Mongolian Journal of Agricultural Sciences, *Vol.16, No.38 (2023)*
https://doi.org/10.5564/mjas.v16i38.3319

Biochemical analysis of radish and beet species

Bayarmaa Jambalsuren*, Purev Dondog

Department of Biology, School of Arts and Sciences, National University of Mongolia, University Street, Ulaanbaatar 14201, Mongolia

*Corresponding author: bayarmaa@num.edu.mn

https://orcid.org/0000-0002-3037-7269

Received: 01.10.2022 Revised: 27.08.2023 Accepted: 15.09.2023

Abstract
The study investigated the biochemical composition and enzyme activities of various radish and beet species, revealing significant variations across different parameters. Reducing sugars, crucial for blood sugar regulation and weight management, showed diverse levels among species, with higher concentrations observed in yellow turnip and white radish. Likewise, fiber content varied, with *Raphanus sativus L. var. niger J. Kern* exhibiting the highest fiber content. Moisture and mineral content also differed across species, with sugar beet displaying the highest mineral content. Regarding vitamin C content, our findings indicated lower levels compared to previous studies, suggesting potential influences of environmental conditions on vitamin accumulation. Flavonoid content, known for its antioxidant properties, was notably high in black radish compared to other species, highlighting the potential health benefits of flavonoid-rich varieties. Enzyme activities, including catalase, peroxidase, and polyphenol oxidase, varied among species, with white radish and beetroot exhibiting higher catalase activity. These findings highlight biochemical variations among radish and beet species, underscoring their potential health benefits and applications.

Keywords: radish and beet, fiber, flavonoids, antioxidant enzymes

Introduction
Vegetables are essential for providing vital macro- and micronutrients, antioxidants, and enzymes necessary for human health. Their consumption is widely acknowledged as integral to maintaining a balanced and healthy lifestyle. Dietary habits are intricately linked to geographical, climatic, agricultural, and demographic factors.
In Mongolia, a shift from nomadic animal husbandry to settled lifestyles has led to increased consumption of agricultural products, including vegetables. While potatoes, cabbage, carrots, and turnips have traditionally dominated vegetable crop production in Mongolia, there has been a growing interest in diversifying the vegetable repertoire. There is a growing interest in the cultivation of black radishes, spurred by research indicating their potential advantages for individuals with diabetes. Consequently, several Mongolian pharmaceutical manufacturers are now domestically cultivating black radishes for tea and biologically active compounds. However, little is known about the biochemical parameters of black radishes grown in Mongolia, prompting the need for this study.
The prevalence of oxidative stress-related health conditions, such as irritable bowel syndrome, inflammatory bowel disease, ulcers, and Crohn's disease, underscores the importance of antioxidant supplements, including antioxidant enzymes, in managing these conditions. While research on antioxidant enzymes in various food sources and their changes during storage and processing is extensive in other countries, such investigations are limited in Mongolia.
Therefore, this paper aims to fill this gap by presenting the results of assessing biochemical parameters and antioxidant enzyme activity in five species of radishes and beets commonly used for food, medical, and animal feed purposes in Mongolia.

Material and Methods

Sample Preparation:
In the spring of 2019, five plant species were cultivated in the same field: *Raphanus raphanistrum sub sp. Sativus (L.) Domin*. (rutabaga), *Raphanus sativus L. var. niger J. Kern*. (black radish), *Raphanus sativus L. var. longipinnatus Bailey*. (white radish), *Beta vulgaris L.* (beetroot), and *Beta vulgaris 'saccharifera 'ALEF* (sugar beet). These plants were allowed to grow under identical environmental conditions until harvested in October. Upon harvest, plant samples were cleaned of soil and green parts, and subsequently stored in a freezer until further analysis.

Measurements:
The study involved the evaluation of various parameters to assess the biochemical composition and enzyme activities in the studied plant species. Moisture and mineral content were determined gravimetrically following established protocols to understand the water and mineral composition of the samples. Fiber content was assessed using standardized methods to measure the dietary fiber content present in the plants. Soluble protein levels were measured spectrophotometrically to quantify the amount of soluble proteins present. Reducing sugar content was determined using the Offner method, providing insights into the sugar composition of the samples. Vitamin C content was assessed via redox titration, allowing for the determination of the vitamin C concentration in the samples. Flavonoid content was determined spectrophotometrically to measure the flavonoid concentration present in the plant species. Additionally, enzyme activities were evaluated, including catalase, peroxidase, polyphenol oxidase, ascorbate peroxidase, and superoxide dismutase. Catalase activity was quantified using the Johnson Temple method, while peroxidase and polyphenol oxidase activities were determined according to K.A.Kozlov's method. Ascorbate peroxidase and superoxide dismutase activities were quantified spectrophotometrically, providing insights into the antioxidant enzyme activity present in the samples.
Each parameter was assessed 3-5 times, and the average value was calculated. Enzyme activities were expressed in units (U) per gram of the sample.

