Comparative Analysis of Ethanol and Methanol Extraction of Flavonoids and Phenolics from different variety of *Moringa Oleifera* Leaves

Abstract

The Moringa tree is recognized for its high nutritional and economic value, boasting leaves rich in minerals, vitamins, and protein. Despite its popularity, the differentiation of phenolic and flavonoid content among various Moringa oleifera varieties remains underexplored, particularly when extracted using different solvents. This study aims to compare the efficiency of ethanol and methanol in extracting phenolics and flavonoids from Moringa oleifera leaves, providing insights into how environmental factors and solvent types influence the phytochemical composition of these leaves. The study's findings could inform the optimal extraction methods for maximizing the nutritional and medicinal benefits of Moringa oleifera. Fresh leaves of five Moringa oleifera varieties were collected from the Winfred Thomas Agricultural Research Station, Alabama A&M University. The leaves were dried, ground, and stored at room temperature. The extraction process involved dissolving the leaves in 70% ethanol and 80% methanol, followed by stirring, filtration, and freeze-drying of the samples. The total phenolic and flavonoid content was then analyzed using high-performance liquid chromatography (HPLC). Statistical analysis involved box plots, scatter plots, bar plots, and t-tests to assess the efficiency of the two solvents in extracting phenolic and flavonoid compounds and the characterization of the total phenolics and flavonoids across the varieties. The descriptive analysis revealed that the Nigerian variety had the highest phenolic content of approximately 822.3 µg/ml when extracted with methanol, compared to 806.3 µg/ml with ethanol. In contrast, the Indian variety showed a significant difference in favor of methanol, with phenolic content at 814.3 µg/ml compared to 647.3 µg/ml in ethanol. For flavonoids, the Nigerian variety showed higher extraction efficiency with ethanol, at 1253.12 µg/ml, compared to 1083.52 µg/ml with methanol. The t-test results indicated that the mean phenolic content extracted using methanol ($M = 798.60$, $SD = 23.567$) was slightly higher than that with ethanol ($M = 790.60$, $SD = 45.183$), though this difference was not statistically significant ($t(28) = 0.608$, $p = 0.382$). Similarly, the mean flavonoid content extracted using methanol ($M = 1068.40$, $SD = 128.641$) was lower than that with ethanol ($M = 1129.40$, $SD = 166.018$), but this difference was also not statistically significant ($t(28) = 1.125$, $p = 0.334$). The scatter plot revealed a strong positive

correlation (Pearson correlation coefficient = 0.95) between phenolic and flavonoid content, indicating a significant linear relationship. This study concludes that both methanol and ethanol are effective solvents for extracting phenolic and flavonoid compounds from Moringa oleifera leaves, with minor variations depending on the variety. Further research should explore the implications of these findings for optimizing extraction processes in industrial applications.

Introduction

The Moringa tree is gaining widespread popularity due to its high nutritional and economic value (Patil et al, 2022; Boopathi & Raveendran, 2021). Its leaves are packed with minerals, vitamins, and protein, making them a stand-out among vegetables (Covington, 2021; Fernández-López *et al*, 2020). Not only are the leaves nutritious, but they also have medicinal properties and can be consumed in various forms such as fresh, dried, in capsules, as tea, or in instant soup. They are also used as green fertilizers and as a supplement for chickens, goats, and cows (Peñalver *et al*, 2022; Islam *et al*, 2021). Moringa is non-toxic and has anti-carcinogenic properties (Abdelkarim & Mohammed, 2015), and it is known as the Miracle Tree due to its worldwide cultivation and high nutritional properties (Elhawary *et al*, 2024). The leaves contain high levels of total anti-oxidative polyphenols, which can reduce the risk of diseases in animals and humans (Hassan et al, 2021). The phenolic and flavonoids content can vary depending on the variety (Ralepele *et al*, 2021; Trigo *et al*, 2020; Zhu *et al*, 2020) and region of cultivation, with regions experiencing drought stress potentially having higher levels (Zhu *et al*., 2020). Additionally, factors such as cultivation area, precipitation, and latitude can greatly affect the accumulation of total flavonoids and phenolic acids in Moringa leaves (Singh *et al*, 2024; Mihai *et al*, 2022; Ralepele *et al*, 2021). The study was designed to investigate the variation of phenolic and flavonoid content in Moringa oleifera leaves from five different countries. This research can provide valuable insights into how environmental factors impact the phytochemical composition of Moringa leaves. While there have been reports of various phytochemicals and bioactive compounds in Moringa leaves, there is little information available on the variety differentiation of phenolic and flavonoid content using high-performance liquid chromatography.

2. Materials and methods

2.1 *Moringa oleifera* leaves

Fresh leaves of five varieties of Moringa were retrieved from the Winfred Thomas Agricultural Research Station (WTARS) at Alabama A&M University. The fresh leaves were allowed to dry at room temperature, ground using a laboratory Micro-Mill (Bel-Art Products, Pequannock, NJ 07440 USA) and kept in sealed air-tight Ziploc bags at room temperature until further analyses.

2.2 Chemicals

Gallic acid, Catechin, Folin & Ciocalteu's phenol reagent, Methanol, Trolox, ABTS salt, Aluminium Chloride, Sodium Hydroxide, Sodium Nitrite, Sodium Carbonate, Acetic acid, Ethanol, Potassium Persulfate, Hydrochloric acid, TPTZ (tripyridyl-S-triazine), DPPH (2,2-diphenyl-1-picrylhydrazyl), Iron Chloride were purchased.

2.3 Sample extraction

For the preparation of extracts, Moringa leaves were dissolved in methanol and ethanol. The mixture was stirred using a magnetic stir bar and VMR Standard Multi-Position Stirrer for 3 hours at room temperature. Each sample was filtered using Whatman filter paper No.4 and the filtrate was evaporated to dryness under reduced pressure using Buchi Rotavapor at 50ºC. The samples were dissolved with deionized water and kept in the -80ºC freezer overnight. The frozen samples were kept in the freezer dryer for 48 hours. The freeze-dried samples were kept at room temperature for further analysis.

2.4 Statistical Analysis

This research work utilized box plots, scatter plot and bar plot to visualize and summarize the distribution of phenolic and flavonoid content in the extracts. T-test was used to determine if there was a significant difference in the extraction of phenolic and flavonoid compounds using 70% ethanol and 80% methanol.

3. Results

3.1 Patterns and trends in phenolic content concentrations

The fig 1 shows the bar chat visualization distribution of total phenolic content in Moringa oleifera leaves across five different varieties (Nigeria, Ghana, Haiti, India, and Tusk). The findings revealed that, when extracted with 80% methanol, the Nigerian variety of Moringa leaves contains approximately 792.2 µg/ml of phenolic content, while in 70% ethanol, it is around 753.3 µg/ml.

The distribution of phenolic content in the two solvents shows a moderate spread, with the methanol solvent resulting in slightly higher phenolic content compared to ethanol (Feng *et al*., 2022; Dabetić *et al*, 2020). In the Ghanaian variation, the amount of phenolic content in methanol is notably higher at about 822.3 µg/ml, while the ethanol solvent demonstrates a content of roughly 806.3 µg/ml. These figures reveal a minor contrast between the solvents, with methanol still managing to extract a slightly higher amount of phenolics (Molole *et al*., 2022; Ezez & Tefera, 2021; Phuyal *et al*, 2020). The Haitian type has a phenolic content of approximately 812.3 µg/ml in methanol and 791.3 µg/ml in ethanol, with only a slight variation between the solvents. Methanol has a slightly higher phenolic content (Ezez & Tefera, 2021). In contrast, the Indian type has a phenolic content of about 814.3 µg/ml in methanol and 647.3 µg/ml in ethanol, with a noticeable difference between the solvents. Methanol shows significantly higher phenolic content compared to ethanol (Morales-Olán *et al*, 2020). The tusk variety exhibits phenolic content levels of approximately 763.0 µg/ml in methanol and 796.3 µg/ml in ethanol. It is notable that, in this instance, ethanol displays marginally higher phenolic content than methanol, which deviates from the norm observed in other varieties (Shopska *et al*, 2021; Hong *et al*, 2020).

