BINDURA UNIVERSITY OF SCIENCE AND EDUCATION

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BACHELOR OF SCIENCE EDUCATION HONORS DEGREE IN CHEMISTRY

TOTAL PHENOLIC CONTENT OF *CURCUBITA MAXIMA* CONSUMED BY MANJOLO COMMUNITY IN BINGA DISTRICT

BY

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B1543383

A DISSERTATION SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS OF THE BACHELOR OF SCIENCE HONORS DEGREE IN CHEMISTRY EDUCATION

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DEDICATION

This effort honors God Almighty, my creator, tower of strength, source of inspiration, wisdom, knowledge and insight. He has been my source of strength throughout this program, and I have only been able to fly on His wings. I also dedicate this work to Prof. Dzomba, my supervisor, who has been supportive throughout the process. To my father Lackson Ncube, my sister Mai Mashy and my friend Maureen Moyo, I appreciate their support. My love for them has no limits.

ABSTRACT

The study was designed to determine total phenolic content of *Curcubita maxima*. *Curcubita maxima* are traditional vegetables which plays an important role in the dietary requirements as well as providing empirical evidence that helps to promote and preserve the cultural heritage of the Manjolo community. The traditional vegetables were subjected to Genesys 10S UV-Vis spectrophotometer at 760nm for the analysis of total phenolic content. Total flavonoid content was determined using aluminum chloride colorimetric assay. Total phenolic content for the samples was between 33 and 35 mg GAE/g. The exact values were 33.04 ± 0.13; 35.11 ± 0.11; 34.10 ± 0.04; 33.44 ± 0.41 and 35.03 ± 0.22 mg GAE/g extract for samples. The total content of flavonoids was 13.44 ± 0.20; 17.66 ± 0.51; 14.01 ± 0.69; 12.77 ± 0.91 and 16.79 ± 0.74 mg QE/g extract for samples. Presence of appreciable levels of phenolic compounds in *Curcubita maxima* shows its importance in the diet as a food that prevents oxidative stress diseases such as cancers and diabetics.

Table of Contents

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1: INTRODUCTION

1.0: Background to the study

There has been a paradigmatic shift in food habits in the last couple of decades as consumers are preferring food that is rich in micronutrients. Consequently, consumption of a variety of traditional vegetables has gained momentum (Benvenuti et al. 2016). Pumpkin leaves have been considered as a traditional vegetable in many countries in Africa and Zimbabwe is among them. The pumpkin plant is a member of the genus *Cucurbita* belonging to the *Cucurbitaceae* family. Around the globe, there are many varieties of the plant. Parts of the plant that are edible are leaves, flowers and the fruits. The plant has a high yield, easy to grow and it is very economical.

Of late the study of bioactive compounds of *Cucurbita maxima* has become popular among food science and technology researchers. Quite a number of studies has proved antioxidant potential, potency in averting such ailments as cardiovascular disease (atherosclerosis, stroke heart attack), cancer as well as neurological disorders like Parkinson's and Alzheimer's disease (Khanama et al, 2012; Kulczyński et al., 2020). Numerous foreign substances are introduced in the human body on a daily basis due to inability to fully metabolize them, for example generation of free radicals and reactive oxygen species. To maintain a healthy diet, one has to be wary of the content and composition of the food they take especially that of plant origin. As mentioned earlier on, majority of consumers are opting organic food of plant origin (Rakass et al., 2018; Kulczyński et al., 2020). Human health heavily depends on their daily antioxidant consumption because they minimize the danger posed by osteoporosis and diabetes among other diseases related to affluence (Indrianingsih et al., 2019).

Inarguably, many vegetables are endowed natural antioxidants that alleviate the undesirable effects of oxidative stress among them flavonoids and phenolic compounds. Pumpkin plant and the fruit possess these compounds and more. It is argued that bioactive compounds found in the plant confer a protective role against coronary heart disease and hypertension and this is particularly important in low-income countries where healthcare is beyond the rich of many (Hagos et al., 2022; Valenzuela et al., 2014; Borde et al., 2022) like in Zimbabwe. Accordingly, it is imperative to determine the content of bioactive compounds in *C. maxima* to provide insights into dietary guidance as well as the quality of the pumpkin leaves. The study evaluates the free radical scavenging activity, total phenolic and flavonoid content of pumpkin leaves consumed by the community in Manjolo, Zimbabwe.

1.1: Problem statement

Consumption of *C. maxima* by people of Manjolo is quite common and it has some cultural significance. The plant has been shown to possess bioactive compounds especially phenolics. Phenolic compounds have numerous health benefits. However, there is a nutritional gap pertaining to total phenolic content of *C.maxima* consumed by the residents of Manjolo community. This is a setback in understanding the possible benefits and nutritional value related to the traditional practices connected to diet. This study addresses the lack of knowledge with regards to total phenolic content of *C. maxima* native to Manjolo community. In a bid to uphold evidence-based dietary practices and protect cultural traditions, investigation and determination of total phenolic content of *C. maxima* cannot be overemphasized

This research is going to add on to existing knowledge about nutritional value of *C. maxima*. The information is useful in creating awareness and in encouraging the consumption of *C. maxima*. In addition, the research may act as a guide for the food industry, health practitioners and policy-

makers in the formulation of potent dietary supplements that are in line with the community's cultural practices and dietary needs.