Results and Discussions

In this study, the biochemical composition and enzyme activities of various radish and beet species were investigated, revealing significant variations across different parameters.
Reducing sugars include mono-, oligo-, and polysaccharides containing free aldehyde and keto groups, which play crucial roles in lowering blood sugar levels and weight loss [16]. In our study, the content of reducing sugars in radishes and beets ranged from 0.48% to 3.14%. More specifically, *Beta vulgaris* 'saccharifera 'ALEF, *Raphanus sativus L. var. niger J. Kern*, and *Beta vulgaris L.* contained relatively small amounts (1.8%, 1.67%, and 0.48% respectively), while *Raphanus raphanistrum sub sp. Sativus (L.) Domin* (yellow turnip) and *Raphanus sativus L. var. longipinnatus Bailey* (white radish) exhibited higher content, estimated at 3.0% to 3.14%.
Dietary fiber plays an essential role in regulating intestinal mobility, enhancing stool excretion, improving gastrointestinal tract function, and aiding in the elimination of harmful waste products from the body. [17]. Our results indicated that *Raphanus sativus L. var. niger J. Kern* had the highest fiber content (2.07%), followed by *Raphanus raphanistrum sub sp. Sativus (L.) Domin* (1.79%), *Raphanus sativus L. var. longipinnatus Bailey* (0.99%), *Beta vulgaris 'saccharifera 'ALEF*, and *Beta vulgaris L.* (0.77% and 0.76% respectively).
Moisture content across the species ranged from 77.62% to 80.92%, while mineral content in radishes and beets varied from 0.83% to 2.01%. Sugar beet (*Beta vulgaris* 'saccharifera 'ALEF) exhibited the highest mineral content at 2.01%, while white radish had the lowest at 0.83%. Turnip, black radish, and beetroot had relatively similar mineral content (1.03%, 1.23%, 1.28% respectively).
As investigated by Duy et al. (2020), the antifungal activity of *Raphanus sativus L.* (white radish) extract and its potential to prolong the shelf-life of cake were examined, with the study reporting the following biochemical composition for white radish: moisture (91.1%±1.35), soluble protein (0.73%±0.12), carbohydrates (2.67%±0.16), and sum of minerals (0.46%±0.27) [18].

Our results closely align with these findings, indicating similar compositions in Table 1.

Table 1.

Biochemical parameters and activity of antioxidant enzymes in radishes and beets

	Parameters	**Species of beets and radishes**				
		Raphanus raphanistrum sub sp. Sativus (L.) Domin	*Raphanus sativus L. var. niger J. Kern.*	*Raphanus sativus L. var. longipinnatus Bailey.*	*Beta vulgaris L.*	*Beta vulgaris 'saccharifera 'ALEF*
1	Moisture, %	79,86	80,92	77,62	77,77	75,71
2	Soluble protein, %	1,48	0,60	1,04	4,20	3,64
3	Reducing sugar, %	3,14	1,67	3,00	0,48	1,80
4	Fiber, %	1,79	2,07	0,99	0,76	0,77
5	Minerals, %	1,03	1,23	0,83	1,28	2,01
6	Vitamin C, µg%	61,60	783,73	44,88	193,60	204,69
7	Flavonoids, mg%	19,48	83,95	21,45	22,66	26,80
8	Catalase, U	17,00	5,95	90,95	90,53	30,18
9	Peroxidase, U	0,32	0,14	0,42	0,43	1,30
10	Polyphenol oxidase, U	0,14	0,10	0,05	1,35	6,40
11	Ascorbate peroxidase, U	2,16	1,55	1,76	0,81	3,07
12	Superoxide dismutase, U	ND	ND	ND	ND	ND

Vitamin C, a water-soluble antioxidant found abundantly in fruits and vegetables, was measured in white radish by Ogunlesi et al. (2010) using cyclic voltammetry and titration, resulting in values of 39.19 mg% and 40.82 mg% [19]. Additionally, Satpathy et al. (2021) determined vitamin C levels in white radish through iodine redox titration, yielding a result of 35.70 mg% [20]. Agarwal et al. (2014) quantified vitamin C content in white radish spectrophotometrically with 2,4-dinitrophenylhydrazine in leaves and roots, resulting in values of 42.9 µg% and 8.25 µg% respectively [21]. Our results revealed a low vitamin C content in the samples. Importantly, the accumulation of vitamin C is known to be influenced by growing conditions [22], with light exposure being a significant factor [23]. This suggests that the observed vitamin C content in our study samples may have been impacted by these environmental conditions.