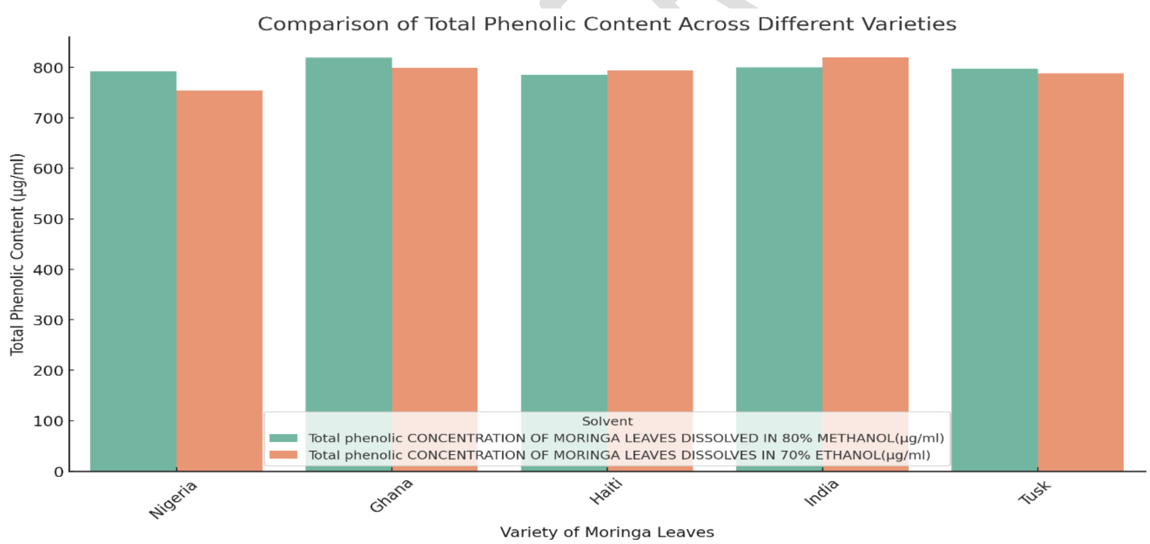


Fig 1: Bar chart showing phenolic distribution with the two solvents

In fig 2, the bar plot shows the extraction efficiency of methanol compared to ethanol for each variety of Moringa oleifera leaves. The extraction efficiency is calculated as the ratio of phenolic content extracted by methanol to that extracted by ethanol. The Nigerian strain of moringa leaves demonstrates a slightly higher extraction efficiency with methanol compared to ethanol (Hikmawanti *et al*, 2021; Tzanova *et al*., 2020), as shown by a ratio slightly above 1. Conversely, the Ghana variety exhibits nearly equal efficiency for both solvents, with a slight edge for methanol (Boateng & Clark, 2024; Owusu, 2022), indicated by a ratio close to 1. In comparison, the Haitian variety also shows a slightly higher efficiency with methanol, as evidenced by a ratio slightly above 1 (Segovia-Hernández *et al*, 2022; Tambawala *et al*, 2022). The Indian strain stands out with significantly higher extraction efficiency with methanol, as indicated by a ratio well above 1 (Sudha & Aranganathan, 2023; Borah *et al*., 2022). Finally, the Tusk variety has a ratio slightly below 1, suggesting that ethanol is actually more efficient than methanol for extracting phenolic compounds in this case (Mini *et al*, 2023; Acharjee, 2021).

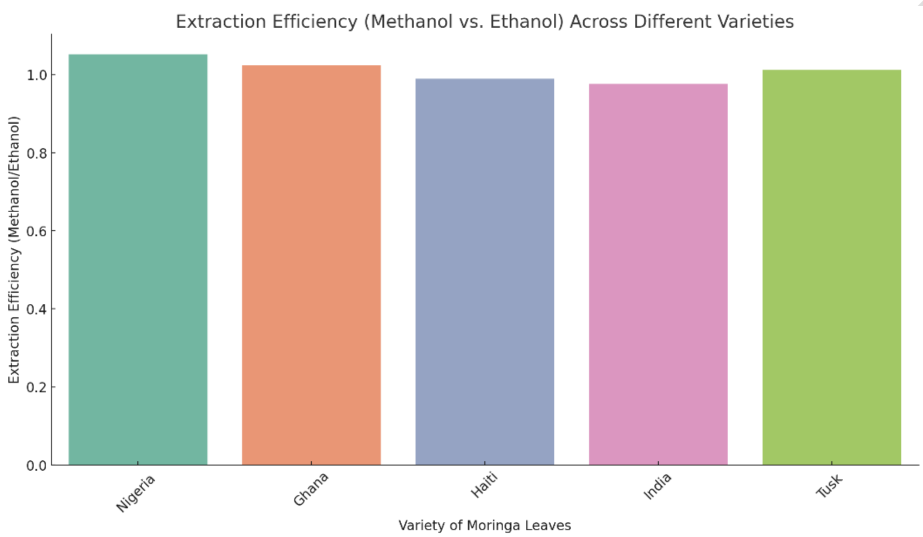


Fig 2: Bar chart showing the extraction efficiency between Methanol and Ethanol

In figure 3, the box plot illustrates the total phenolic concentrations of Moringa Oleifera leaves extracted using two different solvents: 70% ethanol and 80% methanol. In figure 3a, the concentrations are depicted in two distinct colors: red for 70% ethanol and green for 80% methanol. This color differentiation helps in rapidly distinguishing between the two solvents used for extraction. For 70% ethanol, the median phenolic concentration is approximately 775 mg/g, as indicated by the horizontal line within the red box. The box ranges from about 725 mg/g to 800 mg/g, which represents the interquartile range (IQR) and covers the middle 50% of the data. An outlier around 650 mg/g suggests that one variety had a notably lower phenolic concentration when extracted with 70% ethanol. The median for 80% methanol is slightly higher, close to 800 mg/g. The IQR is narrower than that of the ethanol extract, spanning from about 790 mg/g to 815 mg/g, indicating less variability in the phenolic concentration among the varieties when extracted with methanol (Lohvina *et al*., 2021). The whiskers of the red box (70% ethanol) extend further than those of the green box (80% methanol), indicating a wider range of phenolic concentrations in the ethanol extracts compared to the methanol extracts (Hong *et al*, 2020). This suggests that the phenolic content extracted with 70% ethanol might be more influenced by the variety of Moringa Oleifera leaves. The slightly higher median concentration and lower variability in the methanol extracts suggest that 80% methanol might be a more efficient and consistent solvent for extracting phenolic compounds from Moringa Oleifera leaves compared to 70% ethanol (Alara *et al*., 2021; Osorio-Tobón, 2020; Radzali *et al*., 2020). The wider range and the presence of an outlier in the ethanol extract indicate that different varieties of Moringa Oleifera may respond differently to ethanol extraction (Savić *et al*.2024; Maina *et al*.2021; Tovar, 2021).

