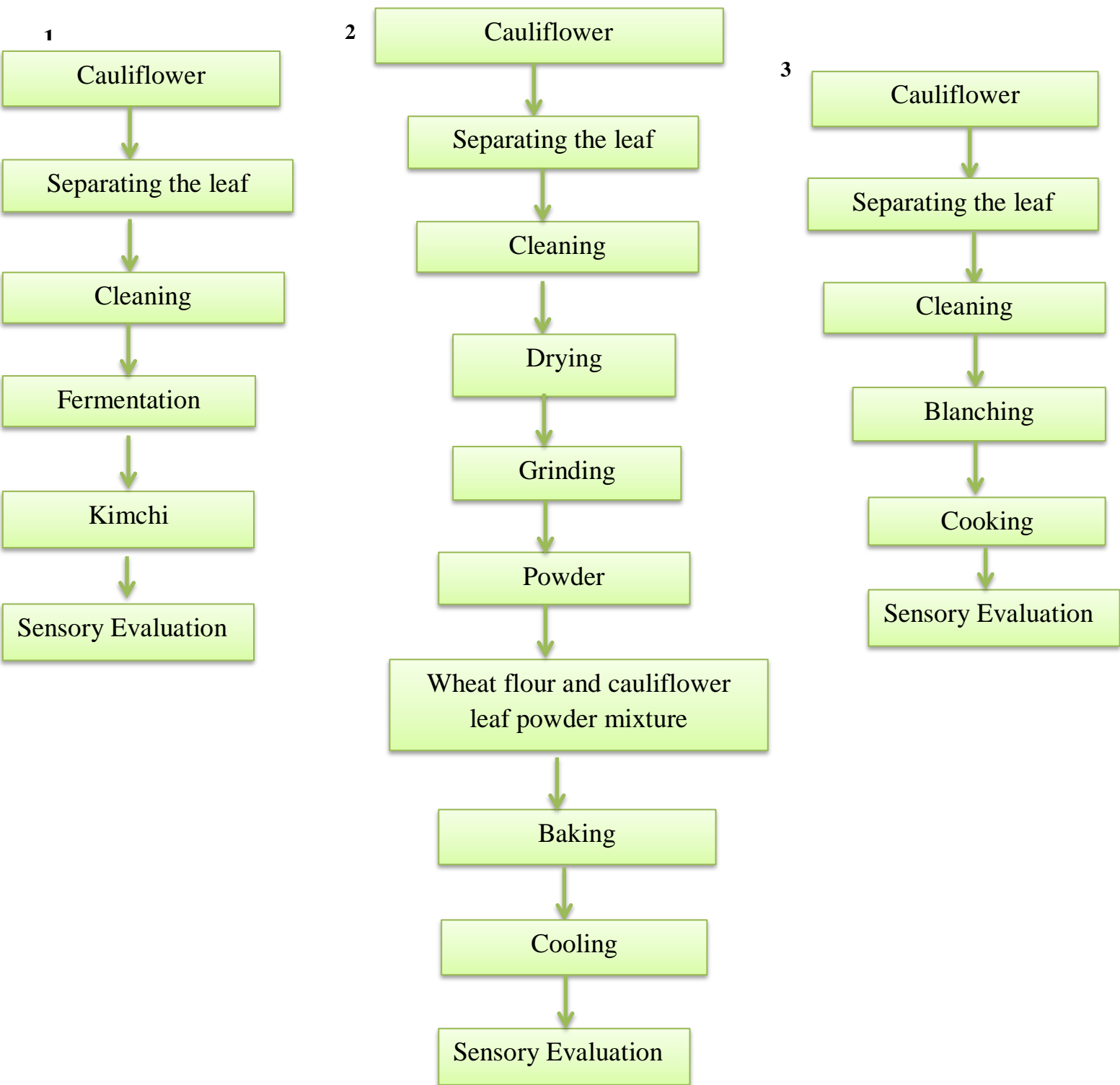


Figure 6: Diagrammatic representation of sample preparation for sensory evaluation, 1-kimchi preparation, 2-Bread preparation and 3-Meal preparation.

3.4 Analytical Method

3.4.1 Determination of Proximate Composition

3.4.1.1 Crude Protein

Crude protein was determined using the three steps known as Digestion, Distillation and Titration (AOAC 2005).

Digestion: 0.5g of sample was placed in digester flask and 6ml of sulphuric acid, hydrogen peroxide and a mixture of copper sulphate and potassium sulphate were added and the flask was placed in the digester machine for 4 hours at 370 °C.

Distillation: it took place after digestion and the digester flask was placed inside the distillation machine and the machine was adjusted to add 25ml of 40% sodium hydroxide, 25ml of boric acid and 25ml of distilled water and an indicator solution and then distillation was over when the total volume reached 250ml.

Finally titration process was done by using 0.1N HCl till it reached reddish colour.

$$\text{Nitrogen (\%)} = \frac{V_{\text{HCl in L}} \times N_{\text{HCl}} \times 14 \times 100}{W}$$

Where; V- Volume of HCl in L consumed to the end point of titration

N- The normality of HCl

W- Sample weight on dry matter basis

14- The molecular weight of nitrogen

The % of Nitrogen was converted to % of protein by using appropriate conversion factor by multiplying the result by 6.25.

3.4.1.2 Crude Fat

Fat content of each sample was determined according to AOAC 2005; 2g of each sample was measured on fat free cotton and placed in extraction thimble. The extraction cylinder was weighted and 50ml of hexane was added before putting the extraction thimble that holds the sample. Then the extraction cylinder was moved into the heating plank and kept in there for 4 hours at 80 °C.

After 4 hours of extraction the extraction cylinder was placed in a drying oven at 70 °C for 30 minutes and then in a desiccator to cool and was measured after cooling.

$$\% \text{ Crude fat} = \frac{(W_2 - W_1)}{W} \times 100$$

Where: W_1 = weight of empty flask,

W_2 = weight of flask and extracted fat, and

W = weight of sample

3.4.1.3 Ash content

Ash content of each sample was determined using AOAC 2005; 2.5g of each sample was measure in a clean porcelain crucible after it was measured. Then the sample was charred on a hot plate until smoking ceased and placed in a muffle furnace at 550°C for 5 hours.

After 5 hours, when the ash is clean and white in appearance, it was cooled at room temperature and weighted.

$$\text{Total ash (\%)} = \frac{M_3 - M_1}{M_2 - M_1} \times 100$$

Where: M_1 - weight of the crucible

M_2 - weight of the crucible after the sample is added

M_3 - weight of the crucible with ash

3.4.1.4 Moisture content

Moisture content of each sample was determined using AOAC 2005; a clean crucible was weighted and 5g of each sample was added on it. The crucible was placed in a drying oven at 105°C for 3hrs.

After 3 hours it was cooled in a desiccator and measured. The sample was again placed in the drying oven for 1 hour and then cooled in a desiccator and measured; this was repeated until a constant weight was obtained.

$$\text{Moisture content (\%)} = \frac{W_2 - W_3}{W_2 - W_1} \times 100$$

Where: W_1 - weight of the crucible

W_2 - weight of the crucible when sample is added

W_3 - the final weight

3.4.1.5 Crude Fiber

Crude fiber of each sample was determined according to AOAC 2005; 12.5% of NaOH and 12.5% of sulphuric acid was prepared in 1000ml volumetric flask. 1g of each sample was placed in a beaker and 200ml of sulphuric acid was added. Then the sample was placed on hot plate and boiled for 37 minutes, after that the acid was drained using vacuum pump. The sample was cooled for 5 minutes and washed three times by using distilled water. The sample was again placed in a beaker and 200ml of 12.5% of NaOH was added and boiled on hot plate for 37 minutes. After 37 minutes the base was drained using vacuum pump.