1.2: Aim

To quantify total flavonoids and phenolic compounds in *C. maxima* leaves

1.3: Objectives

This study seeks to:

1. Extract phytochemicals from *C. maxima* leaves using acetone, ethanol and water
2. Determine total phenolic content in *C. maxima* acetone, ethanol and water extracts
3. Determine the total flavonoid content in *C. maxima* acetone, ethanol and water extracts

1.4: Research questions

1. Which phytochemicals are present in the acetone, ethanol and water extracts of *C. maxima*?
2. How much phenolic compounds and flavonoids are there in the extracts of *C. maxima*?

1.5: Significance of the study

This study is significant to a number of stakeholders. The study will provide an understanding of the nutritional content of the traditional vegetable. This enables the people of the community to make informed choices in relation to their diet. Being knowledgeable about health benefits of the vegetable guides individuals in planning a balanced and nutritious diet. Determination of total phenolic content in *C. maxima* would go a long way in averting chronic diseases within the community as people become aware of the benefits of the vegetable. Furthermore, the study is going to provide empirical evidence that helps promote and preserve the cultural heritage of the community. Above all the study will contribute to the existing body of knowledge pertaining to the phenolic content of *C. maxima* as well as in the fields of food science and nutrition.

1.6: Delimitations

This study is specific to Manjolo community in Binga, Zimbabwe. As such, generalization of the outcomes to other regions or communities would not be possible. The sample size is small due to time and logistical limitations. This further affects standardization and general applicability of the results to the vastness of the Manjolo community. This research also concedes to the fact that there are variations in types of *C. maxima* in Manjolo community.

1.7: Limitations

This investigation may fail to account for the possibility of external factors on total phenolic content of *C. maxima* such as environmental conditions, agricultural practices and post-harvesting care. Furthermore, the study is restricted in including potential variations in total phenolic content in relation to seasons. This is a cross-sectional study which is capturing data at a specific point in time that is *C. maxima* available for consumption in the community as at February 2024 so it is limited in revealing long-term trends in phenolic content of the plant.

1.8: Hypotheses

H_0: There is inconsequential disparity in the total phenolic content among the varieties of *C. maxima* eaten by residents of the Manjolo community.

H_1: There is consequential disparity in the total phenolic content among the varieties of *C. maxima* eaten by residents of the Manjolo community.

CHAPTER 2: LITERATURE REVIEW

2.0: Taxonomic classification

The gourd family of plant species known as *Cucurbitaceae* has renowned pharmacological and nutritional importance around the globe. There are several members of the family that originate from Africa. The family has 800 species and 130 genera whose leaves, seeds and fruits are applicable in many ayurvedic preparations. Figure 1 classifies the *Cucurbitaceae***.**

Figure 1: Classification of *Cucurbitaceae*

Pumpkins have different names in different countries. In Zimbabwe, they are called manhanga in Shona. Common English names include squashes, gourds and butternut. The leaves are called muboora (Zezuru), mutikiti (Ndau) or bhobola (IsiNdebele).

2.1: Production of pumpkins

The *Cucurbita* spp. is amongst the top ten vegetables in the world. Three domesticated species of *Cucurbita* are the most economically important. These are *C. maxima, C. pepo* and *C. moschata* and they are commercially produced on a large scale the world over (Hosen et al., 2021). According to Adhikari et al., (2017), pumpkin is an underutilized crop that possesses significant benefits horticulturally but lack of genetically improved seeds in comparison to maize and wheat has spurred its production.

2.2: Structural differences

The pumpkin plant is a creeper with a stem that can reach up to 10 m in length if the conditions are ideal. The plant reproduces both vegetative and sexually because it possesses stem, roots, tendrils and flowers. The stems are rough and angular. They are capable of producing roots at the

internodes which contain no food reserves. Therefore, *Cucurbita* spp. are short-lived. The fruits vary in terms of size, shape and color amongst the genus and species. They can be globose or oval with different color patterns as shown in figure 2 and 3.