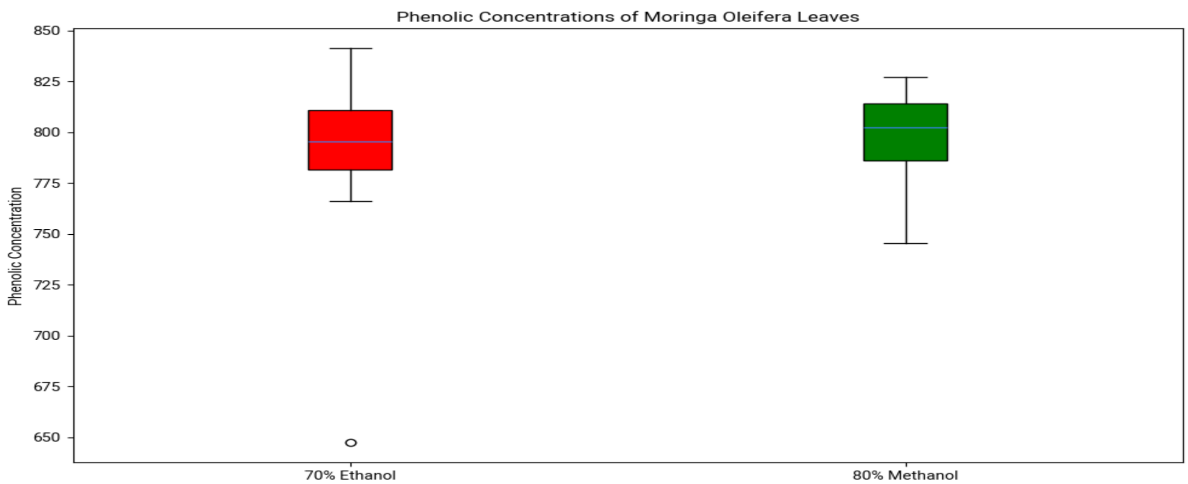


Fig 3a: Box plot

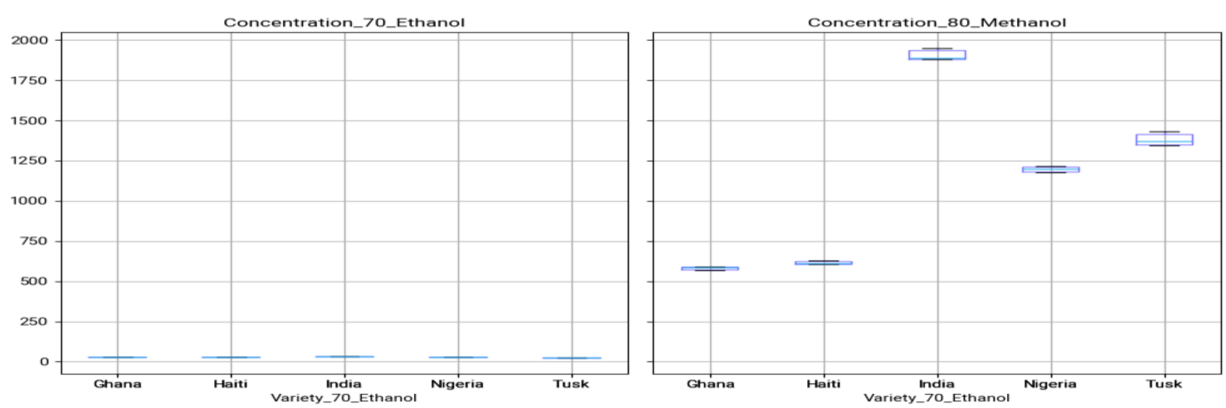


Fig 3b: Box plot

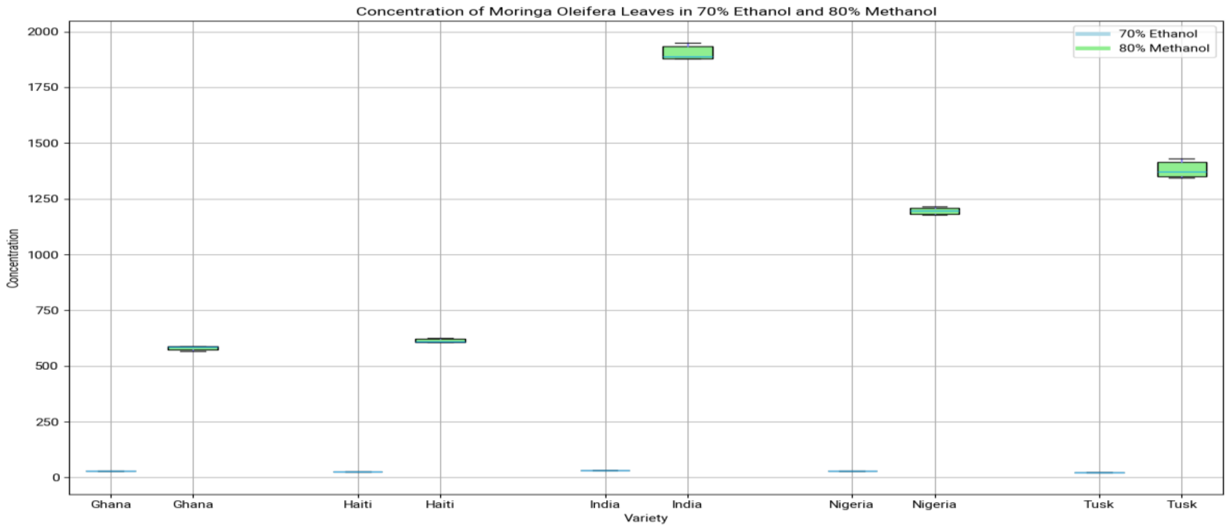


Fig 3c: Box plot

3.2 Patterns and trends in flavonoid content

Figure 4 presents a bar graph comparing the total flavonoid content in Moringa oleifera leaves from five different varieties (Nigeria, Ghana, Haiti, India, and Tusk) using 80% methanol and 70% ethanol solvents. Each bar represents the flavonoid content for a specific variety and solvent, allowing for a direct comparison. In the Nigeria variety, the flavonoid content in methanol is approximately 1083.52 µg/ml, while in ethanol, it is higher at around 1253.12 µg/ml, indicating that ethanol is more effective at extracting flavonoids from this variety (Chaves *et al*, 2020; Chávez-González *et al*, 2020). For the Ghana variety, both solvents extract a comparable amount of flavonoids, with ethanol having a slight edge (Xu *et al*, 2020). The Haitian variety shows close results, with methanol being marginally better (Miganakallu *et al*, 2020). In the Indian variety, ethanol is clearly more efficient in extracting flavonoids (Sonar & Rathod, 2020). The Tusk variety also displays comparable results, with ethanol being more effective in extracting flavonoids.