The residue was placed in a dry and clean crucible and set in oven at 130°C for 2 hours. After 2 hours the crucible was transferred into muffle furnace for 3 hours at 550°C after being measured.

After 3 hours the crucible was cooled at room temperature and weighted.

$$\text{Crude fiber g/100g} = \frac{W_2 - W_3}{W_1}$$

Where: W_1 - weight of sample

W_2 - weight of crucible plus sample after drying in oven

W_3 - weight of crucible plus sample after ashing

3.4.2 Determination of mineral

Mineral analysis was done according to AOAC 2020; 5g of each sample was charred to remove organic matter then ash content was determined by placing the charred samples in the muffle furnace at 550°C for 5 hours, then 3 drops of 1M HNO₃ acid and few drops of distilled water was added. Ash was designated by using 3N and 6N HCl then the digested sample was filtered into sample bottles using whattmans filter paper prior to analysis after filtration made up the volume to 50ml with distilled water.

For Ca determination, 2.5ml of 10% LaCl₃ was added, the Fe, Zn and Ca content of each sample was determined using AAS. Calibration curve was prepared by plotting the absorption or emission values against the metal concentration in mg/100g and finally, reading was done from the graph which represents the metal concentrations that correspond to the absorption or emission value of the samples and the blank.

Metal content was calculated:

$$\text{Metal content (mg/100g)} = \frac{A-B}{10W} \times V$$

Where: A- Concentration of sample solution

B- Concentration of blank solution from curve in ppm

V- Volume of extract

W- Weight of sample

3.4.3 Determination of Antioxidant and Flavonoid

Extraction of the leaf: AOAC 2005 was used for the determination of antioxidant and flavonoid: 5g of the leaf powder was placed in Erlenmeyer flask and 50 ml of methanol was added, then the flask was placed on a shaker and left there for 24hrs.

After 24hrs the sample was filtered and again placed in a shaker for 2 hours. After 2hrs the sample was filtered again and was placed in a round bottom flask after the flask was washed, dried and measured.

The round bottom flask was placed on water evaporator and waited for the entire methanol to evaporate, and then the flask was measured again.

The difference between the empty flask and after it contain the sample and undergo evaporation was calculated and that number was multiplied by 1000 and divided by 50 and the result of that much ml of methanol was added in the flask to rinse the sample on it and was placed in a brown glass in a refrigerator for further analysis.

Total flavonoid content was determined by first adding 20 μ l of leaf extracted into test tubes followed by 980 μ l of methanol and 1 μ l of 10% aluminium chloride. After 30 minutes at room temperature and undergoing vortex the absorbance of the reaction mixture was measured at 415nm using UV-Vis Spectrophotometer.

Antioxidant capacity was determined by preparing mixture that consisted 10 μ l – 160 μ l of leaf extract, 990 μ l - 840 μ l methanol and 4ml DPPH solution into test tubes. The mixture was incubated in the dark for 30 minutes and vortex. The absorbance was measured at 517nm using UV-Vis Spectrophotometer.

3.4.4 Determination of vitamin C

Vitamin C was determined according to AOAC 2005 method: 5 g of each sample was added into 100ml volumetric flask and 50ml of 3% Metaphosphoric acid was added then it was vortex for 5 minutes and sonicate for 10 minutes to mix well. After 10 minutes 50ml of 3% Metaphosphoric was added until it reaches 100ml and shake. The sample was filtered using whatmans filter paper and finally 1ml solution was taken into an injection vial by filtering through syringe filter. The absorbance was measured using HPLC.

3.4.5 Determination of water Absorption capacity

Water absorption capacity was determined according to Aremu *et al.*, (2007). One gram of sample was mixed with 10ml of distilled water in a centrifuge tube and allowed to stand at room temperature for 1 hour. It was then centrifuged at 200 rpm for 30 minutes and the supernatant was transferred into 10 ml gradual cylinder.

Water absorption capacity (ml) = volume of water added – volume of water decanted

3.4.6 Determination of Anti-nutritional Factors

3.4.6.1 Determination of Oxalate

Oxalate was determined by using 1g of cauliflower leaves powder and mixed with 30 ml of 1ml HCl. Each mixture was then shaken in a water bath at 100⁰C for 30 minutes. To each mixture was added 0.5ml 5% Calcium Chloride and thoroughly mixed to precipitate out the calcium oxalate. The suspension was centrifuged at 800rpm for 15 minutes and the supernatant poured out. The pellet was washed twice with 2ml of 0.35M NH₄OH then dissolve in 0.5M H₂SO₄. The solution was titrated with standard solution of 0.1M KMnO₄ with the temperature being maintained at 60⁰C to a faint violet colour that persisted for at least 15 seconds (Abdel-Moemin, 2014). The concentration of oxalate in each sample was obtained from the following calculation: 1 mL 0.1 N KMnO₄ = 0.006303 g oxalate.

3.4.6.2 Determination of Phytate

Phytate content was analyzed using Wade method as described by Latta and Eskin (1980) and later modified by Vaintraub and Lapteva (1988). Series of phytic acid standard solution (4, 8, 16, 24, and 32 µg/g) was prepared using phytic acid (sodium phytate). Then, 3 ml of each standard solution was added into 15 ml of centrifuge tubes and mixed with 2 ml of Wade reagent using vortex. Then the standard curve was plotted using absorbance versus concentration graph. For sample phytic acid analysis, about 1 g of dried sample was extracted

with 10 ml 0.2 N HCl for 1 hour at ambient temperature and centrifuged at 3000 rpm. Then 3 ml of clear supernatant was mixed with 2 ml of Wade reagent using vortex. The absorbance at 500 nm was measured using a UV-Vis spectrophotometer, (UV-Vis, Lambda 9500, Malaysia). The amount of phytic acid was calculated using a phytic acid standard curve and the result was expressed as phytic acid in $\mu\text{g/g}$ fresh weight.

$$\text{Phytic acid in } \mu\text{g/g} = \frac{[(Ab - As) - \text{Intercept}] \times 10}{\text{Slope} \times W \times 3}$$

Where:

As: sample absorbance

Ab: blank absorbance

W: weight of sample

3.4.7 Determination of gluten content

Wet gluten content was determined by using hand washing AACC, 2000 method: 10 grams of each sample was weighed and transferred to plastic mixing chamber and 4.8ml of 2% sodium chloride was added. Mixing and kneading using hands was continued until a rubbery, soft ball of dough was obtained. The dough was washed under running water to wash out starch, water and salt soluble protein via murky like water. Then the gluten was press-dried between hands and rolled in to a ball. The weight of the remaining rolled ball of flour is the amount of gluten per 100 gram.

3.4.8 Determination of molar ratio of phytate to Ca, Zn and Fe

The mole of phytate and mineral was determined by dividing the weight of phytate and minerals with their atomic weight (phytate: 660g/mol, Ca: 40g/mol, Fe: 56g/mol, Zn: 65g/mol). The molar ratio between phytate and mineral was obtained after dividing the mole of phytate with the mole of minerals (Morris & Ellis, 1989).