Figure 2: Varieties of pumpkin fruits

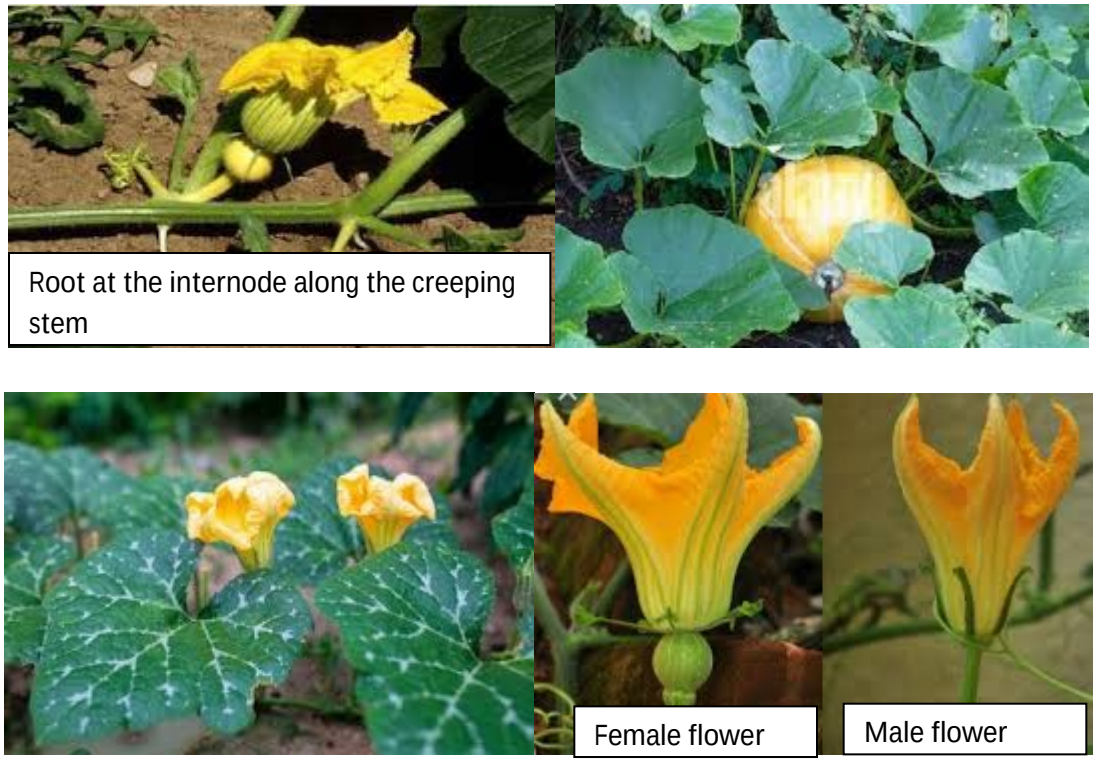


Figure 3: Creeping pumpkin plants and flowers

2.3: Nutritional and chemical composition

Several studies reported nutritional and chemical composition of *Cucurbita* spp. in different varieties of the same species in different parts. Hashash et al., (2017) reported 10.93 %, 14.51 % and 9.22 % carbohydrate content in seeds, fruit flesh and pulp of pumpkin respectively. The study went on to put forward the moisture, β-carotene, mineral, fat and protein content of pumpkin thereby confirming the role of pumpkins in human diet. The presence of β-carotene in pumpkin indicates potential antioxidant activity of the plant. In a separate study, *C. maxima* fruit flesh, peel and seeds extracts were shown to have antioxidant and anti-microbial compounds (Nashath et al., 2017). Amin et al., (2019) state that there is a significant difference in protein and carbohydrate content in seed samples of hybrid and indigenous varieties of *C. maxima* but there was negligible difference in moisture, fat, ash and fiber. Similar observations were obtained by Shahangir (2015).

In that study, lipid profile showed the presence of palmitic, linoleic and stearic acids in small amounts.

Ten different species of *Cucurbita* fruits were found to possess a significant difference in their total protein content, carbohydrate content, crude fiber content, β carotene and lycopene content. β carotene and lycopene content ranged from 0.72-2.48 mg/100g and 0.487– 1.988 mg/100g respectively (Zhou et al., 2017). According to Mohaammed et al., (2014), oxalates, saponins, alkaloids, cyanides, tannins and phytates are found in fruit flesh and pulp of *C. maxima*. Flavonoids and glycosides were found in the aqueous extract whereas phytosterols and terpenoids were found in the methanolic extracts of pumpkins (Gosh and Rana, 2021). The same study reports that there were saturated fatty acids such as lauric, capric, myristic acids and two monounsaturated fatty acids (palmitoleic and oleic) and one polyunsaturated fatty acid (linoleic acid) in pumpkin flowers.

According to Perez (2016), cucurbits contain carotenoids such as neoxanthin, lutein, α- and β-carotenes among many others. The seeds of the plants are also richer in gamma tocopherol than alpha tocopherol. Some studies have showed that mature and immature fruits of cucurbits have flavonoid content below detection limits but shoots and buds had positive results. The average total phenolic content of *C. maxima* as reported Mi et al., (2012) was above 45 mg GAE/100g. This brief review of nutritional and chemical composition of *C. maxima* shows a gap in phenolic content of pumpkin leaves and the phytochemical composition in general. Several factors influence the phenolic content of c. maxima. These include the genetic predisposition of the varieties of *C. maxima*, environmental conditions where the plants are grown (type of soil, seasonal differences and sunlight) and stage of maturity. Furthermore, pre harvesting and post-harvesting handling greatly impact on the total phenolic content of the cucurbits. For example, control of temperature and humidity after harvesting and conditions of storage affect the total phenolic content.

2.4: Potential benefits of consuming *Cucurbits*

2.4.1: The aspect of nutrition

A lot of phytochemical and nutritional analyses have put forward the constituents of various parts of pumpkin. There are studies that have reported high content of vitamins A, E and C, protein, phenolic compounds, fat, carbohydrate, crude fiber and β-carotene in pumpkins. B-carotene confers the characteristic yellow-orange color in pumpkins. The compound is a precursor to vitamin A hence the fruits are rich sources of vitamin A. Other studies have reported pumpkins as important sources of sodium, potassium, magnesium and iron (Hussain et al., 2017; Gosh and Rana, 2021; Zhou et al., 2017).

