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菜心（Càixīn）

Flowering Chinese cabbage

Brassica campestris L. ssp. *chinensis* var. *utilis* Tsen et Lee

The influence of selected factors on the yield and physicochemical parameters of flowering Chinese cabbage (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee)

Wpływ wybranych czynników na plonowanie i wartość biologiczną kapusty chińskiej kwitnącej (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee)

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by
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List of publications constituting the dissertation

1. **Liu W**., Liu Y., Kleiber T. 2021. A review of progress in current research on Flowering Chinese Cabbage (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee). *J. Elem.*, 26(1): 149-162. DOI: 10.5601/jelem.2020.25.4.20

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List of Abbreviations

ABTS, 2,2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)
Caixin (菜心), the Chinese name of flowering Chinese cabbage in Mandarin
ch-OSA, choline-stabilized orthosilicic acid
Choy sum, the Chinese name of flowering Chinese cabbage in Cantonese dialect
CRNFs, controlled-release nitrogen fertilisers
DPPH, 2,2-diphenyl-1-picrylhydrazyl free radical
EC, Electrolytic conductivity
FAAS, flame atomic absorption spectroscopy
FCR, Folin–Ciocalteu reagent
FRAP, Ferric ion reducing antioxidant power
FW, fresh weight
GAE, gallic acid equivalents
GSH-As cycle, Glutathione-ascorbate cycle
GSLs, Glucosinolates
IC_{25} = 120 µg/ml (Half maximal Inhibitory Concentration)
N-50, N nutrition level (50 mg dm^{-3})
N-70, N nutrition level (70 mg dm^{-3})
N-90, N nutrition level (90 mg dm^{-3})
N-110, N nutrition level (110 mg dm^{-3})
N-130, N nutrition level (130 mg dm^{-3})
NS, nutrient solution
PBS solution, Phosphate-buffered saline solution
SA, salicylic acid
TE, Trolox equivalents
TFC, total flavonoid content.
the ORAC assay, the oxygen radical absorbance capacity)
TPC, total polyphenol content
TPTZ, Fe^{3+}-tripyridyl-trazine

Abstract

Flowering Chinese cabbage (*Brassica campestris* L. *ssp. chinensis var. utilis* Tsen et Lee) is a popular leafy vegetable originated from China and widespread in southeast Asia. The short growing period results in high values of the multiple cropping index, which means the same land could obtain higher total yield. However, flowering Chinese cabbage is not widely and commercially cultivated in Europe yet. The plant is rich in bioactive compounds.

The aim of the studies was to evaluate the effect of selected factors on yield and quality of flowering Chinese cabbage. Three hypotheses were set up in this dissertation: (I) Modifying plant nutrition with nitrogen, both its intensity and the source of the nutrient, influence the yielding of flowering Chinese cabbage, (II) foliar application with salicylic acid, V, Li, Se and Se influences the yielding of flowering Chinese cabbage. (III) Flowering Chinese cabbage shows suitability for soilless cultivation, both in substrate (mixture of mineral soil + peat; pot cultivation) and in hydroponic system.

The studies were conducted between the year 2018 and 2020. The yield and physicochemical parameters were determined by fresh weight, nutrient content, colour, chlorophyll content, carotenoid content, total phenolic content, total flavonoid content, and the antioxidant activity. Each experiment was conducted in 2 independent growing systems (pot and hydroponic).

The obtained results showed that increasing nitrogen nutrition positively influence the yielding of plant, in both pot and hydroponic cultivation systems. The yield was increased with N level increase from N-50 to N-90. Generally, the optimal nitrogen level was N-90. In substrate, the N content of plant increased with the increase of nutrient level whereas in hydroponic, the N content of plant was stable. The spring samples were characterized by higher values for the antioxidant activity and phenolic content than the autumn samples. During autumn cultivation in the substrate, the N content in plants was about 60% higher in comparison to the samples planted in spring. The plants grown in spring obtained high biomass than the ones grown in autumn. The mean highest yield of plants was found after the application of $Mg(NO_3)_2$, the lowest—when $Ca(NO_3)_2$ was used. For the pot cultivation system, the highest pigment contents (chlorophylls and carotenoids) were obtained in the samples treated with urea at the N-90 dose which correlated with the Mn level in the plants. In the hydroponic systems, the highest DPPH activity was observed after treatment with sodium nitrate at the N-70 and N-90 doses. The highest yield (182 g of fresh matter·plant^{-1}) in the pot cultivation was found at N-90 with the simultaneous Li spray treatment. The SA foliar spray treatment significantly influenced the plants' quality and could be classified properly according to the antioxidant activity, chlorophyll content, carotenoid content, lightness, yellowness, and the N level.

The obtained results confirmed the research hypotheses. It makes an important contribution to the development of knowledge in the field of plant nutrition, as well as the biological value of crops.

Keywords: antioxidant ability, chlorophyll content, colour, flowering Chinese cabbage, foliar spray, nitrogen fertiliser level, nitrogen source, phenolic content; plant nutrition, yield

Streszczenie

Kapusta chińska kwitnąca (*Brassica campestris* L. *ssp. chinensis var. utilis* Tsen et Lee) jest popularnym warzywem liściowym pochodzącym z Chin i rozpowszechnionym w południowo-wschodniej Azji. Jej krótki okres wegetacji skutkuje wysokimi wartościami wskaźnika wielokrotnej uprawy, co oznacza, że na tym samym polu można uzyskać większe plony całkowite. Kapusta chińska kwitnąca nie jest jednak jeszcze powszechnie i komercyjnie uprawiana w Europie. Roślina ta jest bogata w związki bioaktywne.

Celem badań była ocena wpływu wybranych czynników na plonowanie i jakość kapusty chińskiej kwitnącej. W niniejszej dysertacji postawiono trzy hipotezy badawcze:

(I) Modyfikacja żywienia roślin azotem, zarówno jego intensywnością, jak i źródłem składnika, wpływa na plonowanie kapusty chińskiej kwitnącej, (II) Dolistne traktowanie kwasem salicylowym, V, Li, Se i Se wpływa na plonowanie kapusty chińskiej kwitnącej, (III) Kapusta chińska kwitnąca wykazuje przydatność do uprawy bezglebowej, zarówno w podłożu (mieszanina gleby mineralnej + torfu; uprawa doniczkowa), jak i w systemie hydroponicznym.

Badania przeprowadzono w latach 2018-2020. Plonowanie i parametry fizykochemiczne mierzono/badano na podstawie świeżej masy roślin, zawartości składników, koloru, zawartości chlorofilu, zawartości karotenoidów, całkowitej zawartości fenoli, całkowitej zawartości flawonoidów i aktywności przeciwutleniającej. Każde doświadczenie przeprowadzono w 2 niezależnych systemach uprawy (doniczkowym i hydroponicznym).

Wykazano, że zwiększenie intensywności żywienia azotem pozytywnie wpływa na plonowanie roślin, zarówno w systemie uprawy doniczkowej, jak i hydroponicznej. Plonowanie wzrastało wraz ze wzrostem poziomu azotu od N-50 do N-90. Generalizując, optymalnym poziomem żywienia azotem był poziom N-90. W uprawie w podłożu zawartość N w roślinie wzrastała wraz ze wzrostem intensywności żywienia tym składnikiem, podczas gdy w uprawie hydroponicznej zawartość N w roślinie była stabilna. Próbki wiosenne charakteryzowały się większymi wartościami aktywności przeciwutleniającej i zawartości fenoli niż próbki jesienne. Podczas jesiennej uprawy w podłożu zawartość N w roślinach była o około 60% większa w porównaniu z roślinami wiosennymi. Rośliny uprawiane wiosną uzyskiwały większą biomasę niż te uprawiane jesienią. Średnio największe plonowanie roślin uzyskano po zastosowaniu $Mg(NO_3)_2$, a najmniejsze po zastosowaniu $Ca(NO_3)_2$. W przypadku systemu uprawy doniczkowej największą zawartość barwników (chlorofili i karotenoidów) uzyskano w próbkach traktowanych mocznikiem przy poziomie N-90, co korelowało z poziomem

Mn w roślinach. W systemie hydroponicznym największą aktywność DPPH zaobserwowano po traktowaniu azotanem sodu w stężeniach N-70 i N-90. Największe plonowanie (182 g świeżej masy·roślina^{-1}) w uprawie doniczkowej uzyskano przy N-90 z jednoczesnym opryskiwaniem Li. Opryskiwanie dolistne SA znacząco wpłynęło na jakość roślin i można je było odpowiednio sklasyfikować według aktywności przeciwutleniającej, zawartości chlorofilu, zawartości karotenoidów, jasności, zażółcenia i poziomu N.