3.4.9 Sensory Evaluation

The different breads (F1 to F6) were rated by 10 panellists on a nine point hedonic scale and they were given a rank. The evaluation was done based on appearance, aroma, taste, mouth feel, and overall acceptability as, 1- dislike extremely, 2- dislike very much, 3- dislike moderately, 4- dislike slightly, 5- neither like nor dislike, 6- like slightly, 7- like moderately, 8- like very much and 9- like extremely. Prior to the evaluation, the panellists were semi-trained and were given information about the 9-point hedonic scale and how to utilize it. Each panellist sat in their own private booth, which offered a calm and friendly atmosphere.

3.4.10 Ethical Clearance

Ethical clearance was approved by the College of Natural and Computational Sciences Institutional Review Board on 25/05/2023, Minute No.IBR/05/2015/2023.

3.5 Data Analysis

SPSS, version 20 was used for data analysis using analysis of variance (ANOVA), Duncan and LSD method was used to identify the significant difference between the results.

4. Results and Discussion

4.1 Proximate composition

4.1.1 Moisture content

The results of the proximate composition of raw and processed cauliflower leaves are presented under table 7 in g/100g.

Table 7: Proximate composition of cauliflower leaves under different processing methods (g/100g).

Sample Type	Moisture	Crude Protein	Crude Fat	Crude fiber	Total Ash	Carbohydrate	Energy (Kcal)
Raw	11.95±0.63 ^a	32.45±0.35 ^a	1.75±0.35 ^a	23.5±0.70 ^a	22.8±0.28 ^b	7.85±0.35 ^d	173.76±0.01 ^d
Boiled	11.37±0 ^a	26.21±0.30 ^c	1.25±0.35 ^b	12.5±0.70 ^c	24.4±0.90 ^b	24.07±0.85 ^b	212.37±0.96 ^c
Blanched	8.02±0.11 ^b	29.25±0.25 ^b	1.15±0.35 ^b	9.5±0.70 ^d	20.9±0.84 ^c	31.18±0.36 ^a	252.07±0.86 ^b
Fermented	4.33±0.19 ^c	20.13±0.24 ^d	1.75±0.35 ^a	21.65±0.49 ^b	42.74±0.4 ^a	9.4±0.49 ^c	267.21±0.19 ^a

Note: all data are reported as dry weight basis

All values are the means of duplicate ± SD

a, b, c, d are superscripts given to show the significant difference between means within column

Moisture content of the cauliflower leaves ranged from 4.33–11.95g/100g (Table 7). However, the moisture content of the blanched and the fermented leaves was significantly different from the raw leaves powder ($P<0.05$), and the lowest was observed in the fermented and blanched leaves (Table 7) while the raw leaves were with the highest content. The high moisture content in the raw leaves is not surprising given that moisture is the most single constituent of vegetables (Umerah *et al.*, 2019). The decrease in moisture content after undergoing different processing method might be due to softening of the tissues that occur during boiling, blanching and fermentation, resulting in enhanced moisture removal (Mythili *et al.*, 2021). The lower moisture content in the processed leaves is an indication of the possible positive effect of processing on leafy vegetables shelf-life, quality, freshness and resistance to bacteria (Mythili *et al.*, 2021).

4.1.2 Crude Fat content

The range of fat content in the cauliflower leaves were 1.75–3.25g/100g. The lower fat content in the processed leaves could be due to the loss of water soluble fatty acids and volatile compounds during boiling and blanching. Similar result was reported by Mythili *et al.*, (2021) on crude fat content of cauliflower leaves after blanching. Generally, vegetables are not good sources of fat. Consumption of vegetables may help reduce the high incidence of

obesity, diabetes, cardiovascular diseases and high blood pressure, which are associated with a high intake of fatty foods (Lewu *et al.*, 2009). When compared to other common green leafy vegetables such as in a study by Sheela *et al.*, (2004) where 38 green leafy vegetables were analysed, raw and fermented leaves of cauliflower showed higher fat content than 31 vegetables and lower fat content than to the other 7 vegetables. Greater fat content of the leaves may prove sufficient for tissue repair, regulation of body processes and the biosynthesis of important enzymes and hormones (Mobeen *et al.*, 2021).

4.1.3 Crude Fiber content

The fiber content of the cauliflower leaves ranged from 9.5 – 23.5g/100g (Table 7). The recommended adequate intake for total fiber for adults is 25-38g/day (Hoy & Goldman, 2014), on another study by Storey & Anderson, (2014) states that an intake of 14g dietary fiber per 1000Kcal would promote heart health, it can be said that cauliflower leaves are one good source of fiber. The raw and the fermented leaves were with the highest fiber content when compared to the leaves that underwent blanching and boiling. The result of this study is in line with Yakubu *et al.*, (2012) which showed increment of fiber content in spinach and another green leafy vegetable following fermentation. Amount of fiber content of cauliflower leaves in this study is higher when compared to commonly green leafy vegetables such as Spinach (2.5g/100g), Cabbage (2.8g/100g), Amaranth (4g/100g), Mint (6.3g/100g) and 9 other vegetables in a study by Punna & Rao Paruchuri, (2004). The higher amount of dietary fiber contributes significantly to health since fiber lowers the body cholesterol level (Arasaretnam *et al.*, 2018). In addition, consumption of vegetables with higher fiber content will keep the digestive system healthy, helps healthy body weight and reduces risk of cardiovascular diseases (Kumar *et al.*, 2020).

4.1.4 Crude Protein content

The crude protein content of the raw cauliflower leaves were 32.45 ± 0.35 , which was significantly higher than the leaves that underwent boiling, blanching and fermentation processes (table 7). The various processing techniques caused a significant ($P \leq 0.05$) decrease in the crude protein content. The decrease could be attributed to the fact that some of the proteins were leached off by the water and due to the denaturation and solubilisation during processing (Yakubu *et al.*, 2012; Murcia *et al.*, 1992). However, the crude protein content in this study was within the range of previous report for leafy vegetables with high protein content (Lewu *et al.*, 2009) that makes it one of the cheap source of plant protein.

4.1.5 Total Ash content

The ash content of the cauliflower leaves ranged from 22.8-42.74g/100g with significant difference ($P \leq 0.05$) from the raw leaf after going through blanching and fermentation procedures (Table 7). Compared to the raw leaves, leaves that underwent blanching had the lowest ash composition. The lowest ash composition in the blanched leaves may be because of water absorption that might dilute the ash content and reduce the potential of the leaves to supply essential minerals (Lewu *et al.*, 2009).

4.1.6 Carbohydrate content

The carbohydrate content of the raw cauliflower leaf was 7.85 ± 0.35 g/100g. The low carbohydrate content is not only for cauliflower leaves but it is peculiar to other vegetables (Umerah *et al.*, 2019). However, cauliflower leaves that underwent boiling, bleaching and fermentation processes were with higher carbohydrate content compared to the raw leaves (table 7). The increase in carbohydrate content might be due to the decrease in moisture, fiber, fat and protein content, because amount of carbohydrate was calculated by subtracting the sum of them from 100, their value have an effect on carbohydrate amount. The recommended daily intake of carbohydrate values for people aged 4 years and older is 300g hence, the carbohydrate values of boiled or blanched cauliflower leaves can help meet the daily requirements if consumed often in reasonable quantity (Osum *et al.*, 2013).