2.4.2: The aspect of nutraceuticals

The most economically and culinary significant genera of the vegetable crop are *Cucumis* (cucumber, melon), *Citrullus* (watermelons) and *Cucurbita* (pumpkins and squash). These plants have remained very essential in food and traditional medicine since time immemorial in the history of the African people. *C. maxima* seeds are either roasted, salted and served as a delicacy by the Ndebele and Shona people of Zimbabwe or they are coarsely grounded and molded into balls that are cooked alongside the leaves of the plant and served as relish. Also, flowers of *C. maxima* are mixed with the leaves and consumed as vegetable in Mexico, India and in Zimbabwe as well. *Cucurbits* have found a wide application in numerous folklore medical practices in management of gastro-intestinal parasites and diseases (Salehi et al., 2019). According to Hussain et al., (2017), pumpkin seeds have anthelmintic properties. Pumpkins are said to be anti-cancer, anti-diabetic, hypolipidemic anti-inflammatory. In addition, pumpkins are effective in maintaining the balance of hormones in women who are in the post-menopause phase (Patel, 2013). These reports further

portray extensive research done on pumpkins leaving a gap on investigations to do with the leaves of the pumpkin plant.

2.4.3: Value-addition of *C. maxima*

There are reports of development of value-added products entailing food additives and dietary supplements. Pumpkin flour mixed with rice flour was used to make breakfast functional food (Malkanthi and Hiremath, 2020). Addition of pumpkin flour increases the nutritional value of the functional bread prepared using that flour. In the same vein, Mishra et al., (2019) developed a bread recipe using pumpkin seed flour and similarly the proximate analysis of the product showed that it had elevated nutritional value, a semblance of the pumpkin seeds. In a separate study, Kumari et al., (2020) incorporated germinating pumpkin seed flour in making biscuits but nutritional content of the biscuits was lower because the seed flour reduced the amount of wheat flour used. Similar results were obtained by Khan et al., (2019).

However, incorporation of pumpkin in cakes resulted in higher nutritional value of the supplemented cake (Jesmin et al., 2016). Some studies have reported the production of packaged pumpkin flowers as dietary supplements, but the quality of the packaged flowers deteriorated in five days of packaging (Toro-Velez et al., 2022). Research went on to develop snacks using pumpkin flour, salt, onion powder, vegetable oil, water, shortening and spices. Pumpkin pectin was also isolated, and it exhibited solubility like commercial pectin. Pectin is an effective gelling agent and thickener in the production of jelly or jam (Arachchige et al., 2019; Sanadarani and Prasadi, 2018). Value addition mainly focused on the fruit and seed in foreign countries while neglecting the leaves of the plant hence the current study focuses on the total phenolic content of *C. maxima* leaves in a Zimbabwean context with specific reference to Manjolo community in Binga.

2.5: Phenolic compounds and names

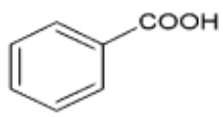
According to Manach et al., (2004), phenolic compounds consists of aromatic rings bearing one or more hydroxyl group, which is either free or involved in ester or ether bonds. Phenolic compounds exist primarily in a conjugated form with one or more sugar residues linked to hydroxyl groups by glycoside bonds. These compounds are also associated with other compounds such as amines, carboxylic acids and lipids (Bravo, 1988). Manach et al., (2004), reiterates that phenolic compounds are divided into different classes which include phenolic acids, flavonoids, stilbenes, tannins and lignans. Cuha et al., (2012) adds that flavonoids consists of subclasses that are based on the oxidation state of the central heterocycle which include flavones, flavonols, flavonones, anthocyanidins and isoflavones.

2.6: Structures of phenolic compounds

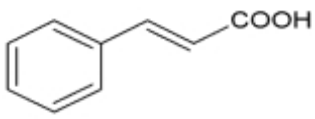
2.6.1: Structure of phenolic acids

Phenolic acids consist of hdroxybenzoic acid with a carboxylic acid functional group which is directly bonded to the phenol ring and hydroxycinnamic acid with a carboxylic acid functional group and a phenol ring are separated by two doubly double bonded carbons (Habauzit and Harcajada, 2008). This is shown below,

Hydroxybenzoic acid



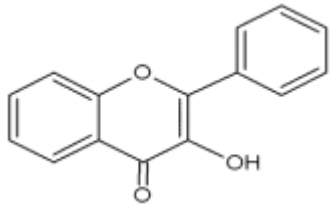
Hydroxycinnamic acid



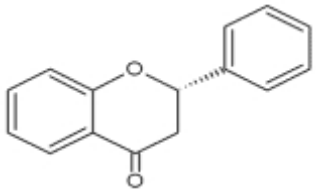
2.6.2: Structure of flavonoids

Flavonoids are further classified into flavones, flavonols, flavonones, anthocyanidins and isoflavones (Greenberg et al., 2008) The structures of these compounds are shown below;