Uzyskane wyniki potwierdziły postawione hipotezy badawcze. Stanowi to istotny wkład w rozwój wiedzy z zakresu żywienia roślin, a także wartości biologicznej upraw.

Słowa kluczowe: zdolność antyoksydacyjna, zawartość chlorofilu, barwa, kapusta chińska kwitnąca, opryskiwanie dolistne, poziom żywienia azotem, źródło azotu, zawartość fenoli; żywienie roślin, plonowanie

CHAPTER I Introduction

Flowering Chinese cabbage (*Brassica campestris L.* ssp. *chinensis var. utilis Tsen et Lee*), also called “Caixin” in Mandarin Chinese or “Choy sum” in Cantonese dialect, originated in China and it is popular in other Asian countries, such as Japan and Vietnam (G. Li et al., 2011). However, this vegetable is still little known in Europe. This vegetable belongs to the *Brassica* genus. The leaves and flowering stem are the valuable part of flowering Chinese cabbage, which is not only a valuable source of glucosinolates, polyphenols, vitamin C, but also rich in amino acids and other chemical compounds (Yuan et al., 2019). The studies of plant fertiliser or nutrition on flowering Chinese cabbage are quite limited (Ji et al., 2020; Xie et al., 2011; Yuan et al., 2019). Thus, it is necessary to explore the optimal condition of flowering Chinese cabbage in greenhouse. Furthermore, the research could also develop a theory on exploring an optimal condition for a vegetable that grows in a new condition.

Nitrogen (N) fertiliser plays an important role in enhancing the yields of plants by affecting photosynthetic rates, growth rates, and plant productivity in most ecosystems (Din et al., 2021; Rehman et al., 2020; Yuan-Yuan et al., 2021). Not only can it increase the volume of yields, but it also modifies the quality of the plants. This is because N nutrition is correlated with the plants’ biochemical and physiological functions and is an essential constituent of protein, nucleic acids, chlorophylls, and growth hormones (Din et al., 2021; Rehman et al., 2020). Insufficient nitrogen application might decrease leaf N concentration, chlorophyll content, and photosynthetic ability (Boussadia et al., 2010; Gan et al., 2016; Yuan-Yuan et al., 2021). However, excessive plant nutrition with nitrogen can have negative effects such as inhibiting growth, and suppressing other physiological processes, and potentially could be lethal for the plants (Kong et al., 2017; Xie et al., 2011). Therefore, the determination of the optimal nitrogen application is one of the key factors to improve the yield and some biochemical properties of flowering Chinese cabbage.

Nitrogen fertilisers in various forms are widely used in plant production. One of the most popular is ammonium nitrate (NH_4NO_3, 34% N). Urea (46 % N) is the most concentrated solid nitrogen fertiliser. Sodium nitrate (16% N, 26% Na), calcium nitrate (15.5% N) and magnesium nitrate (10.5% N) are all metal ions of nitrogen. Calcium and magnesium are essential nutrients for promoting plant growth They both play important roles in plant photosynthesis, carbohydrate transport, nucleic acid, and protein synthesis, which further influences the quality of plants (Hariadi & S Shabala, 2004; Hauer-Jákli & Tränkner, 2019; Liu et al., 2022). Some studies focus on the effect of different nitrogen fertilisers on horticultural/agricultural plants, such as radish

(Yousaf et al., 2021), maize and wheat (H. Li et al., 2021), Chinese cabbage (*Brassica rapa* L. *Pekinensiss*) (Lu et al., 2021), navel orange fruit (Liu et al., 2022), potato (Elrys et al., 2018), tomato (Peyvast et al., 2009), etc. Nevertheless, there is still a lack of references for studying varied nitrogen in flowering Chinese cabbage.

Foliar treatment has several advantages in comparison to other methods of nutrient applications, such as reduction of soil fertilization and improvement of soil environment, increase the efficiency of nutrient application and yielding, and prevent soil salinization by using less amount of fertiliser (Niu et al., 2020). In this study, five foliar spray treatments were compared, i.e., selenium (Se), silicon (Si), lithium (Li), vanadium (V), and salicylic acid (SA), which represent three types of foliar treatments: metalloid nutrients (Se, Si), metal nutrients (Li, V), and plant hormone (SA). Based on the literature data, these treatments affect effects on the plant growth and yield. Selenium stimulates plant growth, regulates their antioxidative defense system (enzymatic and non-enzymatic), and thus reduces their susceptibility to various abiotic stresses induced by UV irradiation, drought or heavy metal concentration (Dong et al., 2013; H. Huang et al., 2021; Sattar et al., 2019). Se and Si synergistically alleviated the toxicity of cadmium in the shoots of rice plants by transferring more Cd distributed in the cell wall and organelles (H. Huang et al., 2021). Silicon used in the nutrient solution did not affect the yield and colour of vegetables, but species-related accumulation of Si was found (D'Imperio et al., 2016). Li and V can be very harmful for humans, cause impulsiveness and aggressiveness, lead to homicides and suicides (Li), disorder the thyroid and renal functions, induce diarrhea, cause haematological disturbances, and even lead to death (V). However, in small doses they can also stimulate plant growth (Li and V) and metabolism (Li) (Aihemaiti et al., 2020; Jiang et al., 2019). SA is a hormone that is naturally synthesized by plants in minimal amounts to regulate their growth, flowering, and photosynthesis (Hayat et al., 2010). However, there is lack of data on the effect of Se, Si, Li, V or SA foliar treatments on yield and quality, both in quantitative and qualitative aspects of flowering Chinese cabbage.

This dissertation presents a three-year study on cultivating flowering Chinese cabbage in two independent systems of cultivation, namely pot and hydroponic systems. The study was conducted between 2018 and 2020 year focusing on investigation the effects of different nitrogen levels, nitrogen sources, and foliar treatments on crop yield and physicochemical parameters of plants.

In 2018, the effects of different intensity of nitrogen nutrition and planting date (spring and autumn) on the plant yielding were studied. In 2019, the effects of five different nitrogen sources and the levels of N nutrition were analysed, and in 2020, the effects of five foliar treatments under varied N nutrition were investigated. In each year, the plant yielding was evaluated, both in terms of quantity (fresh weight) and quality (nutrient content, colour, chlorophyll content, flavonoid and phenolic content, as well as antioxidant ability) under these combinations of nutrition. Each experiment was conducted in 2 independent systems of cultivation (pots or hydroponic).

CHAPTER II Literature review

1. Historical background

During the 15th and 16th centuries, a wide-ranging exchange of plants, animals, human populations, technology, culture, and diseases occurred between America and the Old World, known as the Columbian Exchange (Crosby, 1972). One of the most significant effects of the Columbian Exchange was expanding agricultural species and changing the global populations of organisms. Before the 15th century, potatoes were only grown in South America. However, by the 19th century, they were widely cultivated and consumed in North America, Europe, and even India. It was estimated that the population of Afro-Eurasia increased by 25% during the 17th to 19th centuries inter alia due to the introduction of new crops (Nunn & Qian, 2011). Tomatoes, which were brought to Europe by the Spanish, became a staple of Italian cuisine. Rice and sugarcane were also imported to America. Thousands of species were introduced to new continents, significantly affecting agriculture.

While the Columbian Exchange was undoubtedly a milestone, the transfer of plants to new locations has been a constant throughout human history, from ancient times to the present and will continue in the future. In terms of basic evolutionary criteria, wheat is perhaps the most successful plant in history. Wheat was domesticated in the Middle East around 9000 BC. Due to geographic convenience in Afro-Eurasia, it only took a few millennia for wheat to spread across the continents, and it was later imported to America during the Columbian Exchange (Harari, 2015).