4.1.7 Total Energy content

Cauliflower leaves can be considered as a good source of energy specially when undergoing different processing methods. Table 7 showed that the energy level of cauliflower leaf showed a significant ($P \leq 0.05$) increase from 173.76 ± 0.01 to 267.21 ± 0.19 Kcal when boiled, blanched and fermented. This result showed that cauliflower leaves have a higher energy content when compared to *Amaranthus cruentus*, *Celusia argenta* and *Corchorus olitorius* which have 176.67, 174.93 and 177.55 kcal respectively (Onwordi *et al.*, 2009). Another study by Kumar *et al.*, (2020) where nutritional components of 8 commonly used green leafy vegetables assessed cauliflower leaves when boiled, blanched and fermented showed a higher energy content than spinach, bathua and cabbage. The recommended daily caloric intake for male and female children, male and female adults is 1000-2600Kcal, 1000-2000Kcal, 2000-2800Kcal and 1800-2200Kcal respectively (Faizan & Rouster, 2020). However, children and mothers are reported to have inadequate intakes energy (Abebe *et al.*, 2018); therefore, consuming cauliflower leaves after processing could be one way of changing this report.

4.2 Proximate composition of bread products

The proximate composition of bread made from different proportion of wheat flour (*Var. Triticum aestivum*) and cauliflower leaves powder resulted that, the addition of cauliflower leave powder in to the wheat bread enhanced the crude protein and the crude fiber content from 7.91 to 10.7g/100g) and from 1.75 to 4.5g/ 100g respectively and a decrease in carbohydrate content (74.72 to 68.83g/100g) was observed (table 8).

Table 8: Proximate composition of wheat bread made with different proportion of raw cauliflower leaf powder (g/100g).

Sample type	Moisture	Crude Protein	Crude Fat	Crude Fiber	Total Ash	Carbohydrate	Energy (Kcal)
F1	13.78±0.31 ^a	10.76±0.12 ^a	1±0 ^{bc}	4.5±0.70 ^a	1.22±0.007 ^b	68.83±0.89 ^c	327.4±0.07 ^b
F2	13.47±0.10 ^{ab}	10.4±0 ^a	2.25±0.35 ^a	3.75±0.35 ^{ab}	3.09±0.16 ^a	67.03±0.97 ^d	329.97±0.72 ^b
F3	13.1±0.07 ^b	10.34±0.22 ^a	1.25±0.35 ^{bc}	2.75±0.35 ^{bc}	3.18±0.26 ^a	69.37±0.29 ^c	330.11±0.11 ^b
F4	13.35±0.50 ^{ab}	9.71±0.12 ^c	1.25±0.35 ^{bc}	2±0.70 ^c	3.29±0.11 ^a	70.39±0.54 ^c	331.67±0.49 ^b
F5	13.96±0.05 ^a	9.01±0.37 ^d	1.75±0.35 ^{ab}	2±0 ^c	1.02±0.26 ^b	72.26±0.30 ^b	340.85±0.49 ^a
F6	13.93±0.01 ^a	7.91±0.30 ^b	0.75±0.35 ^c	1.75±0.35 ^c	0.94±0.12 ^b	74.72±0.51 ^a	337.29±0.07 ^a

Note: all data are reported as dry weight basis

All values are the means of duplicate ± SD

a, b, c, d are superscripts given to show the significant difference between means within column

F1-91:9 whole wheat flour: cauliflower leaf powder

F2-93:7 whole wheat flour: cauliflower leaf powder

F3- 95:5 whole wheat flour: cauliflower leaf powder

F4- 97:3 whole wheat flour: cauliflower leaf powder

F5- 99:1 whole wheat flour: cauliflower leaf powder

F6-100:0 whole wheat flour: cauliflower leaf powder

4.2.1 Moisture content

The moisture content of wheat bread enriched with different proportion of cauliflower leaves powder ranged from 13.96 – 13.1g/100g (table 8). Nevertheless, even if the moisture content showed a decrease it was not significantly ($P \leq 0.05$) different in the breads, compared with the whole wheat bread, except for the bread with 95% whole wheat flour (F3) that was with the lowest moisture content. The lowest moisture content in F3 could be an indication of a better shelf life (Odunlade *et al.*, 2017), in breads enriched with the powder compared to the bread that was not enriched. This result goes in line with other researches such as Odunlade *et al.*, (2017), which studied about the chemical compositions and quality characteristics of wheat bread supplemented with different green leafy vegetables and states that the moisture content of the enriched bread significantly ($p < 0.05$) decreased (26.9 to 21.9 %) as the substitution level of fluted pumpkin, African eggplant and amaranth increased.

4.2.2 Crude Fat content

Fat content of bread ranged from 0.75 – 2.25g/100g with different proportion of cauliflower leaves powder and wheat flour (table 8). Even if there was an increase in fat content of all the bread samples that incorporated cauliflower leaf powder when compared to the one that is formed by 100% whole wheat flour the result does not have a consistent pattern so it is hard to say the increase in substitution level could result an increase in fat content as well, most of the results weren't significantly different at $P \leq 0.05$ but the fat content of 100% whole wheat flour bread (F6) is significantly different at $P \leq 0.05$ from F5 (99% whole wheat flour and 1% cauliflower leaf powder) and F2 (93% whole wheat flour and 7% cauliflower leaf powder), this can be an indication that the addition of cauliflower leaf powder happens to increase the fat content of bread, this observation was also reported by Bolarinwa *et al.*, (2019). As another research by Odunlade *et al.*, (2017) states that it means F5 and F2 would be distinctively palatable in taste than the control bread sample because fat improves palatability.

4.2.3 Crude Fiber content

The fiber content consistently increased as the proportion of the cauliflower leaves powder added to the wheat flour increased. The increase was from 1.75 ± 0.35 (F6) to 4.5 ± 0.70 (F1). But compared to the bread made with flour 100% wheat statistically significant increased fiber content was observed in F1 and F2 (Table 8). Large portion of fiber in the leaves of cauliflower was found (table 7), increased amount of fiber content as the substitution level increases shows that the fiber content of the leaves were not degraded by either the fermentation by yeasts or by the baking process, this result was also observed in a study by Nudel *et al.*, (2023) where incorporated *Moringa oleifera* leaf powder and gluten was used in developing nutrient rich and functional wheat bread. The highest amount of fiber content was seen at F1 (91% whole wheat flour and 9% cauliflower leaf powder), F2 (93% whole wheat flour and 7% cauliflower leaf powder) and F3 (95% whole wheat flour and 5% cauliflower leaf powder) respectively, the increase in amount of fiber content when different proportion of cauliflower leaves powder can result in the production of fibrous bread, these kinds of bread have been associated with reduced cardiovascular diseases risk, lower risk of type 2 diabetes and reduced risk of obesity, overweight and high waist to hip ratio (Smith & Tucker, 2011).

4.2.4 Crude Protein content

As indicated under table 8, the crude protein content of the enriched bread showed a steady increment compared to the bread made with wheat flour only and it was significant at $P \leq 0.05$. The increment in the protein content of the enriched bread could be attributed to the high protein content of the cauliflower leaves (Table 7). Addition of protein ingredients to enrich bread could reduce starch digestibility, which helps to balance energy and decrease the glycemic index, which helps to lose weight or keep healthy weight (Prieto-Vázquez del Mercado *et al.*, 2022).

4.2.5 Total Ash content

With the addition of the cauliflower leaves powder, there was a corresponding increase in the ash content of the bread compared to the bread made with the whole wheat flour (Table 8). Particularly F3, F2 and F1 were with statistically higher ash content ($P \leq 0.05$) in relation to F6. The statistically significant increase in the ash content could be due to the high mineral content of cauliflower leaves given amount of ash is an indicator of the vegetable mineral composition that could be provided through its consumption (Mohammed Abdus Satter *et al.*, 2016).