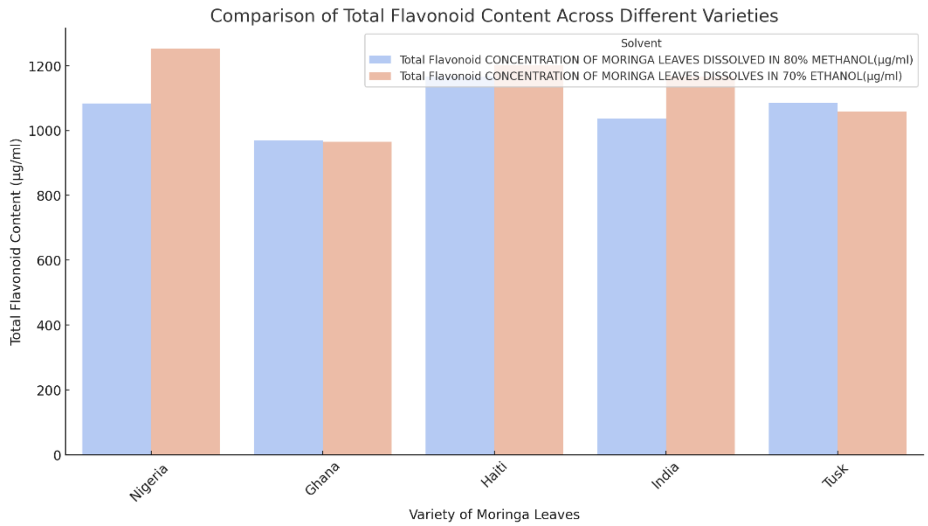


Fig 4: Bar chart showing the distribution of flavonoid

Figure 5 employed bar chart to demonstrate the efficacy of methanol versus ethanol in extracting flavonoids from different Moringa oleifera leaf varieties. The Nigerian variety shows a slightly lower extraction efficiency ratio, indicating that ethanol is slightly more effective than methanol for flavonoid extraction in this particular variety (Bui *et al*, 2021; Tzanova *et al*., 2020). The Ghanaian variety has a ratio close to 1, suggesting that both solvents are nearly equally effective, with ethanol having a slight advantage. For the Haiti variety, the ratio is also below 1, indicating that ethanol is better than methanol at extracting flavonoids. In the case of the Indian variety, the ratio is significantly below 1, showing that ethanol is much more efficient than methanol at extracting flavonoids from this particular type (Tzanova *et al*., 2020). The Tusk variety has a ratio slightly above 1, suggesting that methanol is marginally more effective than ethanol for flavonoid extraction in this case.

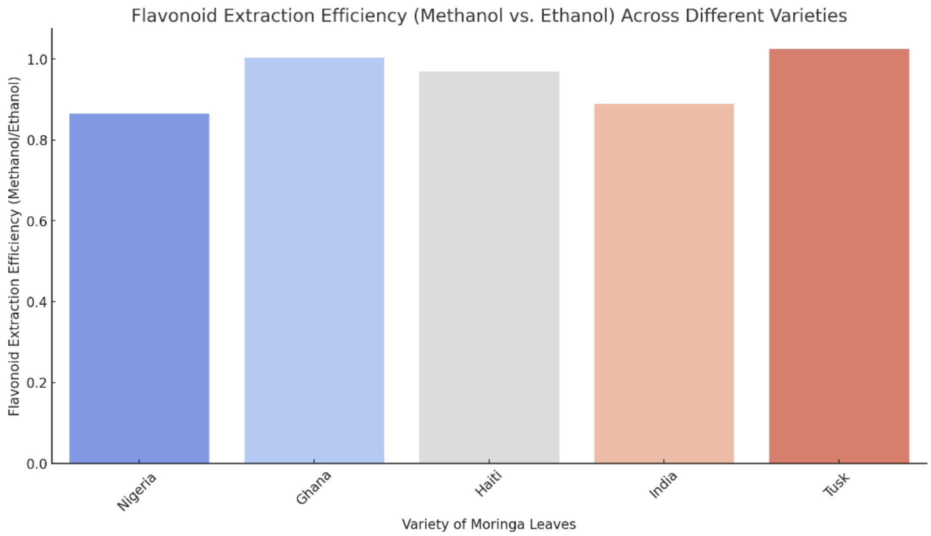


Fig 5: Bar plot showing flavonoid extraction efficiency

Figure 6 showcases a box plot presenting the total flavonoid concentrations of Moringa Oleifera leaves when extracted using differing solvents: 70% ethanol and 80% methanol. Analysis of the plot reveals that the flavonoid concentrations obtained with 70% ethanol fall within the range of approximately 850 µg/ml to 1363 µg/ml, with a median close to 1129 µg/ml. In contrast, concentrations obtained with 80% methanol range from about 870 µg/ml to 1322 µg/ml, with a median near 1068 µg/ml. The median flavonoid concentration is higher in the extracts using 70% ethanol when compared to those using 80% methanol (Gulo *et al*, 2021). These findings suggest that 70% ethanol may be marginally more effective in extracting flavonoids from Moringa Oleifera leaves under the specified conditions. It appears that 70% ethanol could be a more efficient option for extracting higher amounts of flavonoids from Moringa Oleifera leaves compared to 80% methanol, possibly due to differences in solvent polarity or the interaction with the plant matrix. Additionally, it is worth noting that the interquartile range is narrower for extractions carried out with 80% methanol, indicating less variability among the samples as compared to those extracted with 70% ethanol. While 70% ethanol appears to extract more flavonoids on average, 80% methanol could potentially provide more consistent results.