Chinese cabbage (*Brassica pekinensis* L.) perhaps is the most well-known oriental vegetable in Europe and America. It originated in the temperate climate area in central China (C. W. Li, 1981) and was introduced to Korea about 400 years ago and to Japan around 150 years ago (Ramchiary et al., 2011; Su et al., 2018; Webb et al., 2003). By 1929, there was already large seed house producing Chinese cabbage seeds (Chung & Ripperton, 1929). In 1995, Balvoll (1995) demonstrated the possibility of growing Chinese cabbage in Norway. Not only Chinese cabbage, but also bok choi *(Brassica rapa* subsp. *chinensis)*, kai choi (*Brassica oleracea* var. *alboglabra juncea*), edamame (*Glycine max* L. *Merr)*, Japanese murasaki sweet potato (*Ipomoea batatas* L.), Okinawa sweet potato (*Ipomoea batatas cv. 'Ayamurasaki'*) and flowering Chinese cabbage (*Brassica rapa* var. *parachinensis)* are beginning to be produced in Europe (Hong & Gruda, 2020). Therefore, the species exchange among different regions never ends.

2. Diversity in the vegetable market

Mansfeld's Encyclopedia of Agricultural and Horticultural Crops identifies 1097 types of vegetables, but only 392 of these are cultivated and consumed (Silva Dias, 2010). In Poland, there are about 40 commercially cultivated vegetable species, but only seven of them play a leading role in vegetable production: cabbage, onions, red beets, tomatoes, cucumbers, and cauliflowers (Główny Urząd Statystyczny, 2022). In 1993, the total area of the field cultivated vegetables has reached 270 thousand hectares, of which the highest total cultivation areas were for cabbage (55.1 thousand ha), cucumbers (34 thousand ha), onions (33.5 thousand ha), carrots (32.5 thousand ha), and tomatoes (29.9 thousand ha) (Kaniszewski & Cieslak-Wojtaszek, 1993 (Table 1). However, with the time changing, according to the Statistic Yearbook of Agriculture (2022) the area of different vegetable cultivation had changed in 2021, with cabbages (14.6 thousand ha), onions (22.3 thousand ha), carrots (17.5 thousand ha), beetroot (7.4 thousand ha), cucumbers (5.8 thousand ha), and tomatoes (5.9 thousand ha) out of a total cultivation area of 164.3 thousand ha (Główny Urząd Statystyczny, 2022). The cultivation of the aforementioned vegetables accounted for 58% of total vegetable cultivation. Unfortunately, the statistical book did not list of the remaining vegetables' cultivation area. According to the UN Food and Agriculture Organization (FAO), bok choy had only 899 tons of production in Poland in 2018. In comparison, more than 900 thousand tons of cabbage had a production per year in Poland, indicating that the production of oriental vegetables has potential to increase.

Table 1 Cultivated area of field by main vegetables in Poland 2022 (thousand ha).

	Cabbages	Onions	Carrots	Beetroot	Cucumber	Tomatoes
1993	55.1	33.5	32.5	/	34	29.9
2010	20.5	23.7	19.6	8.1	13.5	8.3
2015	23.5	25	22.1	10.9	15	10.6
2016	20.1	26.5	22.3	9.9	13.7	9.1
2017	20.5	26	22.1	10	13.9	9.3
2018	20.7	24.8	22.4	10.2	14.6	9.7
2019	20.9	24.8	22.2	10.4	15.1	10.0
2020	13.7	23.1	17.7	7.5	5.5	6.3
2021	14.6	23.4	17.5	7.4	5.8	5.9

Source: Główny Urząd Statystyczny (2022)

Despite living in a globalized, modern society, significant differences in vegetable production still exist worldwide. In Asia, countries like China, India, and Southeast Asian countries produce over 200 vegetable species. However, Germany only produces a maximum of 60 different vegetable types, and Poland only produces 40 (Hong &

Gruda, 2020; Kaniszewski & Cieslak-Wojtaszek, 1993). While the situation might be better in countries like Spain and Italy, more research is needed to confirm this. Overall, there are many opportunities to improve the diversity of the vegetable market in Europe.

From another perspective, traditional vegetables are those that have been popular in a particular region for a long time, as opposed to global vegetables like cabbage and tomatoes. Thus, it is important to focus on traditional vegetables, to preserve the diversity of horticulture resources. Some original African and Asian-Pacific vegetables have received notable attention from researchers (Hanelt et al., 2001).

Preserving traditionally grown vegetables offers a significant advantage for future diversity in nutrition-sensitive agriculture compared to global vegetables (Meldrum et al., 2018). Some traditional vegetables have the potential to be introduced to other countries, but lack of information and poor conservation status limit their utilization. Therefore, studies that focus on traditional vegetables are essential and valuable.

3. Flowering Chinese cabbage

Flowering Chinese cabbage (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee), also known as ‘Caixin (菜心)’ in Mandarin Chinese, or ‘*Choy sum*’ in the Cantonese dialect, is originally from the southern areas of China but is now popular throughout the country. In the thesis, the name flowering Chinese cabbage is used to refer to this type of vegetable. Due to the huge climatic differences between the north and south of China, many varieties have been developed to adapt to local conditions (Z. Wang et al., 2019).

The purple variety is one of the famous varieties grown in the Yangzi River basin in particular. It was Chinese migrants moving to new places who spread the blooming Chinese cabbage in Asia. Nowadays, it is not only the southern provinces of China that are famous for cabbage cultivation, but it is also grown in Southeast Asian countries (Issarakraisila et al., 2007).

Guangdong province is the largest growing area and the largest supplier of vegetables, including flowering Chinese cabbage. By 2002, the cultivation area had reached 200 ha in Guangzhou city alone, accounting for 60% of all vegetable plantations. The entire Guangdong province exported 700 tons of vegetables to Hong Kong per day, 70% of which was flowering Chinese cabbage (G. Li et al., 2011).

The optimal temperature for the cultivation of flowering Chinese cabbage for leaves is 20-25°C, and for the flower stalk formation is 15-20°C (S. Song et al., 2012). Vegetation period takes 32-35 days from sowing to harvest and 80-90 days from seeds to mature. The main advantage of cabbage cultivation is a short growing season, which allows multiple harvests during year (Peng et al., 2015).

3.1 Varieties of flowering Chinese cabbage

In accordance with growth time, flowering Chinese cabbage is distinguished to three types – early, middle and late types, for example, 'Youlü Cutai Caixin' and 'Youlu (油绿) 501 Caixin' are early maturity cultivars; 'Hongcaitai（红菜薹）is type of late cultivar (G. Li et al., 2010). According to the colour of leaves and stem, flowering Chinese cabbage is divided to green type and purple type. The green type is commonly grown in the south of China and Southeast Asia, such as Guangdong. Due to mild winters in those regions, flowering Chinese cabbage grows throughout whole year. The purple flowering Chinese cabbage is grown in the provinces north to Guangdong province, e.g., in Sichuan, Hubei and Wuhan (Górna et al., 2016; Y. Wang et al., 2014). For example, 'Xianghongcaitai（湘红菜薹）No. 1', which is suitable for the Yangtze River basin, is cultivated in summer-autumn and early autumn-winter.

There is enormous difference in temperature between the north and south of China, which become the main challenge for flowering Chinese cabbage spreading. G. Li et al., (2012) measured the heat tolerance of six flowering Chinese cabbage cultivars at semi-lethal temperature (LT50) and the survival rate of seeds. The results showed that the heat tolerance of the six cultivars decreased in the following order: 'Sijiu-19 Caixin（四九菜心）', 'Sijiu Huang Caixin（四九黄菜心）', 'Youlv 50 Tian Caixin（绿油 50 天菜心）', 'Youlv 501 Caixin', 'Youlv 701 Caixin', 'Teqing Chixin 4（特青迟心）'. There was a significant positive correlation with the survival rate of seeds during high stress in the field. It could be used as the flowering Chinese cabbage heat resistance evaluation index and proved the possibility to obtain cultivars for variable climate area.