Flavonoids, a diverse group of plant polyphenols, are well-known for their antioxidant activity, protecting plants from radiation and pathogens by absorbing UV radiation. In medicinal treatment, flavonoids like quercetin and rutin, known as vitamin P, are widely utilized alongside vitamin C to strengthen blood vessels and enhance blood clotting Moreover, compounds within the flavonoid group exhibit strong antimicrobial properties [24]. A study conducted by Kim et al. (2016) revealed the total content of polyphenols, flavonoids, and antioxidant activity in two varieties (Valentine and Cherry Belle) of white radish. They found the total phenolic content of ethanol extracts to be 160.38 ± 5.0 mg GA/g (Cherry Belle) and 124.46 ± 6.13 mg GA/g (Valentine), while the concentration of total flavonoids in Raphanus sativus L. was 42.93 ± 1.58 mg/g and 16.26 ± 1.84 mg/g (Cherry Belle and Valentine) rutin equivalent per gram dry weight respectively [25]. Our results indicated a relatively high total flavonoid content in black radish (83.95 mg%) compared to similar levels observed in other species (19.48 - 22.66 mg%).

Enkhtsetseg et al. (2019) determined the optimal drying temperature to preserve the antioxidant activity of black radish, employing the DPPH scavenging assay for measuring antioxidant activity, Folin-Ciocalteu reagent for total phenolic content, and spectrophotometry for flavonoid content determination [26]. In contrast to their findings, our study revealed a five-fold higher total flavonoid content in black radish. The difference in our findings could be explained by the fact that we used unpeeled samples, whereas Enkhtsetseg et al. utilized peeled samples. Furthermore, our data exhibited a total flavonoid content 3.7 times higher than that observed in other species (turnip, white radish, beetroot, and sugar beet).

Geographical, climatic, and environmental conditions, as well as sunlight and soil composition, all influence the accumulation of vitamins and plant secondary metabolites [27]. The enzymatic antioxidant system of plants, comprising enzymes such as catalase, peroxidase, polyphenol oxidase, ascorbate peroxidase, and superoxide dismutase, plays a crucial role in this process. In our study, we evaluated the activity of these antioxidant enzymes in various radish and beet species.

Our findings showed that superoxide dismutase activity was undetectable, whereas peroxidase (0.14-1.30 U), polyphenol oxidase (0.05-6.40 U), and ascorbate peroxidase (0.81-3.07 U) activities were low. While, catalase activity ranged from 5.95 to 90.95 U in white radish (*Raphanus sativus L. var. longipinnatus Bailey*) and beetroot (*Beta vulgaris L.*), which was 3-15 times higher than in other species.

In line with our investigation, Motamedi et al. (2022) explored changes in catalase, peroxidase, and glutathione peroxidase activities in radish seedlings subjected to the allelopathic effect of safflower. Their findings revealed that both allelopathic interactions and drought stress contributed to an increase in antioxidant enzyme activity [28]. Similarly, Jayakumar et al. (2007) examined the effects of cobalt stress on various parameters, including growth, biochemical constituents, mineral content, and antioxidant enzyme activity in white radish (*Raphanus sativus L.*). They observed that peroxidase and polyphenol oxidase activities were heightened with increasing cobalt concentration [29]. Additionally, Oudah et al. (2017) investigated the antioxidant response of white radish and cress plantlets to lead exposure, observing a significant decline in catalase and peroxidase activity with reduced oxidative stress [30]. While these prior studies primarily focused on white radish (Raphanus sativus L.), our research extends to encompass yellow turnip, black radish, beetroot, and sugar beet species cultivated under similar conditions, providing valuable insights into their unique biochemical characteristics and enzyme activities.

In summary, our study provides insights into the biochemical composition and enzyme activities of different radish and beet species. We observed variations in parameters like reducing sugars, fiber content, mineral composition, flavonoid content, and antioxidant enzyme activity. These findings highlight the nutritional significance of radishes and beets and offer potential avenues for future research and practical applications in fields like nutrition, medicine, and agriculture.

Conflict of Interests

The authors declare no conflict of interests.

Authors' Contribution

B.J. performed quantitative and qualitative analysis edited and revised the manuscript; P.D. performed statistical analysis of data.