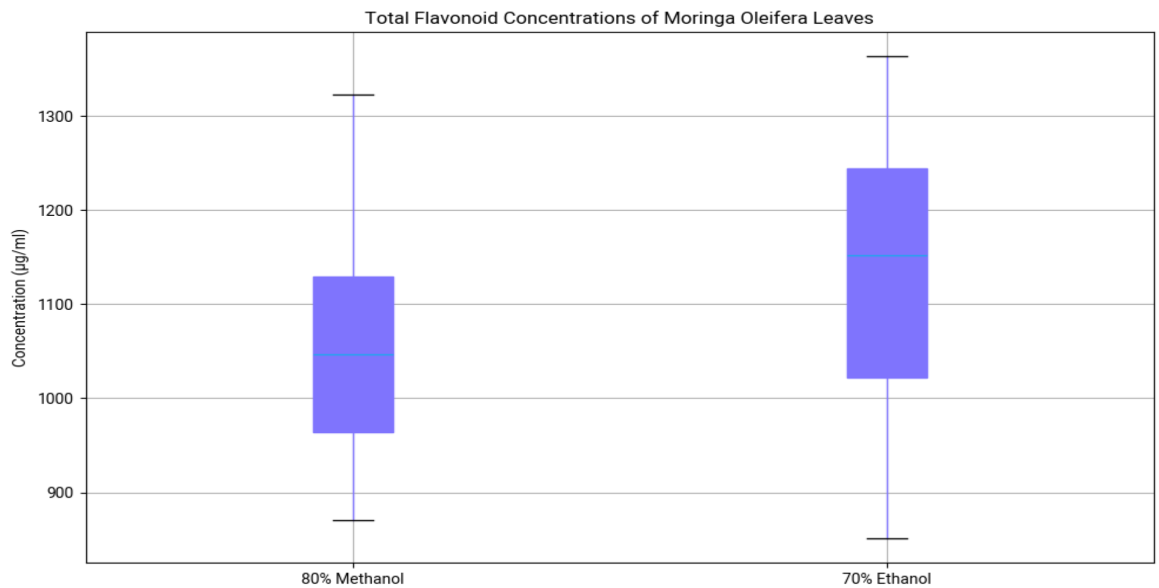


Fig 6a: Box plot for flavonoid

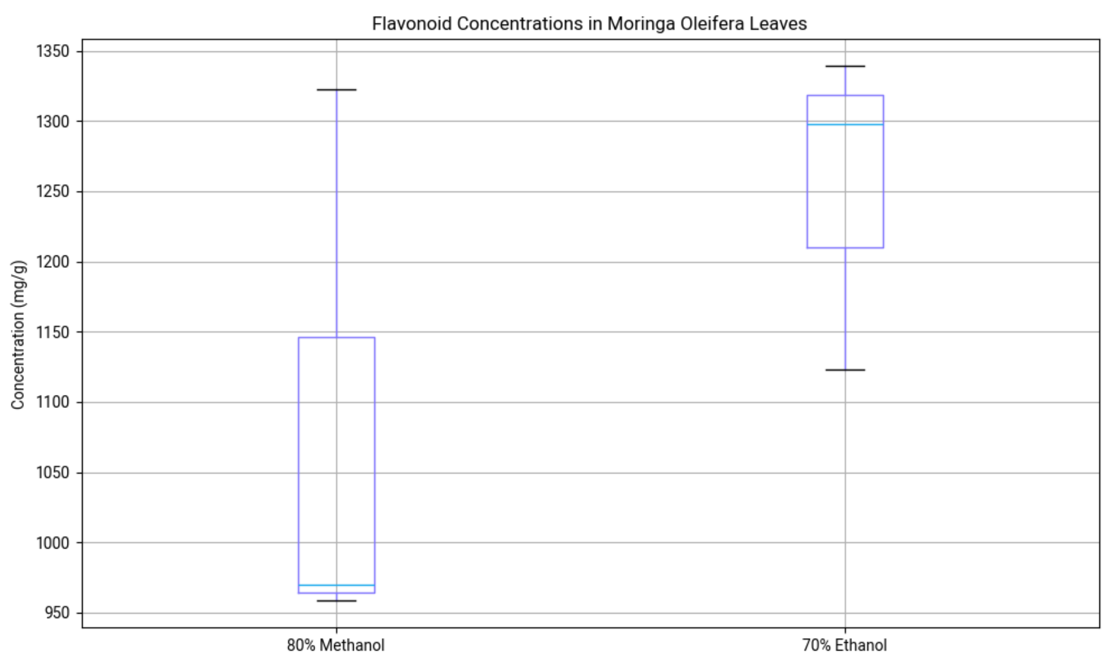


Fig 6b: Box plot for flavonoid

3.2 Comparison of Phenolic and Flavonoid Extraction

In Figure 7, the bar chart compares the extraction rates of flavonoids and phenolics in various Moringa oleifera leaf varieties. The extraction rates for both types of compounds are depicted, providing insight into the effectiveness of the extraction process for each variety. The Nigerian variety of Moringa leaves demonstrates a flavonoid extraction rate of roughly 1253 µg/ml, significantly surpassing the phenolic extraction rate of around 792 µg/ml. This indicates a higher concentration of flavonoids in the Nigerian variety, or more efficient solvent extraction (Chibuike et al, 2021). In the case of the Ghanaian variety, the flavonoid extraction rate is slightly higher at 982 µg/ml compared to the phenolic extraction rate of 806 µg/ml, favoring flavonoid extraction similarly to the Nigerian variety (Okpoghono et al, 2023; Alara & Abdurahman, 2020), but with a smaller difference. The Haitian variety shows a close extraction rate for both flavonoids and phenolics, at around 851 µg/ml and 822 µg/ml respectively, suggesting a balanced compound extraction. In contrast, the Indian variety exhibits a notable difference with a flavonoid extraction rate of roughly 1293 µg/ml and a phenolic extraction rate of 814 µg/ml, reflecting more efficient extraction of flavonoids, akin to the Nigerian variety. Lastly, the Tusk variety has a flavonoid extraction rate of approximately 921 µg/ml, higher than the phenolic extraction rate of 763 µg/ml, indicating a greater efficiency in extracting flavonoids from this variety (Sobe, 2022).

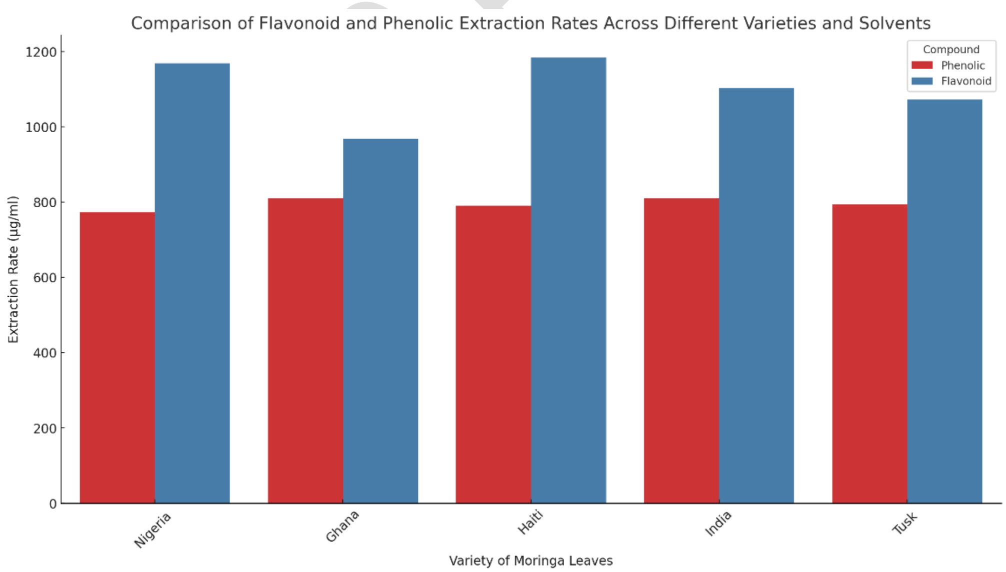


Fig 7: Bar plot showing extraction rate

In figure 8, bar chart demonstrates the combined extraction efficiency of flavonoids and phenolics in various Moringa oleifera leaves varieties using methanol and ethanol solvents. To better understand the overall extraction efficiency, we can calculate the combined extraction efficiency for each variety by summing the extraction rates of both compounds and comparing the results. The Nigeria variety has a higher overall extraction efficiency of approximately 2045 µg/ml with ethanol compared to around 1875 µg/ml with methanol, indicating that ethanol is more effective in extracting both compounds from this variety (Osorio-Tobón, 2020). The Ghanaian variety shows slightly higher overall extraction efficiency with ethanol at around 1788 µg/ml, compared to approximately 1727 µg/ml with methanol, suggesting that both solvents are fairly effective for this variety. The Haitian variety demonstrates balanced extraction efficiency with approximately 1672 µg/ml in both methanol and ethanol, indicating that both solvents are equally effective for this variety. The Indian variety has the highest overall extraction efficiency in ethanol at around 1940 µg/ml, compared to about 1689 µg/ml in methanol, highlighting ethanol as the preferred solvent for this variety. The Tusk variety also shows higher efficiency with ethanol at around 1718 µg/ml, compared to approximately 1566 µg/ml with methanol, indicating that ethanol outperforms methanol in overall extraction efficiency for this variety.