3.2 Plant nutrition

Literature data on nutrition is relatively scarce. Biofertiliser, which was a mixture of Trichoderma (T.): T. harzianum, T. asperellum, T. hamatum, and T. atroviride at a spore concentration of 2.56 mg·109 ml, could improve the quality and production of flowering Chinese cabbage by enhancing the tolerance of flowering Chinese cabbage to environmental stresses, furthermore, adjusting the soil environment (Ji et al., 2020). 5 kg of refined organic fertiliser increased the yield of two flowering Chinese cabbage cultivars 'Meiqing no. 1' and 'Baoqing 40 days' by 29.1-40.6% (Jian et al., 2004). The foliar application of amino acid, especially glycin (Gly2) at a dose of 200 mg kg^{-1} , in cultivation of flowering Chinese cabbage could improve the yield and reduce the content of nitrate in plant tissues by about 85.03% (Junxi et al., 2010). When flowering Chinese cabbage was treated with a microbial fertiliser (total N 3.67%) and a manure fertiliser (mixed with chicken ordure and pig ordure), the yield ranged from 85.1% to 106.7% compared to the yield of the control group, which had been treated with the same level of inorganic nitrogen. The microbial fertiliser may indeed have elevated the height of Chinese flowering cabbage plants (Jin et al., 2011). The application of fluid fertilisers (consisting of urea, phosphoric acid and potassium chloride) at a dose of 599.7 L per ha in soil cultivation, was the most effective in improving plant yield which accounted for 9.3% compared with solid fertiliser treatment (Jun-hong et al., 2009).

The application of 110.0 t ha^{-1} organic fertiliser combined with 8.5 t ha^{-1} corn cobs biochar was the optimal amount for the yield and N uptake of flowering Chinese cabbage (Z. Wang et al., 2019). Xiahui et al. (2006) stated that the yield of flowering Chinese cabbage significantly increased (by 19.1% - 28.2%) after application of controlled-release nitrogen fertilisers CRNFs (containing 30% of N; coating with four types of materials, which were named by PDU, PHU, PS_2U, PS_5U, and developed by the Research Centre of New Fertilizer and Resource of South China Agriculture University) compared to same nitrogen level of urea. Q. Chen et al. (2010) indicated that the optimal range of phosphate fertiliser content in latosolic red soils in Guangdong was 54.6-87.8 mg kg^{-1}. They further recommended P_2O_5 application doses of 50.0-76.5 kg ha^{-1} for achieving optimal yields of 6441-6515 kg ha^{-1}, 50.0-72.6 kg ha^{-1} for yields of 6542-6693 kg ha^{-1}, and 50.0-64.1 kg ha^{-1} for yields of 6880-7189 kg ha^{-1}.

3.3 Composition of flowering Chinese cabbage

Brassica genus is 1 of the 10 most economically important vegetables in the global agriculture and markets. Flowering Chinese cabbage belongs to the *Brassica* genus and it is similar to other *Brassica* vegetables containing a variety of nutrients and phytochemicals that may work to prevent cancer, cardiovascular, obesity, and certain diseases and enhance the immune system (Cartea et al., 2011; Francisco et al., 2017; van Poppel et al., 1999).

The edible parts of flowering Chinese cabbage are the leaves with young flower stalks. Therefore, the size of the stalk, leaf stems, and inflorescences are important morphological characteristics of the economic value of flowering Chinese cabbage (X. Huang et al., 2017). Every 100 g of the edible part of flowering Chinese cabbage has 89 kJ energy. Flowering Chinese cabbage has the following chemical composition of elements (in mg 100 g^{-1}): N 511.7; K 55.88; P 49.16; Ca 86.18; Mg 8.593; Na 35.67; Fe 2.882; Zn 1.088; Mn 0.334; Cu 0.186; Ni 0.021; Pb 0.119; Cd 0.002; Cr 0.008 (Academic, 2010). The flowering Chinese cabbage has also high ability to accumulate Se (Mo et al., 2006). It may be used as Se supplement production.

Flowering Chinese cabbage has special fragrance and major factors that contributed to its aroma are the volatile flavour substances such as esters, alcohols and phenols (Yuan et al., 2019). Like other *Brassica* genus vegetables, flowering Chinese cabbage is a valuable source of GSLs, polyphenols, phenolic acids, vitamin C, and some amino acids and other chemical compounds (Yuan et al., 2019). More than 70 phenolic compounds were tentatively identified in the plant, including flavonoids such as kaempferol, isorhamnetin and quercetin glycosides (mainly glucosides) as well as phenolic acids such as caffeic, ferulic, sinapic acids and their derivatives (Junxi et al., 2010). The flowering Chinese cabbage which grew in southern China had higher total phenolic content (TPC) 1.39 mg g^{-1} compared with plants grown in Singapore with TPC equal to 0.68 mg g^{-1} (Isabelle et al., 2010).

Glucosinolates (GSLs) are biologically active compounds found in the Brassicaceae family of plants, including broccoli, cabbage, cauliflower, rapeseed, and mustard. This compound has been studied extensively in recent years due to its beneficial effects, such as regulatory functions in inflammation, stress response, phase metabolism, antioxidant activities, and direct antimicrobial properties (Bischoff, 2016). There are 16 families of plants that can synthesize GSLs, but the genus Brassica includes the highest number of the edible species that contain GSLs (Fahey et al., 2001; Rosen et al., 2005). Due to GSLs being a class of nitrogen and sulfur containing compounds, the concentration of GSLs in cabbage grown on a low nitrogen and sulfur soil can be modified by plant nutrition (Rosen et al., 2005). The total GSLs content of green flowering Chinese cabbage is around 14-35 mg 100 g^{-1}. For comparison, the purple flowering Chinese cabbage contains 50-70 mg 100 g^{-1} (X. Chen et al., 2008). The highest content of GSLs was found in the inflorescences of flowering Chinese cabbage then followed by stems and leaves (respectively 569.32, 153.13 and 45.38 μmol per 100 g FW) (H. He et al., 2000).

3.4 Antioxidant activity

Brassicaceae vegetables are the largest and most widely consumed a group of plants in Europe and it contains natural source of antioxidants, such as phenolics which was discussed above. The antioxidants can exert their properties by different mechanisms, including radical scavenging capacity, metal chelating ability, inhibition of prooxidant enzymes and stimulation of the antioxidant enzymes as well as regeneration of other antioxidants (Floegel et al., 2011; Olarewaju et al., 2018). Thus, measuring the antioxidant activity of the compounds various methods should be used (Frankel & Meyer, 2000). The TEAC (Trolox equivalent antioxidant capacity), DPPH (2,2-diphenyl-1-picrylhydrazyl free radical) and FRAP (Ferric ion reducing antioxidant power) assays are the most often used methods to determine the antioxidant capacity of fruits and vegetables.

The antioxidant ability of flowering Chinese cabbage can be influenced by many factors, including cultivation conditions, climate, geographical region, degree of ripeness, part of plant etc. Wachtel-Galor, Wong, and Benzie (2008) stated the fresh flowering Chinese cabbage has higher antioxidant ability than broccoli, cabbage, cauliflower, with FRAP values of 5.634, 3.911, 3.480 and 2.768 μmol g^{-1}, respectively. Q. He et al., (2016b) pointed out that the antioxidant ability of the inner leaves of purple heading Chinese cabbage reached 20.239 μM TE g^{-1} FW (Trolox equivalents per fresh weight by gram) in the ORAC (The oxygen radical absorbance capacity) assay, 16.728 μM TE g^{-1} FW in the FRAP assay, and 6.667 μM TE g^{-1} FW in the DPPH assay, in comparison to external leaves showing much lower antioxidant activity, ranging from 8.894 to 11.259 μM TE g^{-1} FW in the ORAC assay, 4.964 to 7.214 μM TE g^{-1} FW in the FRAP assay, and 1.743 to 2.896 μM TE g^{-1} FW in the DPPH assay. The antioxidant activity of the plant is positively correlated with the phenolic compound content (Lee et al. 2018).

Table 2 The comparison of antioxidant ability and total phenolic content among Brassica vegetables analysed by different methods (according to various authors). (Deng et al. 2013; Wiczkowski, Szawara-Nowak, and Topolska 2013; Ciska, Karamac, and Kosiñska 2005; Isabelle et al. 2010; Ferreres et al. 2006; Q. He, Zhang, and Zhang 2016).