4.2.6 Carbohydrate content

The carbohydrate content of wheat bread with different proportion of cauliflower leaf powder is presented under table 8. The content ranges between 67.03-74.72g/100g. However, there was a significant decrease in carbohydrate composition as the proportion of cauliflower leaf powder added to the wheat flour increases. A similar decline in carbohydrate content of wheat bread was also reported by Odunlade *et al.*, (2017) and Bolarinwa *et al.*, (2019) with addition of vegetable powder. The observed decrease might be due to the low carbohydrate content in vegetables as compared to the wheat flour (Ujong *et al.*, 2023).

4.2.7 Total Energy content

Energy content becomes lower as the amount of cauliflower leaves powder incorporated increased. As shown in table 8 energy Kcal ranged between 327.4- 340.85, the lowest amount of energy was observed in the formulation with the highest leaves powder. These decreases in calories may be attributed to the high quantities of dietary fiber in cauliflower leaves powder (Mohammed *et al.*, 2023).

4.3 Anti-nutritional factors

4.3.1 Oxalate and phytate contents

Oxalate and phytate content of cauliflower leaves showed a significant decrease from 2.7 to 0.78mg/100g and from 1.15 to 0.51mg/100g respectively after boiling, blanching and fermentation processes (table 9).

Table 9: Anti-nutritional factors of cauliflower leaf under different processing methods

Sample type	Oxalate (mg/100g)	Phytate (mg/100g)
Raw	2.74±0.13 ^a	1.15±0.02 ^a
Boiled	1.27±0.08 ^b	0.96±0.17 ^b
Blanched	1.93±0.13 ^b	0.82±0.31 ^c
Fermented	0.78±0.22 ^c	0.51±0.07 ^d

Note: all data are reported as dry weight basis

All values are the means of duplicate ± SD

a, b, c, are superscripts given to show the significant difference between means within column

The mean oxalate content of the raw cauliflower leaf was 2.74±0.13mg/100g; the content was significantly lower for leaves that underwent boiling, blanching and fermentation compared to the raw leaves ($P \leq 0.05$), leaves that went through the fermentation procedure showed the least amount of oxalate content (Table 9), a similar result that revealed a decrease in oxalate content of fermented moringa olifera leaves was reported in the findings of Umerah *et al.*, (2019). In a study by Savage *et al.*, (2000), it is stated that cooking reduces the oxalate content of the food by leaching losses into the cooking water. In addition, the range of oxalate content (0.78 – 2.74 mg/100g) in the leaf was lower than what was reported as the toxic range or lethal dose to be between 3-5g per day for human (Ekop *et al.*, 2008). Therefore, oxalate content of cauliflower leaves should not be a risk factor upon consumption.

Likewise, the phytate content of the raw cauliflower leaves were 1.15±0.02mg/100g, but after boiling, blanching and fermentation it showed a significant decrease to 0.96±0.17, 0.82±0.31, 0.51±0.07 mg/100g respectively (table 9). The decrease in phytate content after processing was in agreement with the previous report by Mosha *et al.*, (1995). And, leaching that is enhanced by the high temperature and prolonged time of soaking could be the possible mechanism that decreased phytate in the processed leaves.

4.4 Antioxidant activity, Flavonoid and Vitamin C content

Amount of flavonoid showed a significant decrease after boiling and blanching from 227.45 to 181.7mg/100g and to 70.96mg/100g respectively and showed a significant increase to 341.8mg/100g under fermentation process. Vitamin c amount of the leaves showed a significant decrease after they underwent through different processing techniques when compared to raw form of the leaves (table 10).

Table 10: Flavonoid and Vitamin C content of cauliflower leaf under different processing methods.

Sample type	Flavonoid (mg/100g)	Vitamin C (mg/100g)
Raw	227.45±0.90 ^a	129.9±0.42 ^a
Boiled	181.7±0.72 ^b	9.19±0.72 ^c
Blanched	70.96±0.76 ^c	50.69±0.73 ^b
Fermented	341.8±0.85 ^d	24.51±0.89 ^d

Note: all data are reported as dry weight basis

All values are the means of duplicate ± SD

a, b, c, d are superscripts given to show the significant difference between means within column

4.4.1 Flavonoid content

Flavonoid content of processed and unprocessed Cauliflower leaves is depicted under Table 10. The flavonoid content of the leaves ranged from 70.96mg/100g to 341.8mg/100g though the content showed a significant decrease when boiled and blanched. The decrease in the content of the compounds, following, boiling and blanching, may be due to the loss of total antioxidant given the leaves are with a large surface area that increases the area exposed to the boiling water, which could enhance solubilisation and leaching (Chan *et al.*, 2014). On the other hand, after fermentation, the total flavonoid content significantly increased ($p \leq 0.05$). The increase in total flavonoid content of the leaves following fermentation could be because of the activity of some microorganisms which have the ability to degrade the polyphenol compounds (Dwiputri & Feroniasanti, 2019).

4.4.2 Vitamin C

The average vitamin C value for the raw cauliflower leaf was 129.9±0.42 (Table 10). The value was higher than other leafy vegetables such as Lettuce (8mg/100g); Spinach (51mg/100g); and Beets (34mg/100g) (Salazar *et al.*, 2006). But leaves that were boiled (9.19±0.72), blanched (50.69±0.73) and fermented (24.51±0.89) had lower vitamin C value compared the raw. The decrease in vitamin C content after processing is not surprising considering that vitamin C is water soluble and heat labile (Babalola *et al.*, 2010) . According

to National Institute of Health (2019) the daily recommendation of vitamin C for teen boys 75mg; girls (14-18 years) 65mg; adult men 90mg; adult women 75mg; pregnant teens 115mg; women 85mg breast feeding teens and women 120mg. Therefore, the higher vitamin C content of cauliflower leaf makes it a good candidate to fulfil the daily recommended intakes for people in different age group and physiological state.

4.4.3 Total antioxidant

Cauliflower leaves under different processing methods were tested for their anti-oxidant capacity using DPPH assay and the result of the analysis showed that the raw, boiled, blanched, fermented and ascorbic acid standard oxidation inhibition capacity ranging from 88% to 18%, 80% to 11%, 80% to 18%, 43% to 13% and 97% to 33% respectively (figure 7).

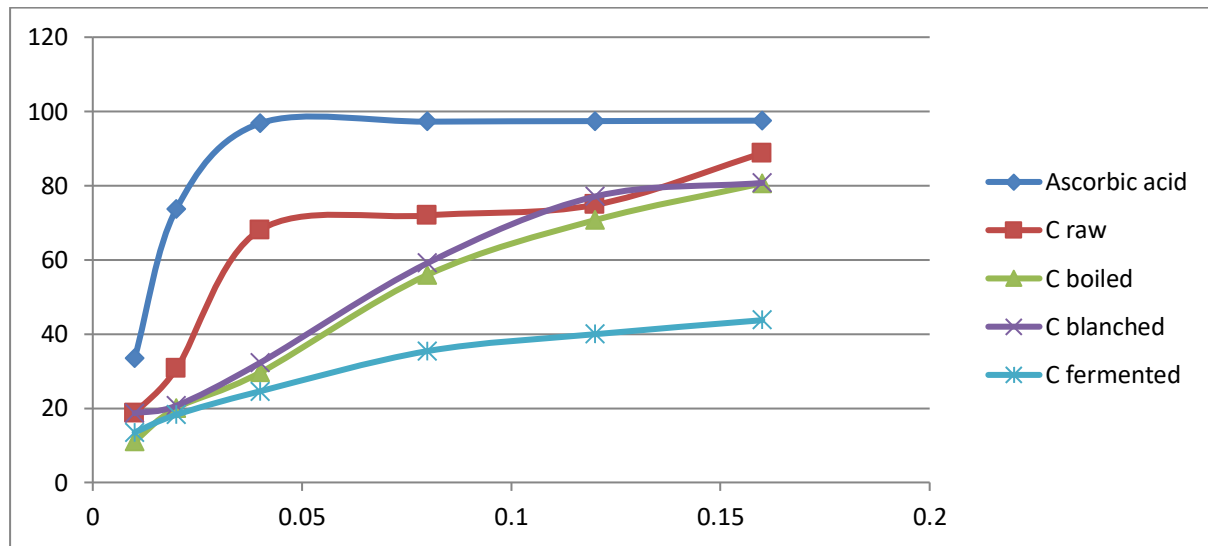


Figure 7: Antioxidant activity of Cauliflower leaves and ascorbic acid standard curve