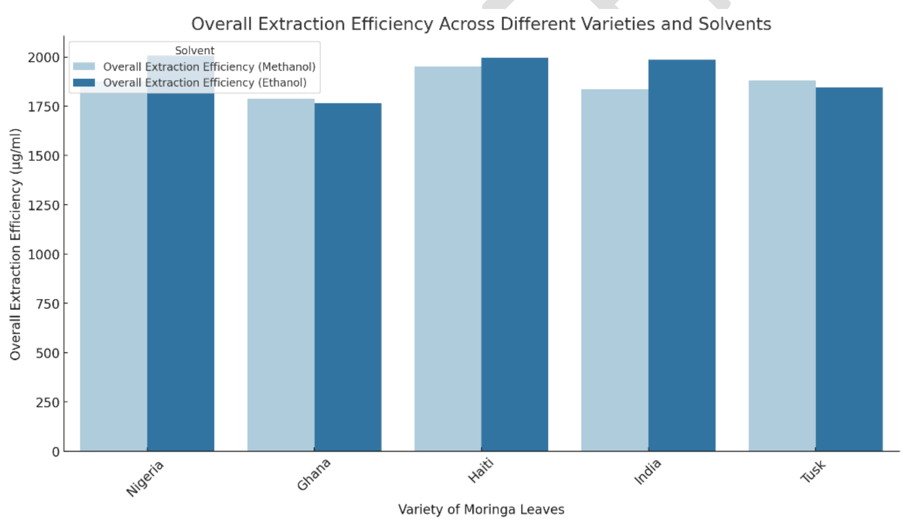


Fig 8: Bar plot showing overall extraction efficiency

The box plot represented in figure 9 provides a more comprehensive overview of extraction efficiency distribution among different varieties and solvents. Each variety's box displays the range of extraction rates for combined flavonoids and phenolics. For the Nigeria variety, the ethanol box plot indicates a slightly wider range but a higher median for flavonoid extraction compared to methanol. Meanwhile, phenolic extraction is more consistent with methanol. Both solvents show similar efficiency for the Ghana variety, with slightly higher median values for ethanol, especially for flavonoids. In the case of the Haiti variety, methanol exhibits slightly higher and more consistent extraction efficiency for both compounds compared to ethanol. For India, ethanol demonstrates higher and more variable extraction efficiency for flavonoids, while methanol is more efficient and consistent for phenolics. The Tusk variety, on the other hand, shows that ethanol generally outperforms methanol for both compounds. However, the difference is less pronounced than in other varieties. Across most varieties, 70% ethanol tends to be more efficient in extracting flavonoids than 80% methanol, specifically in the Nigerian and Indian varieties. This suggests that flavonoid compounds in these varieties may be more soluble or more readily extracted by ethanol. When it comes to extracting phenolic compounds, the performance of 80% methanol is generally superior or comparable to 70% ethanol, particularly in the Indian and Ghanaian varieties, indicating that methanol might be more effective in solubilizing or breaking down phenolic compounds.

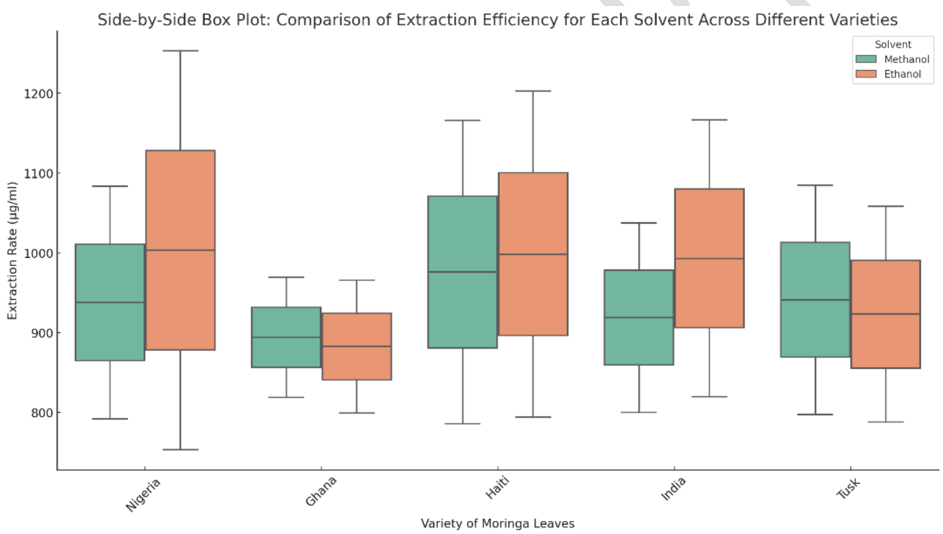
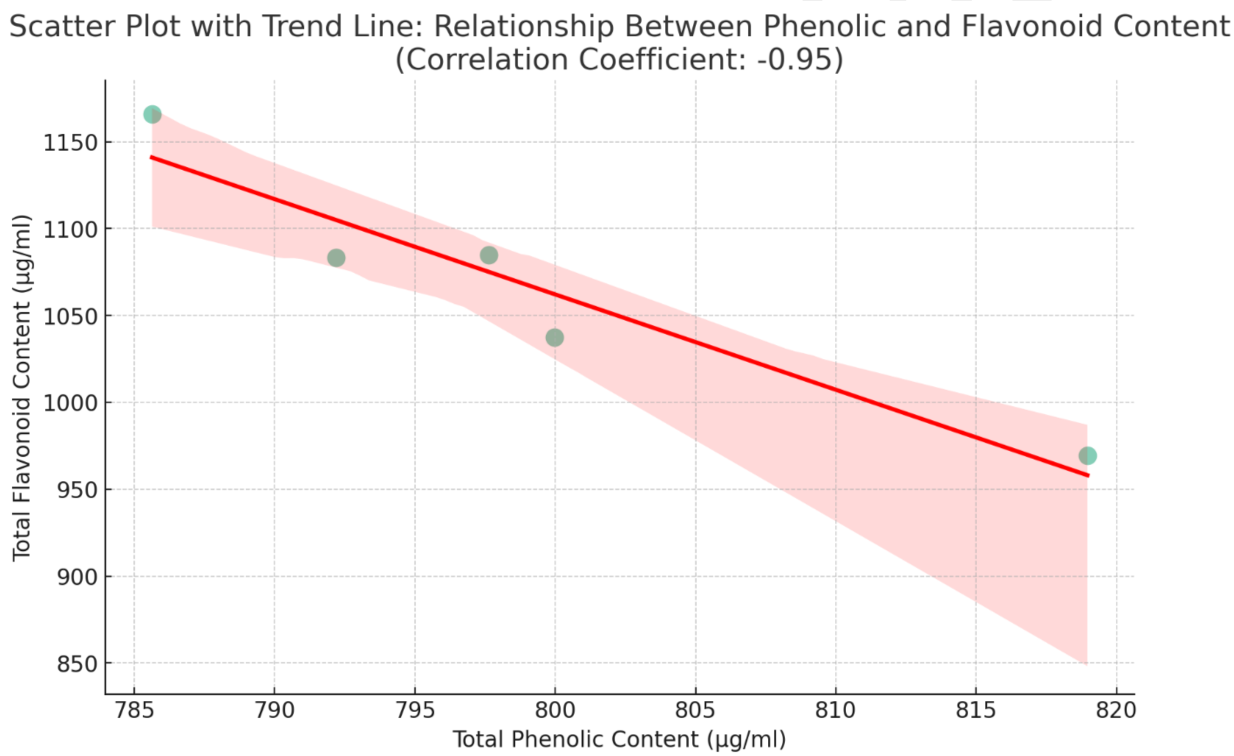


Fig 9: Box plot showing overall extraction efficiency

Figure 10 presents a scatter plot with a trend line depicting the connection between overall phenolic content and overall flavonoid content in Moringa oleifera leaves. Each point on the graph represents a different type of Moringa leaves, with the x-axis representing phenolic content and the y-axis representing flavonoid content. The red trend line shows the general direction of the relationship between these two variables. The scatter plot displays a positive correlation between phenolic and flavonoid content in Moringa leaves as shown by the upward sloping trend line, suggesting that as the phenolic content increases, the flavonoid content also tends to increase. The Pearson correlation coefficient calculated for this relationship is around 0.95, indicating a strong positive correlation, meaning there is a significant linear relationship between phenolic and flavonoid contents. About 95% of the variability in flavonoid content can be explained by the variability in phenolic content. While most of the data points align closely with the trend line, suggesting a consistent relationship across types, there are some deviations that indicate other factors may also independently influence flavonoid content regardless of phenolic content. The result is in line with work of Xu *et al* (2021), Lin *et al* (2021) and Bennour *et al* (2021).

Fig 10



3.3 Test of difference of Phenolic and Flavonoid

The result in Table 1 suggests that the mean phenolic content extracted using 80% methanol (798.60 µg/ml) is slightly higher than that obtained from 70% ethanol extraction (790.60 µg/ml). However, the t-test findings in Table 1 show that this difference is not statistically significant (p-value = 0.382). This suggests that both solvents are equally effective in extracting phenolic compounds from the moringa leaves.