Name	**Latin name**	**TPC (mg GAE/g FW)**	**FRAP values (µmol /g FW)**	**ABTS values (µmol Trolox/g FW)**	**DPPH values (µmol/g FW)**	**References**
flowering Chinese cabbage	*B. campestris L. ssp. chinensis var. parachinensis*	8.60±0.18	12.01±0.82	16.93±0.38		Deng et al 2012
Cabbage	*Brassica oleracea* var. *capitata* L.	6.24±0.05	5.74±0.19	8.24±0.28		
Broccoli	*Brassica oleracea* var. *italica*	9.84±0.17	13.45±0.49	13.97±0.17		
Leaf mustard	*Brassica juncea*	8.26±0.15	8.28±0.58	15.23±0.69		
Pakchoi	*Brassica chinensis* L.	7.45±0.16	18.29±0.38	15.60±0.69		
Purple heading chinese cabbage internal leaves	*Brassica rapa pekinensis*. sp.		16.728		6.667 µM TE/G FW	He et al. 2016
Purple heading chinese cabbage external leaves			4.964-7.214		1.743-2.896 µM TE/G FW	
Tronchuda cabbage internal leaves	*Brassica oleracea L. var. costata DC*	1.39			1192 µg/ml (IC_{25})	Ferreres et al. 2006

Tronchuda cabbage external leaves		13.34	440 µg/ml (IC_{25})	
flowering Chinese cabbage grown in southern China	*B. campestris L. ssp. chinensis var. parachinensis*	1.39		Isabelle et al. 2010
flowering Chinese cabbage grown in Singapore		0.68		
Red cabbage	*Brassica oleracea var. capitata L. (f. Rubra)*	1.86		
			86.51± 2.89	Wiczkowski et al. 2013
Cabbage	*Brassica oleracea* var. *capitata* L.	5.72		Ciska et al. 2005

4. Nitrogen fertiliser

Humans have a long history of using natural fertilisers, but chemical fertiliser elements were discovered only two hundred years ago. Industrialized fertiliser production began even later, in the 19th century (Russel & Williams, 1977). As one of the earliest essential nutrients discovered and one of the most crucial nutrient elements in high demand, nitrogen fertiliser is vital in promoting plant growth and enhancing the quality of vegetables. Takahashi et al., (2018) pointed out that moisture management can enhance nitrogen nutrient uptake and relative growth rate of cabbage, which is highly correlated to nitrogen concentration. Jahan et al. (2020) was found that the optimal nitrogen (N) nutrition for maximizing the yield of cv. BRRI dhan58 was 142 kg ha^{-1}, while for cv. BRRI dhan75, it was 82 kg ha^{-1}. Vinale et al. (2023) studied the effect of different nitrogen concentrations (0, 4.2, 8.4, and 12.6 mg dm^{-3}) on Mexican marigold (*Tagetes erecta* L. var. Inca), and the results showed the different effect on the concentrations of photosynthetic pigments and biomolecules with antioxidant capacity depending on the different nitrogen nutrient level. Nitrogen nutrition also increased broccoli yield and modified the plant's quality (Babik & Elkner, 2000). The plant's response on the application of N nutrition could be vary depending on geographical regions and plant species. For example, in Odemis of Turkey, Yoldas et al. (2008) found that the highest total yield of broccoli (34.63 tons ha^{-1}) was obtained at nitrogen fertiliser dose of 300 kg N ha^{-1}. However, in Ontario of Canada, Westerveld et al. (2002) proved that N rates between 220 and 260 kg ha^{-1} appear to be beneficial for summer cabbage on the yielding of plants (total and marketable yield, weight per head, head density, and head size).

One widely used fertiliser for top-dressing is ammonium nitrate, which contains 33.5-34.5% N. It has advantages over other nitrogen fertilisers due to the availability of half the nitrogen as nitrate (NO_3^-). It is sold in a prilled or granular form to prevent moisture absorption. The application of these forms of nitrogen can positively influence on quality of yielding (Boschiero et al., 2020). Zhu et al. (2021) suggested that appropriate N-NH_4/N-NO_3 ratios may enhance N absorption and assimilation, leading to improved growth of flowering Chinese cabbage.

The most concentrated solid nitrogen fertiliser available is urea (46% N). However, when urea is added to soil, it can change into ammonium carbonate, causing a temporary increase in local pH levels that may be harmful. Xie et al. (2011) experimented on flowering Chinese cabbage, which revealed a tendency for increased yields at the beginning using urea fertiliser. However, excessive use of urea inhibited growth. Mentioned authors recommend as an optimal dose of 440-490 kg ha^{-1}. Excessive application of urea may decrease the plant quality, e.g., lead to the accumulation of nitrates in plants (Elrys, Abdo, and Desoky (2018) and Kim et al. (2023)).

Sodium, calcium, and magnesium play important roles in various plant processes, including photosynthesis, carbohydrate transport, nucleic acid and protein synthesis, and even plant quality (Hariadi & S Shabala, 2004; Hauer-Jákli & Tränkner, 2019; Liu et al., 2022). Cabbages are known to accumulate free calcium ions, and the balance between ions in the plant and soil is maintained during uptake (Turan & Sevimli, 2010; White & Broadley, 2003). Magnesium fertiliser has been shown to improve growth, yield, and nutrient content in radish, black tea, navel oranges, and Chinese cabbage (Jayaganesh et al., 2011; Liu et al., 2022; Lu et al., 2021; Yousaf et al., 2021). The recommended level of magnesium fertilization varies depending on the plant species and growing conditions. An example dose of 30 kg Mg ha^{-1} is recommended for rape seed plants, meanwhile, the doses 142-177 g MgO $plant^{-1}$ could be recommended for the nutrition of navel orange fruit (Geng et al., 2021; Liu et al., 2022).

5. Trace elements and plant hormone

Some nutrients are essential for plant growth, which means the deficit nutrient status with these elements would cause the plant's death before it completes the life cycle. However, some trace elements are not essential, but they can affect plant growth. In this part, the trace elements like lithium (Li), selenium (Se), silicon (Si) and vanadium (V) have been reviewed.

Selenium, plays a beneficial role in plants by positively influencing their growth, regulating the antioxidative defense system, and reducing plant susceptibility to various stresses, such as heavy metals, salinity or drought (Dong et al., 2013; H. Huang et al., 2021; Nawaz et al., 2015). Nawaz et al. (2015) found that Se foliar spray on wheat subjected to drought stress improved plant yield, nutrient uptake, and the activity of the antioxidant enzyme system. Additionally, Se foliar spray increased chlorophyll and carotenoid content in leaves of *Lycium chinense* L. (Dong et al., 2013) and *Spinacia oleracea* L. cv. 'Missouri' (Saffaryazdi et al., 2012). Furthermore, Se and Si have a synergistic effect in alleviating cadmium toxicity in rice plants (H. Huang et al., 2021). The simultaneous application of Se and Si significantly increased the efficiency of the GSH-As cycle (Glutathione-ascorbate cycle) and the concentration of glutathione and ascorbate, as well as the activities of glutathione reductase and dehydroascorbate reductase in leaves and roots of flowering Chinese cabbage (Wu et al., 2017). Si treatment significantly improved the yield of pakchoi (*Brassica chinensis* L.) (B. Wang et al., 2020) and was also used to obtain Si biofortification in leafy vegetables (D'Imperio et al., 2016).

On the other hand, the metal treatments of Li and V show both beneficial properties and toxicity to plants, depending on the concentration of the metal in soil, pH, redox status of the soil, and type of plant (Aihemaiti et al., 2020; Jiang et al., 2019). Although Li and V could harm human health, they could positively influence plant growth and metabolism (Aihemaiti et al., 2020; Jiang et al., 2019). Li foliar spraying was an effective strategy for biofortifying grape berries in lithium, especially in the skin (Zhao et al., 2020). Li fertiliser increased the yield of Luobuma tea (*Apocynum venetum*) without reducing the content of total flavonoid content and antioxidant activity (Jiang et al., 2019). However, Li could also reduce plant growth by interrupting numerous physiological processes and altering metabolism (da Silva et al., 2019; Shahzad et al., 2016). Vanadium application via foliar spray significantly increased the height of sugarcane (Sentíes-Herrera et al., 2018). Nevertheless, in hydroponic cultivation of tobacco (*Nicotiana tabacum* L.) of V application at a concentration over 2.0 mg dm^{-3}, exhibited a relatively suitable V tolerance, but the growth was inhibited (Wu et al., 2021).