Table 11: Antioxidant capacity of cauliflower leaf under different processing methods

Sample type	Antioxidant Capacity (mg/100g)
Raw	58.81±0.61 ^a
Boiled	44.66±0.48 ^a
Blanched	48.09±0.91 ^a
Fermented	29.23±0.27 ^b

Note: all data are reported as dry weight basis

All values are the means of duplicate ± SD

a, b, c, d are superscripts given to show the significant difference between means within column
The loss in antioxidant capacity after processing (table 11) could be due to the large surface area of the vegetables that would be in contact with the boiling water that results in

solubilisation and leaching. The higher content of the antioxidant in cauliflower leaf could be beneficial for anti-aging and anti-inflammatory activities that makes them scientifically interesting compounds (Zehiroglu & Ozturk Sarikaya, 2019).

4.5 Iron, zinc and calcium contents

Cauliflower leaves showed a significant difference ($p \leq 0.05$) on zinc composition (0.54-2.67), calcium (166.87-498.12) and iron (6.65-9.91) content when treated under different processing methods (table 12).

Table 12: Iron, zinc and calcium composition of raw and processed cauliflower leaf

No	Sample type	Zinc (mg/100g)	Calcium(mg/100g)	Iron (mg/100g)
1	Raw	1.55±0.09 ^c	283.94±0.84 ^c	7.34±0.01 ^b
2	Boiled	1.93±0.14 ^b	425.31±0.94 ^b	9.68±0.52 ^a
3	Blanched	0.54±0.09 ^d	166.87±0.59 ^d	6.65±0.02 ^b
4	Fermented	2.67±0.04 ^a	498.12±0.18 ^a	9.91±0.49 ^a

Note: all data are reported as dry weight basis

All values are the means of duplicate ± SD

a, b, c, d are superscripts given to show the significant difference between means within column

4.5.1 Zinc

The zinc content of the cauliflower leaves is presented under table 12. The average zinc composition of the raw leaves was 1.55±0.09 which is higher than the commonly known leafy vegetables like lettuce (0.16mg/100g) and Spinach (0.22 mg/100g) (Salazar *et al.*, 2006). But the composition reduced after boiling (1.93±0.14) and blanching (0.54±0.09) processes. On the contrary, zinc content of the fermented samples was significantly higher (2.67±0.04) ($P \leq 0.05$) than the raw and the other processed leaves. The higher zinc content in the fermented leaves may be because of the increase in the activities of microfloral enzymes that hydrolyses the zinc that is bound to protein to release the zinc for utilization (Umerah *et al.*, 2019).

4.5.2 Calcium

Raw cauliflower leaves contained 283.94±0.84g/100g calcium on average (Table 12) which is higher than the other leafy vegetables such as Lettuce (35 mg/100g), Spinach (106 mg/100g) and Beets (110 mg/100g) (Salazar *et al.*, (2006). The content is still higher than the vegetables after boiling (425.31±0.94); blanching (166.87±0.59) and fermentation (498.12±0.18) processes. Calcium in the body is used as a structural components of bones and teeth, which represent about 40% of all minerals present in the body (Osum *et al.*, 2013). According to NIH (2023) the daily recommended amount of calcium for teens (14-18years) is

1300mg and for adults (19-50years) is 1000mg, this numbers shows that cauliflower leaves can be used as one sources of calcium.

4.5.3 Iron

The iron content of the raw cauliflower leaf was 7.34 ± 0.01 mg/100g on average. The content was higher than the commonly known vegetables such as Lettuce (2 mg/100g); Spinach (3.1 mg/100g); and Beets (2.7 mg/100g) Salazar et al., (2006). Conversely, the iron content of the leaf under different processing methods ranged from 6.65 – 9.91 mg/100g. The highest amount of iron was observed after fermentation and the lowest was observed following blanching processes (6.65 ± 0.02) (table 12). The higher iron content in the fermented leaves could be caused by lactic acid bacteria (LAB) that results in the release of minerals from their chelated complex compounds. On the contrary, the low level of mineral in the blanched leaves could be due to leaching out through the discarded blanching medium (Mongwaketse, 2021). Assuming low bioavailability, the average iron in 100g of the processed cauliflower leaf could provide more than 70% of the recommended daily iron for teen boys and girls as well as for adult men and women respectively NIH (2023), which will contribute to reduce iron deficiency anemia that is affecting 60% of children in Africa (Osum *et al.*, 2013).

4.6 Molar ratios of Phytate to Ca, Zn and Fe

Mineral molar ratio is used to estimate relative mineral bioavailability. Cauliflower leaves showed different molar ratios after being boiled, blanched and fermented; these results are presented under table 13 below.

Table 13: Cauliflower leaf molar ratios of Phy to Ca, Zn, and Fe under different processing methods

No	Sample type	Phy/Ca	Phy/Zn	Phy/Fe
1	Raw	0.00024	0.0731	0.0132
2	Boiled	0.00014	0.0489	0.0083
3	Blanched	0.00004	0.0939	0.0065
4	Fermented	0.00006	0.0041	0.0009

According to Fordyce *et al.*, (1987) phytate/Fe above 1 has a negative effect, phytate/Zn higher than 15 has low estimated bioavailability and phytate/Ca above 0.17 can impair calcium bioavailability. So in this study we can see that all of the desirable molar ratio of phy/Fe, phy/Ca and phy/Zn were achieved.

In foods that contain high calcium content there is a chance that the calcium could exacerbate the negative effect of phytate on zinc absorption, a molar ratio of phytate \times calcium to zinc has been suggested as a better predictor of zinc absorption and a molar ratio higher than 200 may have a negative effect on zinc bioavailability (Fordyce *et al.*, 1987). As table 13 shows PhyCa/Zn ratio in raw, boiled and blanched exceeded this limit so we can conclude that cauliflower leaves being a high calcium containing plant affects the bioavailability of zinc but when treated under fermentation the leaves will have a high estimated zinc bioavailability.

4.7 Functional Properties

Cauliflower leaves powder showed different results on the water absorption capacity, wet gluten content and wet gluten to protein content when incorporated with wheat flour. These results are presented below in table 13.

Table 14: Functional properties of bread made with wheat flour and different proportions of cauliflower leaf powder.