References:

[1] "Introduction to the agricultural sector 2021", Bulletin. National Statistical Commission. Ulaanbaatar, 2022, pp 82-95. (in Mongolian)

[2] B.K. Singh, S.R.Sharma, B.Singh, "Antioxidant enzymes in cabbage: Variability and inheritance of superoxide dismutase, peroxidase and catalase", Scientia Horticulturae. Vol.124, No.1, pp.9-13. Feb. 2010. https://doi.org/10.1016/j.scienta.2009.12.011

[3] M.Patnaik, U.C.Basak, "Enzymatic antioxidant activities in eight wild edible fruits of Odisha", Tropical Plant Research, Vol.1, No.3, pp.36-42. Dec. 2014. Corpus ID:54179777

[4] S.J.Kim, D. Han, M.H.Park, J.S.Rhee, "Screening for Superoxide Dismutase-like Compounds and Its Activators in Extracts of Fruits and Vegetables", Bioscience, Biotechnology and Biochemistry. Vol. 58, No.12, pp.2263-2265, May 1994. https://doi.org/10.1271/bbb.58.2263

[5] P.Tsouvaltzis, J.K.Brecht, "Changes in quality and antioxidant enzyme activities of bunched and topped radish (Raphanus sativus L.) plants during storage at 5 or 10oC", Journal of Food Quality, Vol.37, No.3, pp 157-167, Apr.2014. https://doi.org/10.1111/jfq.12082

[6] B.I.Erguder, A.Avci, E.Devrim, I.Dubak, "Effects of Cooking Techniques on Antioxidant Enzyme Activities of Some Fruits and Vegetables", Turk J Med Sci, Vol. 37, No.3, pp151-156, Jun. 2007.https://journals.tubitak.gov.tr/medical/vol37/iss3/7.
[7] Z.Zeng, X.He, C.Li, H.Chen,L.Liu, X.Feng, "Oral delivery of antioxidant enzymes for effective treatment of inflammatory disease", Biomaterials, Vol.271, pp 4843-4860, Apr.2021. https://doi.org/10.1016/j.biomaterials.2021.120753
[8] "Chemical analysis of medicinal plants", edited by N.I.Grinkevich and L.N.Safronovich,"Vuishaya Skola", Moscow, 1983, pp 89-93, 171-173. (in Russian)
[9] B.P.Pleshkov, Practicum on plant biochemistry, "Kolos", Moscow, 1985, pp 15-17, 104-108. (in Russian)
[10] O.M.Afuwape, O.M.Ayodele, T.G.Obe, "Quantitative determination of ascorbic acid content in some fruit peels obtained in Ado-Ekiti by redox titration", World Journal of Advanced Research and Reviews, Vol.15, No.1, pp 306-310, 9 Jul. 2022. https://doi.org/10.30574/wjarr.2022.15.1.0659
[11] B.Pandey, M.Rajbhandari, "Estimation of Total Phenolic and Flavonoid Contents in Some Medicinal Plants and Their Antioxidant Activities", Nepal Journal of Science and Technology, Vol. 15, No.1, pp 53-60, Oct.2014. https://doi.org/10.3126/njst.v15i1.12010
[12] D.Purev, J.Bayarmaa, "Enzymology", "Udam Soyol", Ulaanbaatar, 2012, pp 371. (in Mongolian)
[13] F.Kh. Khaziev, "Methods of soil enzymology", "Nauka", Moscow, 2005, pp 41-43. (in Russian)
[14] M.Mizuno, M.Kamei, H.Tsuchida, "Ascorbate peroxidase and catalase cooperate for protection against hydrogen peroxidase generated in potato tubers during low-temperature storage", Biochemistry and Molecular Biology International. Vol. 44, No. 4, pp 717-726, Apr.1998. https://doi.org/10.1080/15216549800201762
[15] J.M.McCord, I.Fridovich, "Superoxide dismutase. An enzymatic function for erythrocuprein (hemocuprein)", Journal of Biological Chemistry, Vol.244, No.22, pp 6049-6055, Nov.1969. https://doi.org/10.1016/S0021-9258(18)63504-5
[16] M.Zaitoun, M.Ghanem, S.Harphoush, "Sugars: Types and Their Functional Properties in Food and Human Health", International Journal of Public Health Research, Vol.6, No.4, pp 93-99, Oct. 2018. http://www.openscienceonline.com/journal/ijphr ISSN: 2381-4829 (Print); ISSN: 2381-4837 (Online).
[17] T.M. Barber, S.Kabisch, A.F.H. Pfeiffer, M.O.Weickert, "The Health Benefits of Dietary Fibre", Nutrients, Aug.2020, 12, 3209. https://doi.org/10.3390/nu12103209
[18] H.H.Duy, P.T.K. Ngoc, L.T. H.Anh, D.T. A.Dao, D.C.Nguyen, V.T.Than, "In Vitro Antifungal Efficacy of White Radish (Raphanus sativus L.) Root Extract and Application as a Natural Preservative in Sponge Cake", Processes, Vol.7, 549, 2019, https://doi.org/10.3390/pr7090549
[19] M.Ogunlesi, W.Okiei, L.Azeez, V.Obakachi, M.Osunsanmi, G.Nkenchor, "Vitamin C contents of tropical vegetables and foods determined by volta metric and titrimetric methods and their relevance to the medicinal uses of the plants", Int. J. Electrochem. Sci., Vol.5, pp105-115, Jan. 2010, https://doi.org/10.1016/S1452-3981(23)15270-9
[20] L.Satpathy, N.Pradhan, D.Dash, P.P.Baral, S.P.Parida, "Quantitative Determination of Vitamin C Concentration of Common Edible Food Sources by Redox Titration Using Iodine Solution", Letters in Applied Nano Sciense, Vol.10, No.3, pp 2361- 2369, 2021. https://doi.org/10.33263/LIANBS103.23612369
[21] K.Agarwal, R.Varma, "Radical Scavenging Ability and Biochemical Screening of a Common Asian Vegetable Raphanus sativus L.", Int. J. Pharm. Sci. Rev. Res., Vol.27, No.1, pp 127-134, Jul.-Aug. 2014.
[22] E.Bimenindavy, O.A.Tymofeeva, "Influence of ecological and geographical condition on the content of phenolic compounds and ascorbic acid in medicinal plants", Biomics, Vol 12, No 4, pp 475 - 479, 2020. https://doi.org/10.31301/2221-6197.bmcs.2020-41
[23] C.Paciolla, S.Fortunato, N.Dipierro, A.Paradiso, S.DeLeonardis, L.Mastropasqua, M.Concetta de Pinto, "Vitamin C in Plants: From Functions to Biofortification. Review", Antioxidants, 8, 519, Oct.2019. https://doi.org/10.3390/antiox8110519
[24] A.Ullah, S.Munir, S.L.Badshah, N.Khan, L.Ghani, B.G.Poulson, A.H.Emwas, M.Jaremko, "Important Flavonoids and Their Role as a Therapeutic Agent", Molecules, Vol. 25, No.22, 5243. Nov.2020. https://doi.org/10.3390/molecules25225243