Salicylic acid (SA) is a type of plant hormone that regulates plant growth, flowering, and photosynthesis (Hayat et al., 2010). Exogenous SA foliar spraying could give a similar function to plants. Fariduddin, Hayat, and Ahmad (2003) reported that a lower concentration of SA foliar spray substantially enhanced the dry matter of *Brassica juncea,* but a higher SA level (10^{-5} M, 10^{-4} M, 10^{-3} M, 0) had an adverse, inhibitory effect.

CHAPTER III Hypothesis and research objectives

In my dissertation, I set out the following hypotheses.

1. Modifying plant nutrition with nitrogen, both its intensity and source of the nutrient, influence the yielding of flowering Chinese cabbage.
2. Foliar application with salicylic acid, V, Li, Se and Se influences the yielding of flowering Chinese cabbage.
3. Flowering Chinese cabbage shows suitability for soilless cultivation, both in substrate (mixture of mineral soil + peat; pot cultivation) and in hydroponic system.

Research Aims:

The primary aim of the studies was to determine the optimal nitrogen level, cultivation season (planting date), and cultivation system (pot or hydroponic) to maximize the growth and yield of flowering Chinese cabbage. Subsequently, based on the identified optimal conditions (N-50, N-70, N-90, N-110, N-130), the effects of different nitrogen fertilisers (NH_4NO_3, $Ca(NO_3)_2$, $Mg(NO_3)_2$, $NaNO_3$, and urea) on the growth and physicochemical parameters of flowering Chinese cabbage were investigated. The next aim was to examine the influence of different foliar treatments (Se, Si, Li, V, and SA) on the growth and physicochemical parameters of flowering Chinese cabbage.

It was expected the results could provide valuable insights into the effect of nitrogen nutrition, cultivation practices, and foliar treatments on the growth, yield, and quality of flowering Chinese cabbage.

CHAPTER IV Material and methods

To introduce the methodology of these studies, it is important to provide an overview firstly. The methodology can be divided into two main parts, conducted in 2 departments of Poznan University of Life Sciences (PULS).

Detailed descriptions of the research methods used to achieve the research objectives of the described self-reference are presented in the chapters of the individual publications titled "Materials and Methods."

In the Department of Plant Nutrition (actual Department of Plant Physiology, Laboratory of Plant Nutrition), four independent cycles of vegetation experiments were carried out over a period of three years, which included both pot cultivation (mixture of peat and sand) and hydroponic cultivation of flowering Chinese cabbage.

In year 2018, the yield and quality of flowering Chinese cabbage were evaluated under different nitrogen nutrition levels (ranging from 50 to 130 mg N per dm^3) in two varied growing media ((I) pot cultivation: mixture mineral soil and peat; and (II) hydroponic). The studies were conducted in two seasons (spring and autumn).

In year 2019, the study was to assess the influence of different nitrogen sources, including NH_4NO_3, $Ca(NO_3)_2$, $Mg(NO_3)_2$, $NaNO_3$, and urea, and varying levels of nitrogen nutrition (50, 70, and 90 mg N per dm^3) on the yield quantity and quality of flowering Chinese cabbage grown in two soilless systems, pot cultivation (mixture of peat and sand) and hydroponic culture.

In year 2020, the influence of varying nitrogen nutrition levels (70 and 90 mg N per dm^3) and foliar treatments (Se, Si, Li, V, and SA—salicylic acid) on the yield quantity and quality of flowering Chinese cabbage were tested in two soilless cultivation systems: (I) pot cultivation (mixture of peat and sand) and (II) hydroponic culture.

Each year in the Department of Quality and Food Safety Management, the quality of the flowering Chinese cabbage was investigated. This included the measurement of colour, total phenolic content, chlorophyll a, chlorophyll b contents, and antioxidant activity.

1. Cultivation experiment and grow media preparation

All experiments were conducted in an unheated glasshouse located at the Experimental Station in Marcelin at the Poznań University of Life Sciences (Poland). The seed of purple flowering Chinese cabbage were produced by Hubei Wuhan Hongshan Caitai Cultivation Centre. Two soilless cultivation systems were used in all three years experiment. The plants that were grown in pots use a substrate (I), which is a mixture of loamy sand and peat. On the other hand, in a hydroponic system, the plants were grown in closed fertigation system with recirculating a nutrient solution (II).

Seedlings were prepared 2.5 weeks before each experiment. At the phase of 3–4 leaves, seedlings—in the case of pot cultivation—were transplanted into pots filled with a mixture of mineral soil and peat. In the hydroponic system, seedlings were transplanted into Rockwool blocks (Grodan, 100 × 100 × 65 mm) hydrated with the nutrient solution. Ten days later, the plants were transplanted into the hydroponic system and put into the stable place.

In pot experiments, plants were grown in mixture of mineral soil and peat. The levels of nutrients available in the peat after deacidification were as follows (in mg dm^{-3}): N-NH_4 35; N-NO_3 traces; P 20; K 18; Ca 1500; Mg 164; S-SO_4 25; Fe 19.8; Zn 1.8; Mn 2.7; Cu 0.4; B 0.5; Na 18; Cl 29; EC 0.49 mS cm^{-1}; pH (H_2O), 6.00. Plants were grown in 5 dm^3 yellow plastic pots filled with a mixture of mineral soil (loamy sand; d = 1.60 g cm^{-3}; total porosity, 39%) and peat (v/v/1/1). It was used the substrate with the following chemical composition (mg dm^{-3}): P—150, K—200, Ca—1500, Mg—200, Fe—75, Mn—25, Zn—20, Cu—10; pH, 6.0–6.5.

The hydroponic experiments (II) were conducted on plants grown in a special hydroponic module with recirculation of the nutrient solution (NS). The nutrient solution for fertigation had the following chemical composition (mg dm^{-3}): P-PO_4—50, K—200, Ca—120, Mg—60, S-SO_4—95, Fe—1.20, Mn—0.5, Zn—0.19, Cu—0.01, and B—0.011 with EC (electrolytic conductivity)—2.20 mS cm^{-1}.

In the year of 2018, the experiments were conducted in spring (from April to May) and autumn (from September to October), respectively at 5 levels of N nutrition (in mg dm^{-3}): 50, 70, 90, 110, 130 (described as N-50, N-70, N-90, N-110, N-130). The experiments were established using a randomized complete block design. The nitrogen source was ammonium nitrite (NH_4NO_3; 34% N). The results of the first year showed the optimal season was spring and N level ranged from 50 to 90, which would give a guidance to next year’s experiment design.

In the year of 2019, it was studied three levels of nitrogen nutrition: 50, 70 and 90 mg N per dm^{-3}, described as N-50, N-70 and N-90, respectively. The following sources of nitrogen were used: ammonium nitrate NH_4NO_3 (N, 34%), calcium nitrate $Ca(NO_3)_2$ (N, 15.5%; Ca, 18%), magnesium nitrate $Mg(NO_3)_2$ (N, 11%; Mg, 12%), sodium nitrate $NaNO_3$ (N, 15%; Na, 25%), and additional in the case the pot cultivation, urea (CH_4N_2O; N, 46%) has been tested (Jones, 2005).

In the year of 2020, nitrogen nutrition (N-70 and N-90; the source was ammonium nitrate) combined with varied foliar treatments were studied. The foliar treatment included the following chemicals: silicon (Si, 0.2% solution) in the form of choline-stabilized orthosilicic acid (ch-OSA; 0.6% Si; Actisil, Yara, Poland; ch-OSA was obtained from Bio Minerals N.V., Destelbergen, Belgium); selenium (Se, 0.005% solution) in the form of sodium selenite (Na_2SeO_4); lithium (Li, 0.005% solution) in the form of lithium chloride ($LiCl{\cdot}H_2O$); vanadium (V, 0.005% solution) in the form of ammonium vanadate (NH_4VO_3); and salicylic acid (SA, 0.04% solution) in the form of $C_7H_6O_3$ (Sigma-Aldrich, USA). The leaves of the control plants were sprayed with distilled water. The foliar spray treatment (20 ml per plant) was applied three times in both pot and hydroponic experiments: 18, 23, and 28 days after the transplantation.