Sample type	Water Absorption capacity (ml)	Wet Gluten content (%)	Wet Gluten/Protein
F1	2 \pm 0.70 ^a	13.60 \pm 0.03 ^a	1.26
F2	1.5 \pm 0.70 ^{ab}	13.05 \pm 0.12 ^{abc}	1.25
F3	1.5 \pm 0 ^{ac}	13.28 \pm 0.04 ^{ab}	1.28
F4	2.25 \pm 0.35 ^{ad}	12.68 \pm 0.45 ^{bc}	1.30
F5	1.75 \pm 0.35 ^a	12.49 \pm 0.43 ^c	1.38
F6	1.25 \pm 0.35 ^{ae}	12.85 \pm 0.27 ^{bc}	1.62

Note: all data are reported as dry weight basis

All values are the means of duplicate \pm SD

a, b, c, d, e are superscripts given to show the significant difference between means within column

F1-91:9 whole wheat flour: cauliflower leaf powder

F2-93:7 whole wheat flour: cauliflower leaf powder

F3- 95:5 whole wheat flour: cauliflower leaf powder

F4- 97:3 whole wheat flour: cauliflower leaf powder

F5- 99:1 whole wheat flour: cauliflower leaf powder

F6-100:0 whole wheat flour: cauliflower leaf powder

4.7.1 Water Absorption Capacity

Wheat breads made with different proportion of cauliflower leaves powder showed no significant difference in water absorption capacity with the bread made without the powder. But studies show that cauliflower leaves powder have a high potential for use in food products such as bakeries, beverages, gravies, soups and sauces due to its high water absorption capacity (Mythili *et al.*, 2021).

4.7.2 Gluten Content

The gluten content of wheat bread made with different proportion of Cauliflower leaf powder is presented in table 14. But the content was not significantly different from bread without the powder except in F1 (91:9 whole wheat flour: cauliflower leaf powder) that was with highest gluten (Table 14). The increment of gluten in F1 could be because of the increased protein content with the addition of the cauliflower leaf powder. According to Violeta *et al.*, (2010), wet gluten and protein ratio (WG/P) ranging between, 2.7 – 3.0 have optimal baking characteristics, while WG/P ratio closer to 2.3 could have strong gluten characteristics. But wheat breads made with different proportion of cauliflower leaf were having WG/P ratio of below 2.0 that shows the low gluten characteristics of the breads which will be a good preference for people that are interested in foods with low gluten content. High quality evidences supports the need for gluten avoidance to prevent disease other than those specifically known to cause immune-mediated response to gluten (Niland & Cash, 2018).

4.8 Sensory Evaluation

4.8.1 Bread



Figure 8: Panellists

Next to bread made with 100% wheat flour (F6), F5 and F4 were the second and third most preferred breads for their sensory quality (colour, odour, flavour and overall acceptability) (table 15). Although, overall acceptability of the bread samples decrease with increasing addition of cauliflower leaf powder, most of the bread products with cauliflower leaf were superior in crude protein, crude fat, crude fiber and total ash content compared to bread with 100% wheat flour (F6). However, in order to fulfil bread suitability in terms of nutrition and acceptability it will be reasonable to provide people bread with the combination of 99% wheat flour and 1% cauliflower leaf powder and 97% wheat flour and 3% cauliflower leaf powder.

Table 15: Sensory evaluation results of bread made with different proportion of Cauliflower leaf powder

Sample Type	Evaluation criteria			
	External colour	Odour	Flavour	Overall acceptability
F1	3.7	4	4.4	4.7
F2	4.5	4.7	5.4	5.1
F3	5.9	5.8	6	6.1
F4	7.1	7.2	6.9	7.7
F5	8	8.1	8.2	8.2
F6	8.7	8.4	8.1	8.6

Note: All values are the means of results of 10 panellists
F1-91:9 whole wheat flour: cauliflower leaf powder
F2-93:7 whole wheat flour: cauliflower leaf powder
F3- 95:5 whole wheat flour: cauliflower leaf powder
F4- 97:3 whole wheat flour: cauliflower leaf powder
F5- 99:1 whole wheat flour: cauliflower leaf powder
F6-100:0 whole wheat flour: cauliflower leaf powder



Figure 9: Bread. (A) Breads with different proportions of wheat flour and cauliflower leaf powder before being baked (F1-F6, from left to right), (B) Breads with different proportions of wheat flour and cauliflower leaf powder after baking (F1-F6, from left to right), (C) Small portion of each bread sample as presented for the panellists for sensory evaluation.

4.8.2 Meal like Cabbage

Table 16 shows the result of the sensory evaluation of a meal made with cauliflower leaf and Ethiopian Spinach which was the control. Almost similar average scores were given for both products for their colour, odour/aroma, flavour and overall acceptability. But the panellists commented that the external colour of cauliflower leaf meal was a bit darker when compare with the control group. Overall, the Cauliflower leaf can be made in to a meal like Cabbage and is acceptable for the consumers. Therefore, the product can be introduced to the wider consumer as an alternative means of using the cauliflower leaf.

This sensory evaluation was done in order to show another way of consuming the leaf and as table 16 shows cauliflower leaves were accepted at a high rate when cooked as a meal, it was preferred by the panellists as the meal that was made by using Ethiopian spinach (control group) except that most panellists comment.



Figure 10: Prepared meal (A) small portion of meal like cabbage made from cauliflower leaves as presented for the panellists, (B) small portion of meal like cabbage made from Ethiopian spinach as presented for the panellists

Table 16: Sensory evaluation results of a meal made with cauliflower leaf and Ethiopian Spinach

Sample Type	Evaluation criteria			
	External Colour	Odour/Aroma	Flavour	Overall Acceptability
Meal like cabbage made from Cauliflower leaves	8	8.6	8.4	8.2
Meal like cabbage made from Ethiopian spinach	8.8	8.8	9	8.8

Note: All values are average score of the result of sensory evaluation by the 10 panellists.

4.8.3 Kimchi

Another way of consuming cauliflower leaves could be its fermented form as kimchi. To determine the acceptability, a sensory evaluation was done on the kimchi made with the cauliflower leaf by making Kimchi made with Napa cabbage, which has a real kimchi test, as a control group. As presented under table 16, the panellists better liked the control group compared to kimchi that was made with the Cauliflower leaves. The panellists commented that the test in both samples was not familiar to them but what they most liked about both samples was the taste of the spices. However, there was no a wide gap in the average score of the kimchi that was made from cauliflower leaves and Napa Cabbage in all evaluation criteria but there is even a possibility to enhance the external colour, odour/aroma and flavour using different natural flavouring and colouring agents.

Table 17: Sensory evaluation results of Kimchi made with Cauliflower leaf and Napa Cabbage.

Sample Types	Evaluation criteria			
	External colour	Odour/Aroma	Flavour	Overall acceptability
Kimchi made from cauliflower leaves	8.2	5.5	6.1	6.9
Kimchi made from Napa cabbage	8.8	7.5	7.6	7.9

Note: All values are average score of the result of sensory evaluation by the 10 panellists.



Figure 11: Kimchi, (A) Kimchi made up of china cabbage, (B) Kimchi made up of cauliflower leaf powder, (C) small amount of both samples as presented for the panellists for sensory evaluation

5. Conclusion and Recommendation

Results of this study indicated that cauliflower leaf is a good source of fiber, protein, vitamin C, minerals such as Ca, Zn, Fe, total antioxidant and flavonoid and can be stated that the leaf is one of nutritious green leafy vegetables. Undertaking the leaf into different processing methods showed different results on the leaf but in terms of anti-nutritional factors, which could be a risk for consuming it, processing methods like boiling, blanching and fermentation reduced oxalate and phytate content of the leaf. The acceptability of the leaf was good specially when cooked as meal like cabbage and as bread with small amount of leaves' powder. Incorporating the leaf's powder with wheat flour showed a good impact to provide fiber rich bread, to increase acceptance of the bread with higher inclusion of cauliflower leaves powder further sensory analysis should be carried out by adding some flavours like vanilla and by also sweetening the bread using artificial sweeteners such as Aspartame might show a different result on the acceptability. Generally it can be conclude that consuming cauliflower leaf could be one way of reducing our low vegetable consumption habit, decrease nutritional deficiency in the country and contribute in reducing postharvest loss of the plant.