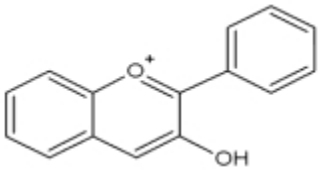
Flavonols



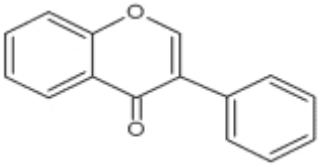
Flavonones



Anthocyanidins



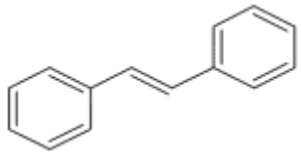
Isoflavones



2.6.3: Structure of stilbenes

According to Amic et al., (2003), stilbenes consist of phenolic compounds with two phenol units linked by doubly bonded carbons. There are several examples of stilbenes which are resveratrol, pterostilbene and piceatonnol. The structure of stilbenes is shown below,

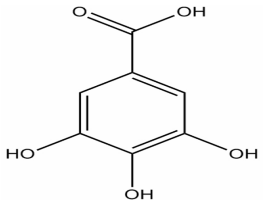
Stilbenes



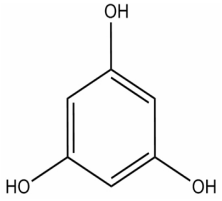
2.6.4: Structure of tannins

Tannins are structured to bind and precipitate proteins and amino acids. They are further classified into hydrolysable tannins and complex tannins (Okuda et al., 1989). The structure of hydrolysable tannins and complex tannins are shown below,

Hydrolysable tannins



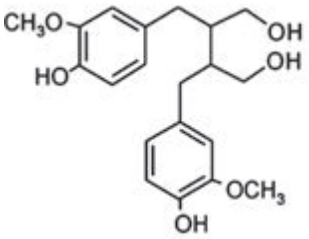
Complex tannins



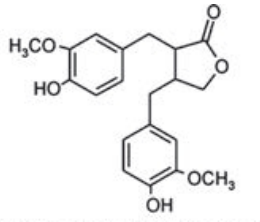
2.6.5: Structure of Lignans

Lignans consists of two phenol units linked by four carbons. The examples of lignans include matairesinol and secoisolariciresinol (Saleem et al., 2005). The structure of lignans is shown below,

Secoisolariciresinol lignans



Matairesinol lignans



2.7: Importance of phenolic compounds to diet

Phenolic compounds play an integral part in the human diet since they are present in all plant organs. According to Cicerale et al., (2012), high intakes of fruits, vegetables, whole grains, tea lowers the risks of chronic diseases such as cancer cardiovascular diseases, chronic inflammation as well as degenerative diseases. For example, apples contain phenolics such as flava-3-ols, phenolic acids, dihydrochalcnes and flavonols. Mango fruits also contain several bioactive compounds such as vitamins, cateronoids, terpenoids and phenolic compounds. Phenolic compounds are one of phytochemicals present in cereals which play an essential role to health. According to Manach et al., (2004), phenolic acids such as coumari, ferulic, gallic, hydroxybenzoic, vanillic, syringic and sinapic are found in the whole grain cereals like wheat, barley, oat, rice and corn.

CHAPTER 3: METHODOLOGY

3.0: Reagents and chemicals

Sulphuric acid, Folin’s reagent, gallic acid, sodium nitrite, sodium carbonate, aluminum chloride, quercetin, sodium hydroxide.

All chemicals that were used in this research were analytical grade.

3.1: Equipment

Analytical weighing balance, centrifuge, micropipettes, UV-Visible spectrophotometer Genesys 10S, glassware.

3.2: Collection of samples

Pumpkin leaves were collected from five different fields around Manjolo community, Binga district, Zimbabwe in the February to March period of year 2024. Mature full-grown soft edible leaves were collected into khaki envelopes with labels. The samples were separately cleaned in tap water, rinsed thrice in distilled water, chopped and left to dry at room temperature on a working table. The dry samples were separately crushed into fine powders that were stored in labelled airtight plastic containers under refrigeration.

3.3: Preparation of the extracts

10g of each sample were separately weighed and placed in 100 ml of 80% hydroethanolic solution. The samples were left on an orbital shaker for 24 hours at medium speed. The extracts were filtered to remove plant residue; filtrates were concentrated on a rotor vapor and dried at 40 ℃ in an oven. The extract powders were weighed and kept in a refrigerator until time of use. The percent yield was calculated as follows:

$$\%\ yield = \frac{mass\ of\ dry\ extract}{mass\ of\ plant\ sample\ measured\ for\ extrction} \times 100$$

3.4: Qualitative phytochemistry of *C.Maxima* extracts

Phytochemical constituents of *C. maxima* were tested as summarized in table 1.