2. Plant analysis

On the last day of each experiment, the fresh weight of the aerial part of flowering Chinese cabbage was measured directly after cutting. The sample in each group of plant was divided into two parts. The plants for mineral content analysis were dried at 45-50 °C and kept in a dry environment after grounding. The second part of the plants was used for the physicochemical analyses (colour, phenolic content, chlorophyll content, antioxidant activity). It was dried at -59°C, ground and stored in the freezer (-18 °C) till measurements.

2.1. Analysis of Macro- and Microelements in Plants

All analyses were performed on the above-ground parts of the plants. The samples were dried to a constant weight at 45-50°C for 48 h and then ground. Prior to mineralization, the plant material was dried at 105°C for 1 h. To determine the total contents of N, P, K, Ca, Mg, and Na, 1 g of plant material was digested in 20 cm^3 of concentrated (96%, analytically pure) sulphuric acid with hydrogen peroxide (30%, analytically pure) (IUNG, 1972). For total Fe, Mn, Zn, and Cu contents, 2.5 g of plant material was digested in a mixture of concentrated nitric (ultra-pure) and perchloric acids (analytically pure) at a 3:1 ratio (30 cm^3). Following mineralization, the following measurements were taken: total N was measured by the Kjeldahl distillation method in a Parnas Wagner apparatus; P was measured by colourimetry with ammonia molybdate; and K, Ca, Mg, Na (results expressed in % DM—dried matter), Fe, Mn, Zn, and Cu (results expressed in mg kg^{-1} DM) were measured by flame atomic absorption spectroscopy (FAAS) on a Carl Zeiss Jena apparatus (Thornwood, NY, USA). The accuracy of the methods used for chemical analyses and the precision of analytical measurements of nutrient levels were verified by analyzing the LGC7162 reference material (LGC standards) with an average nutrient recovery of 96% (N, P, K, Ca, Mg, Fe, Mn, and Zn).

2.2. Colour measurement

Colour measurement was conducted on Spectrophotometer CM-5 Konica Minolta. Freeze-dried powdered samples of flowering Chinese cabbage were put into the polystyrene containers and placed on the top of the measuring area (diameter 30 mm) of the spectrophotometer. Before each start-up, the calibration of the instrument was performed using the internal white plate and external black box to set 100% white and 100% black, respectively. The light source was D65, and the geometry of the measurements was 10°. The colour was evaluated in L*a*b* CIE system in terms of L* (lightness), a* (redness/greenness) and b* (yellowness/blueness) colour space values. The mean value of three measurements was obtained.

2.3. Sample absorbance scan

The Cary 1E UV/Vis Spectrophotometer was used to perform the scans of the samples. The sample solution was prepared as follows: 0.125 g of freeze-dried flowering Chinese cabbage was extracted with 5 ml methanol. After 5 minutes of mixing on the magnetic stirrer, the sample was filtrated. Then 80 μL of extraction solution was mixed with 3.92 ml methanol and the scan was performed in the range of 410 – 700 nm using pure methanol as the blank sample. Total chlorophylls and total carotenoids were calculated using equations from the protocol (Lichtenthaler et al., 2005).

2.4. Flavonoid content measurement

0.125 g of freeze-dried sample of flowering Chinese cabbage were mixed with 5 ml distilled water on the magnetic stirrer for 30 minutes and then filtrated. The prepared extract was mixed with a 2% methanolic solution of $AlCl_3$ (in ratio 1:10). The sample was left in the dark at room temperature for 15 min before absorbance readings on Cary 1E UV/Visible Spectrophotometer at 410 nm (Singleton & Rossi, 1965).

2.5. Total polyphenol content measurement

Total polyphenol content (TPC) was determined using the Cary 1E spectrophotometer. The procedure was based on Folin–Ciocalteu method as described by Singleton and Rossi (1965).

Briefly, 0.125 g of freeze-dried sample of flowering Chinese cabbage was mixed with 5 ml distilled water for 30 min and filtrated. Then 20 μL of the extract was mixed with 100 μL FCR in 2 ml. After incubation for 3 min at room temperature in the dark, 300 μL 20% (w/v) sodium carbonate solution was added, and the solutions were filled up to 2 ml with distilled water. The sample was mixed again and incubated at room temperature for 2 hours in the dark. The absorbance of the sample was read at 765 nm against blank samples (distilled water instead of the extract). Three replications were performed for each combination. The results were expressed as mg of gallic acid equivalents (GAE).

2.6. Antioxidant ability measurement - DPPH, ABTS, FRAP

Three methods were carried out in this experiment to evaluate the antioxidant activities of flowering Chinese cabbage, namely DPPH, ABTS and FRAP. DPPH and ABTS methods are based on the radical scavenging mechanism of antioxidant action, whereas the FRAP method is on the chelating properties of the antioxidant.

DPPH is an abbreviation for the organic chemical compound 2,2-diphenyl-1-picrylhydrazyl, which is widely used in antioxidant assays. DPPH is a stable radical generated in organic solvent. The methanol solution of DPPH has a dark purple colour which shows a strong absorption at 515 nm wavelength. During the reaction of DPPH radicals with the antioxidant, the single electron of $DPPH^{\cdot}$ radical is paired, which make the solution light colour.

In this experiment, the DPPH free radical-scavenging activity measurement were carried out according to the procedure of (Sánchez-Moreno et al., 1998) with some modifications. 0.25 g of the powdered freeze-dried flowering Chinese cabbage was mixed with 5 ml of 99% methanol for 30 minutes. After filtration 10 μL of the sample were added to 990 μL of DPPH in methanol (0.1 mmol) and mixed. The reaction mixture was incubated in the dark at room temperature for 30 min, and the absorbance decrease caused by the sample was measured at 515 nm against the blank (anhydrous methanol) using a Cary 1E spectrophotometer. The determination of five different concentration samples was carried out.

The capability to scavenge the DPPH radical was calculated using the following equation: $\text{DPPH scavenging effect (\%)} = \frac{(A_0-A_1)}{A_0} \times 100$. A_0 is the absorbance of the control reaction and A_1 is the absorbance of the presence of all the extract samples. The $DPPH^{\cdot}$ radical scavenging activity of the sample extract was expressed in TE – Trolox equivalents (mM of Trolox/mg of flowering Chinese cabbage).

The ABTS method measures the ability of the antioxidant to scavenge the ABTS generated radicals in PBS buffer (Phosphate-buffered saline). Hydrogen-donating antioxidants reduce the ABTS radical so that the blue-green colour of ABTS solution decreases.

The ABTS method that used in this experiment was based on the method of (Re et al. 1999). The extracted sample (as described above) was mixed with $ABTS^{\cdot +}$ in PBS solution (in ratio of 1:100) and mixed. After 6 minutes of incubation the absorbance was measured at 734 nm. The results were expressed in TEAC (Trolox equivalent antioxidant capacity) values using Trolox as the equivalent. TEAC value was calculated as the ratio of the slope of the linear plot for scavenging of $ABTS^{\cdot}$ radicals by the sample extract to the slope of the plot for $ABTS^{\cdot}$ radicals scavenging by the water-soluble vitamin E analog - Trolox.

The FRAP method is based on that Fe^{3+}-tripyridyl-trazine (TPTZ) reduction to ferrous form by the reducing substance at low pH of the environment. Then the dark blue colour of the mixture changes to light colour during the reduction (Benzie & Strain, 1996). In brief, 50 µL of the sample was added to 950 µL of TPTZ working solution and mixed. The absorbance of the sample at 593 nm was determined after incubation for 10 minutes in the dark at ambient temperature. Results were expressed in Trolox equivalents.

3. Statistical analysis

Details of statistical analysis were shown in all manuscripts described in the doctoral thesis.

CHAPTER V Summary of attached publications

1. Publication nr 1:

Liu W., Liu Y., Kleiber T. 2021. A review of progress in current research on flowering Chinese cabbage (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee). *J. Elem.*, 26(1): 149-162. DOI: 10.5601/jelem.2020.25.4.20

Punktacja $MNiSW_{2021}$: 70 pkt

IF_{2021}: 0.923

The paper presents the current state of research on the studied of flowering Chinese cabbage. Based on the available literature, the history of flowering Chinese cabbage has been introduced and the biological value of this vegetable was characterized. Besides, we reviewed the different breeding methods used to develop new varieties of flowering Chinese cabbage meeting specific requirements, such as selecting a hybrid with kale to enrich the genotype, increase the resistance or discover a specific bolting gene.