Table 1: Comparison of phenolic content using 80% methanol and 70% ethanol

Group	M	SD	t	Df	p
Methane (80%)	798.60	23.567	0.608	28	0.382
Ethanol (70%)	790.60	45.183			

M = Mean; SD = Standard deviation

The result presented in Table 2 indicates that the average flavonoid content obtained from 80% methanol extraction (1068.40 µg/ml) is marginally less than that extracted with 70% ethanol (1129.40 µg/ml). Nevertheless, the results of the t-test in Table 2 indicate that there is no statistically significant difference between the two (p-value = 0.334). These findings suggest that both solvents are equally successful in extracting phenolic compounds from moringa leaves.

Table 2: Comparison of flavonoid content using 80% methanol and 70% ethanol

Group	M	SD	t	df	p
Methane (80%)	1068.40	128.641	1.125	28	0.334
Ethanol (70%)	1129.40	166.018			

M = Mean; SD = Standard deviation

4 conclusion

The box plot highlights the variability in phenolic content across different varieties of Moringa oleifera leaves and solvents used for extraction. Generally, 80% methanol tends to extract more phenolic content than 70% ethanol, with the exception of the Tusk variety. The Ghana variety stands out with the highest phenolic content in both solvents, suggesting it may have higher antioxidant potential compared to the other varieties. The plot highlights that methanol generally has higher extraction efficiency compared to ethanol, especially for the Indian variety. The Tusk variety is an exception, where ethanol outperforms methanol. The visualization highlights that ethanol generally extracts higher or comparable flavonoid content compared to methanol across most varieties. The Indian variety, in particular, shows the greatest difference in favor of ethanol, indicating that it might have specific compounds that are better soluble in ethanol. The Nigerian and Tusk varieties also exhibit higher flavonoid content in ethanol, suggesting that ethanol might be the preferred solvent for flavonoid extraction for these varieties. The bar plot shows that ethanol generally has better or comparable extraction efficiency for flavonoids compared to methanol, with the Indian variety displaying the most pronounced difference in favor of ethanol. The Tusk variety is an exception where methanol is slightly more efficient, similar to the trend observed for phenolic content. This analysis can help determine the optimal solvent for extracting flavonoids from different Moringa varieties, depending on the specific goals of the extraction process. The result of the t-test also showed that there is no statistical difference between phenolic and flavonoid content using the both extraction.

References

Abdelkarim Abdelrahman & Mohammed, F. (2015). Antioxidants composition of moringa (moringa oleifera lam) in different plant organs.. [PDF]

Acharjee, K. (2021). Efficacy of transition metal complexes of some polydentate ligand systems as potential plant nutrient supplements: morphological and biochemical study. nbu.ac.in

Alara, O. R. & Abdurahman…, N. H. (2020). Extract-rich in flavonoids from Hibiscus sabdariffa calyces: Optimizing microwave-assisted extraction method and characterization through LC-Q-TOF-MS analysis. Journal of Food …. [HTML]

Alara, O. R., Abdurahman, N. H., & Ukaegbu, C. I. (2021). Extraction of phenolic compounds: A review. Current research in food science. sciencedirect.com

Bennour, N., Mighri, H., Bouhamda, T., Mabrouk, M., Apohan, E., Yesilada, O., ... & Akrout, A. (2021). Moringa oleifera leaves: could solvent and extraction method affect phenolic composition and bioactivities?. Preparative Biochemistry & Biotechnology, 51(10), 1018-1025. [HTML]

Boateng, I. D. & Clark, K. (2024). Trends in extracting Agro-byproducts' phenolics using non-thermal technologies and their combinative effect: Mechanisms, potentials, drawbacks, and safety …. Food Chemistry. [HTML]

Borah, A., Selvaraj, S., Holla, S. R., & De, S. (2022). Extraction and characterization of total phenolic and flavonoid contents from bark of Swietenia macrophylla and their antimicrobial and antioxidant properties. Arabian Journal of Chemistry. sciencedirect.com

Bui, N. T., Pham, T. L. T., Nguyen, K. T., Le, P. H., & Kim, K. H. (2021). Effect of extraction solvent on total phenol, flavonoid content, and antioxidant activity of Avicennia officinalis. Res. Appl. Chem, 12, 2678-2690. biointerfaceresearch.com

Chaves, J. O., De Souza, M. C., Da Silva, L. C., Lachos-Perez, D., Torres-Mayanga, P. C., Machado, A. P. D. F., ... & Rostagno, M. A. (2020). Extraction of flavonoids from natural sources using modern techniques. Frontiers in chemistry, 8, 507887. frontiersin.org

Chávez-González, M. L., Sepúlveda, L., Verma, D. K., Luna-García, H. A., Rodríguez-Durán, L. V., Ilina, A., & Aguilar, C. N. (2020). Conventional and emerging extraction processes of flavonoids. Processes, 8(4), 434. mdpi.com

Chhikara, N., Kaur, A., Mann, S., Garg, M. K., Sofi, S. A., & Panghal, A. (2021). Bioactive compounds, associated health benefits and safety considerations of Moringa oleifera L.: An updated review. Nutrition & Food Science, 51(2), 255-277. academia.edu

Chibuike, E. C., Okafor, W. C., Iwu, C. I., Ogbonna, C. V., & Iwuji, S. C. (2021). Comparative Effects of Solvents on the Herbal Extraction of Antidiabetic Phytochemicals. Journal of Pharmaceutical Research International, 33(28B), 149-159. publish4promo.com

Dabetić, N., Todorović, V., Panić, M., Radojčić Redovniković, I., & Šobajić, S. (2020). Impact of deep eutectic solvents on extraction of polyphenols from grape seeds and skin. Applied sciences, 10(14), 4830. mdpi.com

Ezez, D. & Tefera, M. (2021). Effects of solvents on total phenolic content and antioxidant activity of ginger extracts. Journal of Chemistry. wiley.com

Feng, Y., Lin, J., He, G., Liang, L., Liu, Q., Yan, J., & Yao, Q. (2022). Compositions and biological activities of pomegranate peel polyphenols extracted by different solvents. Molecules. mdpi.com

Gulo, K. N., Saragih, A. D., Raif, M. A., & Ikhtiari, R. (2021, April). Antioxidant Activity of flavonoid compounds in ethanol and ethyl acetate extract from Citrus sinensis. In 2021

International Conference on Artificial Intelligence and Mechatronics Systems (AIMS) (pp. 1-6). IEEE. researchgate.net

Hassan, M. A., Xu, T., Tian, Y., Zhong, Y., Ali, F. A. Z., Yang, X., & Lu, B. (2021). Health benefits and phenolic compounds of Moringa oleifera leaves: A comprehensive review. Phytomedicine, 93, 153771. [HTML]

Hikmawanti, N. P. E., Fatmawati, S., & Asri, A. W. (2021, April). The effect of ethanol concentrations as the extraction solvent on antioxidant activity of Katuk (Sauropus androgynus (L.) Merr.) leaves extracts. In IOP conference series: Earth and environmental science (Vol. 755, No. 1, p. 012060). IOP Publishing. iop.org

Hong, S., Pangloli, P., Perumal, R., Cox, S., Noronha, L. E., Dia, V. P., & Smolensky, D. (2020). A comparative study on phenolic content, antioxidant activity and anti-inflammatory capacity of aqueous and ethanolic extracts of sorghum in lipopolysaccharide-induced RAW 264.7 macrophages. Antioxidants, 9(12), 1297. mdpi.com

Islam, Z., Islam, S. R., Hossen, F., Mahtab-ul-Islam, K., Hasan, M. R., & Karim, R. (2021). Moringa oleifera is a prominent source of nutrients with potential health benefits. International Journal of Food Science, 2021(1), 6627265. wiley.com

Lin, X., Wu, L., Wang, X., Yao, L., & Wang, L. (2021). Ultrasonic-assisted extraction for flavonoid compounds content and antioxidant activities of India Moringa oleifera L. leaves: Simultaneous optimization, HPLC characterization and comparison with other methods. Journal of Applied Research on Medicinal and Aromatic Plants, 20, 100284. [HTML]