Table 1: Preliminary phytochemical screening of samples

Phytochemical	Test procedure	Positive result
Alkaloids	2 ml extract + 2 drops Mayer's reagent along the sides of the test tube	Creamy or yellow precipitate
Flavonoids	2 ml extract + 10% Ferric chloride solution	Green precipitate
Phenolic compounds	1 ml extract + 4 drops dilute iodine solution	Transient red color
Tannins	0.5 g plant extract + 10 ml bromine water	Bromine is decolorized
Phytosterols	3 ml extract + a pinch of Sulphur	Sulphur sinks to the bottom
Terpenoids	5 ml plant extract + 2 ml chloroform + 3 drops sulphuric acid boiled on water bath	Grey colored solution
Saponins	2 ml extract + 5 ml distilled water, shake vigorously	Stable foam

3.5: Quantitative phytochemical analysis of *C. maxima*

3.5.1: Total phenolic content

The Folin method using gallic acid as a standard was applied. 1 ml of sample solution was mixed with a 10-fold dilution of a mixture of 7.5 % sodium carbonate and Folin-Ciocalteu reagent. The samples were dark-incubated for half an hour after which the absorbance were measured using a Genesys 10S UV-Vis spectrophotometer at 760 nm. The total phenolic content was expressed as milligrams of gallic acid equivalents per gram of extract (mg GAE/g extract).

3.5.2: Total flavonoid content

Total flavonoid content of samples was determined using the aluminum chloride colorimetric assay. The standard used was quercetin. 0.2 ml of 5% sodium nitrate solution were diluted with 3 ml of distilled water and mixed with 1 ml of extract. After 5 minutes, 0.4 ml of 10% aluminum chloride were added. The mixtures were allowed to stand for 5 minutes, and 1,5 ml of 1 M sodium hydroxide were added followed by 1 ml distilled water. The mixtures were centrifuged at 3500 rpm for 5 minutes. The supernatants were collected and their absorbance measured at 415 nm against an 80 % hydroethanolic blank containing everything else except the plant extract. The total flavonoid content was expressed as milligrams of quercetin equivalents per gram of extract (mg QE/g extract).

CHAPTER 4: RESULTS

4.0: Extraction yield

Table 2 is a record of the dry extract yields obtained for each sample. The yields were slightly different across all the samples. Graphical presentation of yields is given in figure 4.

Table 2: Yield of extracts obtained for each sample.

Sample	Field m	Field n	Field o	Field p	Field q
% yield	11.35	11.91	11.81	11.19	11. 54

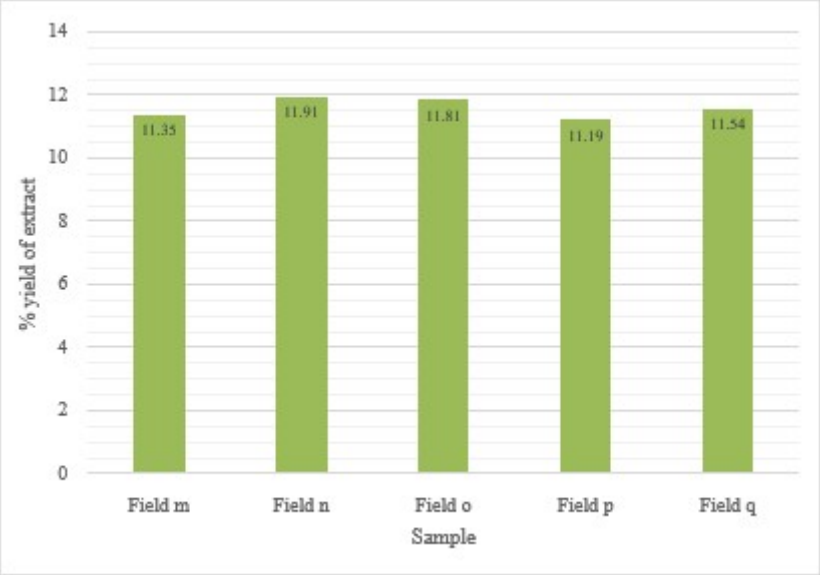


Figure 4:Comparison of percent yield of extracts

4.1: Preliminary phytochemical screening

Extracts of the various samples of *C.maxima* were screened for their phytochemical composition as described in section 3.4. Table 3 summarises the results that were obtained. Five samples were screened for phytochemicals and all of them tested positive in all the tests that were conducted.

Table 3: Preliminary phytochemical analysis of samples of *C. maxima*

Phytochemicals	Test used	Result				
		Field m	Field n	Field o	Field p	Field q
Alkaloids	Mayer's test	+	+	+	+	+
Flavonoids	Ferric chloride test	+	+	+	+	+
Phenolic compounds	Iodine test	+	+	+	+	+
Tannins	Bromine test	+	+	+	+	+
Phytosterols	Sulphur test	+	+	+	+	+

Terpenoids	Salkowski test	+	+	+	+	+
Saponins	Foam test	+	+	+	+	+

Key: + means present; - means not present

4.2: Total phenolic and flavonoid content

Phenolic compounds and flavonoids were quantified. The numerical values of the results of the quantitative phytochemical analysis are shown in table 4. Figure 5 and 6 are calibration curves for quantification of phenolic and flavonoid compounds respectively. Figure 7 is a graphical presentation of quantitative phytochemical analysis of *C. maxima* obtained from different fields in Manjolo.