Based on the study it is recommended that

- Awareness within the society about the usefulness of green leafy vegetables should develop
- Further studies should be carried to investigate additional nutritional values of the raw and processed cauliflower leaf
- Effect of different processing methods other than boiling, blanching and fermentation should be explored
- Studies on the raw cauliflower leaf, if it also have other anti-nutritional factors should be observed
- Other ways of consuming cauliflower leaf and increasing the acceptability of the leaf should be further studied
- Microbial load of the fermented cauliflower leaf should be investigated

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Annex 1: Information sheet for sensory evaluation (English and Amharic)

Information sheet

Hello everyone, currently, I am conducting my MSc research on the title ‘ Nutritional and anti-nutritional factors of cauliflower leaf under different processing methods and its acceptability as bread nutrient enhancer’, the purpose of this study is to evaluate the acceptability of cauliflower leaf that undergo different processing methods by using it as an ingredient.

I. Procedure

Take about 30 minutes at each session. You are presented with, different cooked cauliflower leaf, kimchi made using cauliflower leaf and bread incorporated with cauliflower leaf powder, samples separately and asked to rank certain sensory characteristics of the samples. As a panellist, it is critical for you to complete your session. The research is conducted in food science and nutrition sensory laboratory.

II. Benefits/ Risks

Your participation in the project will provide information about sensory properties of cauliflower leaf. There are no identifiable risks associated with tasting this different cauliflower leaf samples.

III. Extent of Anonymity and Confidentiality

The results of your performance are strictly confidential. Individual panellists will not be referred to any publications or reviews.

IV. Compensation

There is no monetary compensation for participation in this study

V. Freedom of withdraw

If after becoming familiar with the sensory project you chose not to participate, you may withdraw. It is essential to the success of the sensory panel that you complete each session, however, there may be circumstances under which that you may chose not to complete your session and withdraw at any time.

Consent form for ‘Nutritional and Anti-nutritional factors of Cauliflower Leaf under Different Processing Methods and its Acceptability as Bread Nutrient Enhancer’

I have been informed about the objectives, risks and benefits of the study. I have also been informed about my rights not to participate in the study and withdraw any time without any consequences. I have been able to ask questions about the study and my questions have been answered to my satisfaction. I understand that taking part in the study involves sensory testing.

I understand the information collected will not be shared beyond the study. However, I agree that may be shared with other researchers for future research studies that may be similar to this study.

Based on the information provided above, I have agreed to participate in the study

Name of participant -----

Signature of the participant-----

Date-----

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Annex 3: Checklist for bread sensory evaluation

Hedonic Scale Instruction

You will receive a total of 6 different breads formed by different proportion of cauliflower leaf powder and wheat flour. Please indicate your opinion about the characteristics being tested (colour, odour, flavour, and overall acceptability).

You are asked to evaluate each sample in the order they laid out from left to right. Remember to cleanse your palates between samples with the bottled water provided.

Bread sample with different proportion of cauliflower leaf powder and wheat flour

Rating acceptance test using semi-trained panellists

Direction: please rate the acceptance of each bread sample provided with hedonic scale of 1- dislike extremely to 9- like extremely, as indicated at the score sheet below

a) Score sheet

Panellist code _____ Sample Code _____ Date _____

Sensory Attribute	Description	Panellist's Response								
		1	2	3	4	5	6	7	8	9
External colour										
Odour/ Aroma										
Flavour										
Overall acceptability										

Note: 1= Dislike Extremely, 2= Dislike Very Much, 3= Dislike Moderately, 4= Dislike Slightly, 5= neither Like nor Dislike, 6= Like Slightly, 7= Like Moderately, 8= Like Very Much, 9= Like Extremely

Comment: _____

Feel free to include comments to describe any noticeable attribute

b) Ranking

Please give ranks for each bread samples prepared by different proportion of cauliflower leaf powder and wheat flour based on the given sample code

Sample Code	Rank			
	External colour	Odour/ Aroma	Flavour	Overall acceptability

Annex 4: Checklist for meal like cabbage sensory evaluation

Hedonic Scale Instruction

You will receive cauliflower leaf cooked like cabbage. Please indicate your opinion about the characteristics being tested (colour, odour, flavour, and overall acceptability).

Remember to cleanse your palates between samples with the bottled water provided.

Cauliflower leaf cooked like cabbage

- a) Rating acceptance test using semi-trained panellists

Direction: please rate the acceptance of the sample provided with hedonic scale of 1- dislike extremely to 9- like extremely, as indicated at the score sheet below

- b) Score sheet

Panellist code _____ Sample Code _____ Date _____

Sensory Attribute	Description	Panellist's Response								
		1	2	3	4	5	6	7	8	9
External colour										
Odour/ Aroma										
Flavour										
Overall acceptability										

Note: 1= Dislike Extremely, 2= Dislike Very Much, 3= Dislike Moderately, 4= Dislike Slightly, 5= neither Like nor Dislike, 6= Like Slightly, 7= Like Moderately, 8= Like Very Much, 9= Like Extremely

Comment: _____

Feel free to include comments to describe any noticeable attribute

Annex 5: Checklist for kimchi sensory evaluation

Hedonic Scale Instruction

You will receive kimchi prepared by cauliflower leaf. Please indicate your opinion about the characteristics being tested (colour, odour, flavour, and overall acceptability).

Remember to cleanse your palates between samples with the bottled water provided.

kimchi prepared by cauliflower leaf

- a) Rating acceptance test using semi-trained panellists

Direction: please rate the acceptance of the sample provided with hedonic scale of 1- dislike extremely to 9- like extremely, as indicated at the score sheet below

- b) Score sheet

Panellist code _____ Sample Code _____ Date _____

Sensory Attribute	Description	Panellist's Response								
		1	2	3	4	5	6	7	8	9
External colour										
Odour/ Aroma										
Flavour										
Overall acceptability										

Note: 1= Dislike Extremely, 2= Dislike Very Much, 3= Dislike Moderately, 4= Dislike Slightly, 5= neither Like nor Dislike, 6= Like Slightly, 7= Like Moderately, 8= Like Very Much, 9= Like Extremely

Comment: _____


Feel free to include comments to describe any noticeable attribute

Annex 6: Ethical clearance

COLLEGE OF NATURAL & COMPUTATIONAL SCIENCES

Addis Ababa University

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Ref.No. CNCSDO/758/15/2023
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Date June 09, 2023
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To Whom It May Concern


The College of Natural and Computational Sciences Institutional Review Board (CNS-IRB) Committee in its meeting held on 25/05/2023, Minute No. IRB/05/2015/2023 has examined the project proposal entitled “Nutritional and Anti-nutritional factors of Cauliflower Leaf under Different Processing Methods and its Acceptability as Bread Nutrient Enhancer” by Aklesia Haileysus from the Addis Ababa University.

The proposal is approved for implementation for one year, effective June 09, 2023.

With regards,



Tileye Feyissa, (Prof)
Dean, College of Natural & Computational Sciences
Addis Ababa University



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Please Quote our reference number in your correspondence

“Examine all things; hold fast that which is good”

“ሁሉን መርምሩ መልካሙን ያከ”