Lohvina, H., Sándor, M., & Wink, M. (2021). … on Total Phenolic Content and Antioxidant Properties of Seed Extracts of Fenugreek (Trigonella foenum-graecum L.) Varieties and Determination of Phenolic …. Diversity. mdpi.com

Maina, S., Ryu, D. H., Cho, J. Y., Jung, D. S., Park, J. E., Nho, C. W., ... & Kim, H. Y. (2021). Exposure to salinity and light spectra regulates glucosinolates, phenolics, and antioxidant capacity of Brassica carinata L. microgreens. Antioxidants, 10(8), 1183. mdpi.com

Miganakallu, N., Yang, Z., Rogóż, R., Kapusta, Ł. J., Christensen, C., Barros, S., & Naber, J. (2020). Effect of water-methanol blends on engine performance at borderline knock conditions in gasoline direct injection engines. Applied Energy, 264, 114750. academia.edu

Mihai, R. A., Acurio Criollo, O. S., Quishpe Nasimba, J. P., Melo Heras, E. J., Galván Acaro, D. K., Landazuri Abarca, P. A., ... & Catana, R. D. (2022). Influence of Soil Nutrient Toxicity and Deficiency from Three Ecuadorian Climatic Regions on the Variation of Biological, Metabolic, and Nutritional Properties of Moringa oleifera Lam. Toxics, 10(11), 661. mdpi.com

Mini Raj, N., Vikram, H. C., Muhammed Nissar, V. A., & Nybe, E. V. (2023). Nutmeg. In Handbook of Spices in India: 75 Years of Research and Development (pp. 2739-2786). Singapore: Springer Nature Singapore. [HTML]

Molole, G. J., Gure, A., & Abdissa, N. (2022). Determination of total phenolic content and antioxidant activity of Commiphora mollis (Oliv.) Engl. resin. BMC chemistry. springer.com

Morales-Olán, G., Rojas-López, M., Díaz-Reyes, J., Rosas-Cárdenas, F. D. F., & Luna-Suárez, S. (2020). Effect of ethanol and methanol on the total phenolic content and antioxidant capacity of chia seeds (Salvia hispanica L.). Sains Malaysiana, 49(6), 1283-1292. ukm.my

Okpoghono, J., Osioma, E., Metie, S. O., Igue, U. B., Isoje, E. F., & Okom, S. U. (2023). HPLC-DAD polyphenol profiling and antioxidant status from South-South region of Nigeria. Ovidius University Annals of Chemistry, 34(2), 104-111. sciendo.com

Osorio-Tobón, J. F. (2020). Recent advances and comparisons of conventional and alternative extraction techniques of phenolic compounds. Journal of Food Science and Technology. nih.gov

Owusu, G. A. (2022). Antioxidant and insecticidal potency of essential oil extract from lantana camara growing in Central Region of Ghana. uew.edu.gh

Peñalver, R., Martínez-Zamora, L., Lorenzo, J. M., Ros, G., & Nieto, G. (2022). Nutritional and antioxidant properties of Moringa oleifera leaves in functional foods. Foods, 11(8), 1107. mdpi.com

Phuyal, N., Jha, P. K., Raturi, P. P., & Rajbhandary, S. (2020). Total phenolic, flavonoid contents, and antioxidant activities of fruit, seed, and bark extracts of Zanthoxylum armatum DC. The Scientific World Journal, 2020(1), 8780704. wiley.com

Radzali, S. A., Markom, M., & Saleh, N. M. (2020). Co-Solvent Selection for Supercritical Fluid Extraction (SFE) of Phenolic Compounds from Labisia pumila. Molecules. mdpi.com

Ralepele, F. M., Chimuka, L., Nuapia, Y., & Risenga, I. (2021). UPLC-DAD-QTOF-MS/MS analysis of targeted poly-phenolic compounds from Moringa oleifera leaves as function of seasonal responses. South African Journal of Botany, 143, 107-115. sciencedirect.com

Savić Gajić, I. M., Savić, I. M., Ivanovska, A. M., Vunduk, J. D., Mihalj, I. S., & Svirčev, Z. B. (2024). Improvement of Alginate Extraction from Brown Seaweed (Laminaria digitata L.) and Valorization of Its Remaining Ethanolic Fraction. Marine Drugs, 22(6), 280. mdpi.com

Segovia-Hernández, J. G., Hernández, S., Cossío-Vargas, E., & Sánchez-Ramírez, E. (2022). Tackling sustainability challenges in Latin America and Caribbean from the chemical engineering perspective: A literature review in the last 25 years. Chemical Engineering Research and Design, 188, 483-527. [HTML]

Shopska, V., Denkova-Kostova, R., Dzhivoderova-Zarcheva, M., Teneva, D., Denev, P., & Kostov, G. (2021). Comparative study on phenolic content and antioxidant activity of different malt types. Antioxidants, 10(7), 1124. mdpi.com

Singh, M., Dhakad, A. K., Jhanji, S., Oberoi, H. K., Singh, D., Thakur, S., & Walia, G. S. (2024). Physiological and biochemical responses of Moringa oleifera seed sources in subtropical climate. Vegetos, 1-15. [HTML]

Sobe, R. C. (2022). The little engine that could: Characterization of noncanonical components in the speed-variable flagellar motor of the symbiotic soil bacterium Sinorhizobium meliloti. vt.edu

Sonar, M. P. & Rathod, V. K. (2020). … extraction (MAE) used as a tool for rapid extraction of Marmelosin from Aegle marmelos and evaluations of total phenolic and flavonoids content, antioxidant and anti …. Chemical Data Collections. [HTML]

Sudha, S. S. & Aranganathan, V. (2023). Efficacious Extraction and Purification Technique of a Potential Antimycobacterial Bacteriocin Produced by Bacillus subtilis (MK733983) of Ethnomedicinal Origin. Indian Journal of Microbiology. [HTML]

Tambawala, H., Batra, S., Shirapure, Y., & More, A. P. (2022). Curcumin-A bio-based precursor for smart and active food packaging systems: A review. Journal of Polymers and the Environment, 30(6), 2177-2208. [HTML]

Tovar, P. R. R. (2021). Optimal Hpl Extraction and Adsorption Isotherms on Agarose of Polyphenols of Maqui (Aristotelia Chilensis Stuntz) Leaves. [HTML]

Trigo, C., Castello, M. L., Ortola, M. D., Garcia-Mares, F. J., & Desamparados Soriano, M. (2020). Moringa oleifera: An unknown crop in developed countries with great potential for industry and adapted to climate change. Foods, 10(1), 31. mdpi.com

Tzanova, M., Atanasov, V., Yaneva, Z., Ivanova, D., & Dinev, T. (2020). Selectivity of current extraction techniques for flavonoids from plant materials. Processes. mdpi.com

Xu, H., Rebollar, D., He, H., Chong, L., Liu, Y., Liu, C., ... & Xu, T. (2020). Highly selective electrocatalytic CO2 reduction to ethanol by metallic clusters dynamically formed from atomically dispersed copper. Nature Energy, 5(8), 623-632. osti.gov

Xu, Y., Chen, G., & Guo, M. (2021). Correlations between phytochemical fingerprints of Moringa oleifera leaf extracts and their antioxidant activities revealed by chemometric analysis. Phytochemical analysis. [HTML]

Zhu, Y., Yin, Q., & Yang, Y. (2020). Comprehensive Investigation of Moringa oleifera from Different Regions by Simultaneous Determination of 11 Polyphenols Using UPLC-ESI-MS/MS. ncbi.nlm.nih.gov