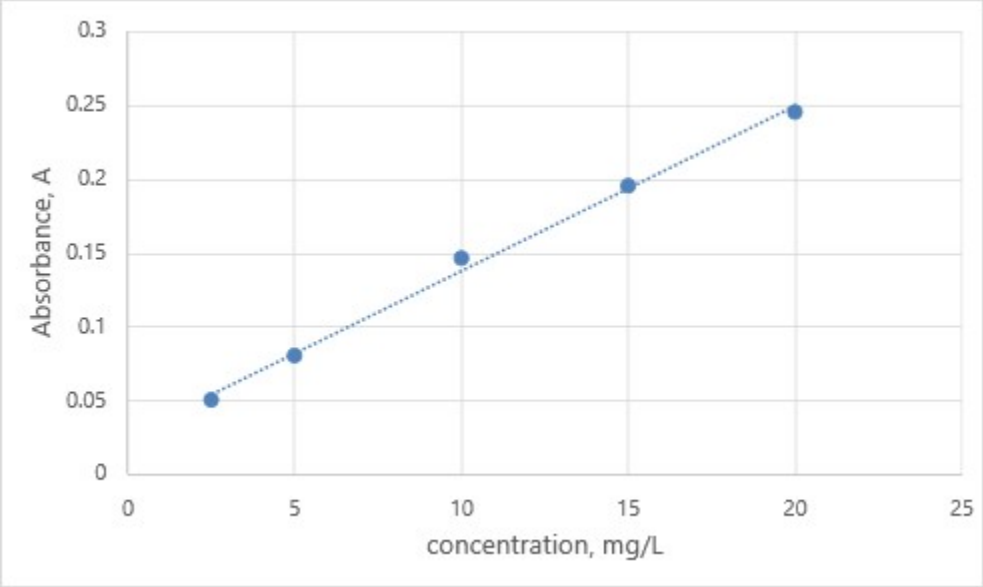


Figure 5: Calibration curve for quantification of phenolic compounds

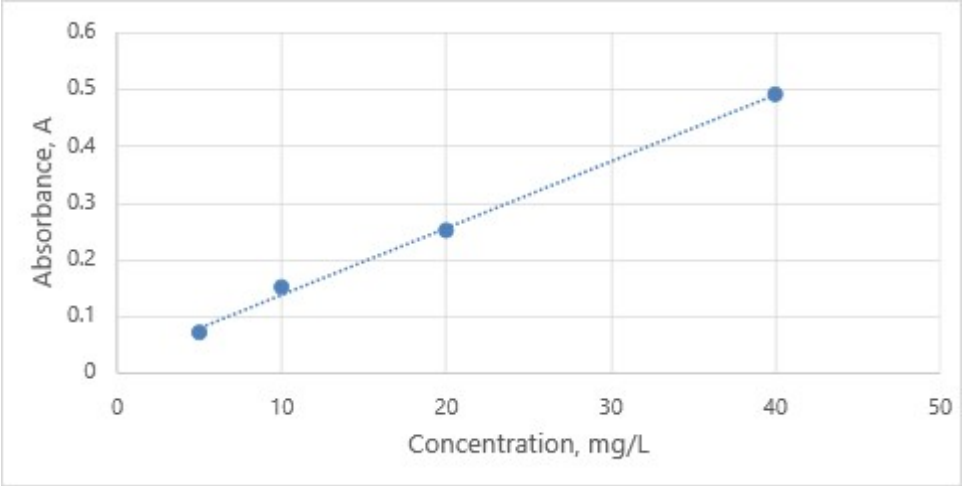


Figure 6: Calibration curve for quantification of flavonoids

Table 4: Total flavonoid and phenolic content for the samples collected from field m, n, o, p and q.

Sample	Total phenolic content (mg GAE/g extract)	Total flavonoid content (mg QE/g extract)
Field m	33.04 ± 0.13	13.44 ± 0.20
Field n	35.11 ± 0.11	17.66 ± 0.51
Field o	34.10 ± 0,04	14.01 ± 0.69
Field p	33.44 ± 0.41	12.77 ± 0.91
Field q	35.03 ± 0.22	16.79 ± 0.74