[25] J.K.Kim, T.B.Baskar, S.U.Park, "Total Phenolic and Flavonoid Contents and Antioxidant Activities of Two Raphanus sativus L. cultivars (Cherry Belle and Valentine)", Biosciences Biotechnology Research Asia, Vol. 13, No.1, pp31-36, Mar.2016. https://doi.org/10.13005/bbra/1999

[26] L.Enkhtsetseg, E.Lkhamsuren, E. Enkhtsetseg, "Study of factors affecting the antioxidant activity of black radish (Raphanus sativus L. var. niger)", KhulelTogoot-2019. Materials of scientific conference, "Erdem" printing, Ulaanbaatar, 2019, pp 41-46. (in Mongolian)

[27] P.Pant, S.Pandey, S. Dall'Acqua, "The Influence of Environmental Conditions on Secondary Metabolitesin Medicinal Plants: A Literature Review", Chemistry and Biodiversity, Nov.2021, 18. https://doi.org/10.1002/cbdv.202100345

[28] M.Motamedi, H.Karimmojeni, F.G.Sini, M.M.Majidi, "Changes in activities of antioxidant enzymes in radish (Raphanus sativus) seedlings in response to allelopathic effect of safflower (Carthamus tinctorius)", Brazilian Journal of Pharmaceutical Sciences, 58; e19017, Jan.2022, http://dx.doi.org/10.1590/s2175-97902022e19017.

[29] K.Jayakumar, C.A.Jaleel, P.Vijayarengan, "Changes in Growth, Biochemical Constituents, and Antioxidant Potentials in Radish (Raphanus sativus L.) under Cobalt Stress", Turk J BiolVol.31, pp127-136, Jan.2007.Corpus ID:85578129.

[30] H.K.Oudah, M.M.Jawad, E.H.Abed, "Role of enzymes catalase, peroxidase and amino acid (proline) in Raphanus sativus and Lepidium sativusin exposure levels different water pollution of ion lead", Journal of Science, Jan.2017, Vol. 58, No.2A, pp592-599, https://ijs.uobaghdad.edu.iq/index.php/eijs/article/view/6078