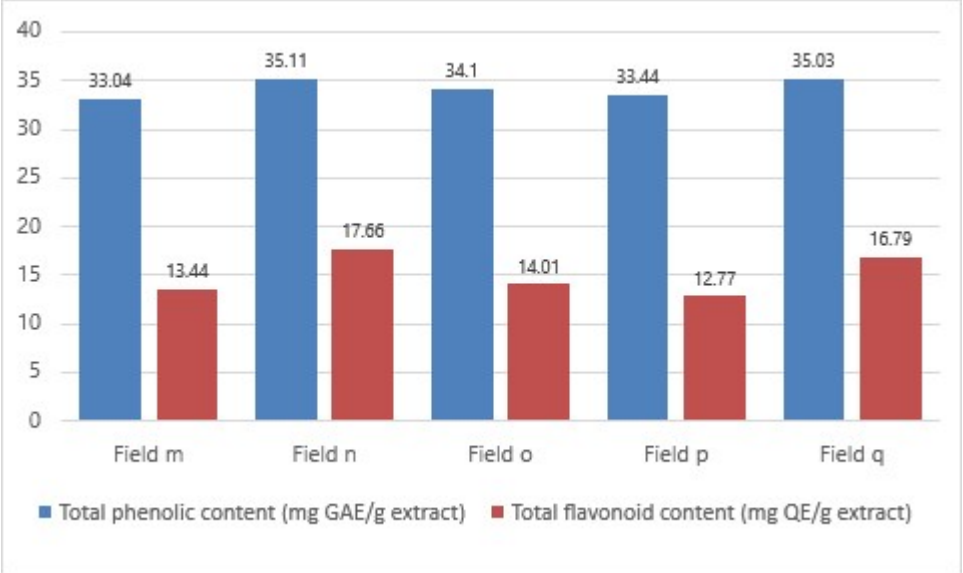


Figure 7: Quantifiable phenolic and flavonoid compounds in *C. maxima*

CHAPTER 5: DISCUSSION, CONCLUSION AND RECOMMENDATION

5.1: Discussion

This work evaluated phytochemical constituents and total phenolic and flavonoid content of 80 % ethanol extracts of five samples of *C. maxima* leaves collected from Manjolo community was evaluated using gallic acid and quercetin standards respectively. Phytochemical screening of the samples revealed the presence of alkaloids, saponins, terpenoids, flavonoids, phenolics, tannins and phytosterols (table 3). From human physiology point of view, phenolic compounds are paramount in anchoring the body's defense systems because they have anti-proliferative, antioxidant and anti-inflammatory properties. In addition, the other phytochemicals are known to equally important to humankind in the same sense as phenolics.

Total phenolic content was evaluated using gallic acid as a standard. The curve parameters (shown in figure 5) were $R^2 = 0.9958$ and the equation was $y = 0.112x + 0.0258$. According to the results obtained by this study, total phenolic content for the samples was between 33 and 35 mg GAE/g. The exact values were 33.04 ± 0.13; 35.11 ± 0.11; 34.10 ± 0.04; 33.44 ± 0.41 and 35.03 ± 0.22 mg GAE/g extract for samples m to q in that respective order (table 4). Quantification of flavonoids was done using quercetin as the standard. Parameters of the standard calibration curve were as follows: the equation was y = 0.0118 +0.0196 and the correlation coefficient was 0.9974 (figure 4). The total content was 13.44 ± 0.20; 17.66 ± 0.51; 14.01 ± 0.69; 12.77 ± 0.91 and 16.79 ± 0.74 mg QE/g extract for samples m to q respectively (table 4). The difference in the values obtained in this study were not statistically significant at 5 % confidence interval.

According to Jarungjitaree and Naradisorn (2019) significant differences in total phenolic content were observed in different methanolic extracts of *C.maxima*. The values obtained in that study were different from those of the current study. They were between 57 and 92 mg GAE/ 100 g extract. In a separate study, Bochnak and Swieca (2020) investigated the total phenolic content of dried pumpkin. Very low values ranging from 4- 11 mg GAE/g extract were obtained. Wanna (2019) used the Folin method to quantify phenolics in pumpkin peel and flesh and discovered that the total phenolic content in peel was 110.45 mg GAE/g while in flesh it was 116.66 mg GAE/g.

Singh et al. (2016) investigated total flavonoid content of *C. maxima* fruits and obtained values between 13.81 and 14. 62 mg QCE/100 g in 70%. In the same study, pumpkin pulp 6.90 mg QCE/100 g in 70 % methanol while the same concentration of ethanol yielded 7.73 mg QCE/100 g from pumpkin peel. According to Asif et al. (2017), the total flavonoid content in methanol extracts (65, 80, and 99.9%) of puree and peel of pumpkin were between 0.51 and 0.72 mg CE/100 g and 0.23 and 0.41 mg CE/100 g in that respective order. There was very little agreement between

literature values and values obtained in the current study mainly because the solvents that were used in extraction were different. In addition, the parts that were investigated varied from pulp, peel and puree while in the current study leaf extracts were studied. However, all the scholars are in agreement that generally *C. maxima* contains quantifiable phenolics and flavonoids regardless of the part of plant or fruit investigated.

5.2: Conclusion

The current study involved phytochemical screening quantification of flavonoids and phenolics extracted from leaves of *C.maxima* consumed by Manjolo community. Pumpkin leaves contain appreciable amounts of phenolics and flavonoids. The results indicate that the total phenolic and flavonoid content of *C. maxima* consumed by Manjolo community is partially different from each other. The data obtained in the current study is a revelation that *C. maxima* leaves are an excellent source of phenolics and the study has provided an impetus for full utilization of this valuable plant which is underutilized in Zimbabwe. The study also serves a precursor to advanced research on value addition of the plant as well as formulation of nutraceuticals.

5.3: Recommendations

The fact that *C.maxima* leaves possess biologically important phytochemicals makes the plant an accurate, adaptable and convenient source of ingredients for industries such as food, medicine and nutraceutical. Extraction and isolation of the plant's phytochemicals for development of functional foods and food products is recommended as these may help in preventing age related disorders as well as a number of medical conditions. Extensive research in value addition of *C. maxima* is recommended; for example, commercializing dried leaves, making flour and fortified foods.

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