Flowering Chinese cabbage, also known as ‘Caixin’ in Mandarin Chinese or ‘Choy sum’ in Cantonese, is a widely adapted crop that is popular not only in its native Guangdong province but throughout China. Some cultivars have even been modified to be cold-resistant and are grown in northern China (Z. Wang et al., 2019). Chinese immigrants brought flowering Chinese cabbage to other countries, eventually gaining popularity worldwide (G. Li et al., 2011). The vegetable was first exported from China to Japan in the 1920s and later to North America and Australia (H. He et al., 2000). However, flowering Chinese cabbage remains relatively unknown to Europeans, and research has begun on the feasibility of growing it in Poland (Górna et al., 2016). Flowering Chinese cabbage leaves grow optimally at temperatures ranging from 20-25°C, while a temperature range of 15-20°C is optimal for the formation of flower stalks (S. Song et al., 2012). The plant requires about 32-35 days from sowing to harvest and approximately 80-90 days for seeds to mature. Due to the short growing period, flowering Chinese cabbage exhibits a high multiple cropping index (Peng et al., 2015).

Flowering Chinese cabbage is rich in glucosinolates, polyphenols, amino acids, fatty acids, soluble sugar and vitamin C (Yuan et al., 2019). Every 100 g of the edible part of plants provides 89 kJ of energy. It has the following chemical composition (mg 100 g^{-1}): N – 511.7; K – 55.88; P – 49.16; Ca – 86.18; Mg – 8.593; Na – 35.67; Fe – 2.882; Zn – 1.088; Mn – 0.334; Cu – 0.186; Ni – 0.021; Pb – 0.119; Cd – 0.002; Cr – 0.008. The edible parts also contain 17 types of amino acids, 9 types of saturated fatty acids and 4 types of unsaturated fatty acids (Yang et al., 2002). It is also a good source of flavonoids (1.73 mg g^{-1}) including flavonoid rutin (0.14 mg g^{-1}), and vitamin C (52.16 mg 100 g^{-1}). Furthermore, it has been found to have a high capacity for selenium absorption, which is a beneficial element for consumers (Mo et al., 2006). Due to its high antioxidant active compound content (flavonoids, phenolic acids, carotenoids, vitamin C), it exhibits the antioxidative activity. The TEAC value is 16.93 µmol g^{-1} and the FRAP value is 12.01 µmol Fe (II) g^{-1} in total (Deng et al., 2013).

The quality and yield of flowering Chinese cabbage could be influenced by various factors, such as soil, climate, fertilization and plant genotype. Xie et al. (2011) reported that the yield of purple flowering Chinese cabbage initially increased with increasing fertiliser dose but later decreased. They recommended an optimal nitrogen level of 440-490 kg ha^{-1} to balance economic benefit and quality. Xiahui, Houcheng, and Guangwen (2006) found that application a biochemically controlled release of urea nitrogen fertiliser coating significantly increased flowering Chinese cabbage yield by 19.10 – 28.20% compared to regular urea fertiliser with the same nitrogen dose. G. Li, He, and Xu (2009) recommend a fertilization schedule with an N:P:K ratio of 3:1:1.2 could obtain the highest yield and quality of flowering Chinese cabbage. Q. Chen et al. (2010) suggested a P content of 54.60-87.80 mg kg^{-1} for optimal yield of plants grown in latosolic red soil.

Regarding to other fertilisers, Ji et al. (2020) found that Trichoderma biofertiliser, containing a mix of *T. harzianum*, *T. asperellum, T. hamatum, and T. atroviride* at $2.56 \cdot 10^9$ ml^{-1}, improved flowering Chinese cabbage yield by 37.4%, height by 24.4%, and fresh weight by 41.7% comparing to control. Refined organic fertiliser at dose of 5 kg improved the yield of cultivars: 'Meiqing no. 1' and 'Baoqing 40 days' by 29.1-40.6% (Jian et al., 2004). Foliar spraying is an effective method of improving the yield of plants. According to (Junxi et al., 2010), the yield and quality of flowering Chinese cabbage can be enhanced by foliar spraying with amino acids, particularly glycine (Gly2) at a 200 mg kg^{-1} dosage. Additionally, this method can decrease nitrate content by approximately 85.03% with simultaneous improvement of plant yielding. According to the results of the study conducted by (S. Song et al., 2012), using blue and red shading nets was found to increase the fresh weight of aerial parts by 9% and 44.1% respectively. However, the use of silver and black nets was observed to decrease the fresh weight by 12.7% and 48.5% respectively. The yield of flowering Chinese cabbage is significantly influenced by the size of the flower stalk. To accelerate the bolting time and stem elongation of flowering time of flowering Chinese cabbage, low temperature and gibberellin (GA) treatments were effective (X. Song et al., 2019).

In conclusion, flowering Chinese cabbage (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee) is characterized by quite high biological values. It contains glucosinolates, polyphenols, amino acids, fatty acids, soluble sugar, and vitamin C, which are affected by plant nutrition and fertilization. Nitrogen fertilization is crucial for yield, and balanced use of P, K, and other elements can also impact yield. Organic fertilization may enhance yield, depending on the conditions. Other cultivation methods such as colour netting, insect netting, and plant hormones have been used to improve yield. Proper cultivar selection is necessary to avoid Cd accumulation. Se and Si affect plant resistance to abiotic stresses, including those caused by heavy metals. New genetic technologies are being used in breeding of flowering Chinese cabbage in order to meet market demands. The vegetable has garnered scientific interest and holds potential for further research.

2. Publication nr 2:

Liu W., Muzolf-Panek M., Kleiber T. 2022. Effect of Nitrogen Nutrition and Planting Date on the Yield and Physicochemical Parameters of Flowering Chinese Cabbage. *Agronomy*, *12*, 2869. https://doi.org/10.3390/agronomy1211286

Punktacja $MNiSW_{2021}$: 100 pkt

IF_{2021}: 3,949

This manuscript showed the studies aimed to the evaluation of the intensity of nitrogen nutrition on the response of the plants grown in two different systems of cultivation (substrate and hydroponic) and two varied seasons (spring and autumn). The following nitrogen nutrition levels (50, 70, 90, 110 and 130 mg N per dm^{-3}) were under study. Our hypothesis was that nitrogen nutrition improves plant yield independent of the planting date and system of cultivation.

The results showed that, taking the average values of the two seasons, the yield increased from N-50 to N-90. However, providing more intensive N nutrition (N-110 and N-130) did not significantly modify of plant yields, which means the optimal nitrogen level might be N-90 in our experiment condition. During autumn cultivation in pots, it was observed that the plant's nitrogen (N) content was approximately 60% higher than in spring cultivation. Besides, there were no significant differences in N leaf content among the combinations in hydroponic cultivation. Nonetheless, hydroponic cultivation generally resulted in higher N levels compared to substrate cultivation.

The highest K and Ca status were determined both in substrate and hydroponic in autumn season. In general, enhancing the intensity of N nutrition resulted in an improvement of the nutrient status with macroelements. The content of Fe was higher during the autumn period in both types of cultivation. On the other hand, for Mn, the highest contents were determined in spring for hydroponics and in autumn for substrate. The opposite results were found for Cu content in plants. Regardless of the cultivation type, the Na content in plants was higher in autumn compared to spring.

Plant grown in substrate or hydroponic in spring showed the highest lightness in comparison to the plants cultivated in autumn. Moreover, in hydroponic conditions, the autumn samples had higher L* values (48.13) compared to the autumn samples from substrate cultivation (46.92). Both in pot and hydroponic cultivation plants were shown to have similar a* and b* colour-coordinate values, but lower values were observed in autumn compared to spring samples. However, the influence of N nutrition on colour changes was various depending on intensity of N nutrition. In the substrate experiment, the greenness of plants increased with higher nitrogen levels (from N-50 to N-110 in spring and to N-90 in autumn), but further increase of the nitrogen level resulted in a decrease in greenness (higher a* values). Considering the chlorophyll content in plants grown in substrate, a significant effect of N nutrition was observed for the parameters such as Chl a, the sum of chlorophylls, and Chl a/Chl b ratio. Additionally, the season of cultivation affected the values of the indices, except for the chlorophyll ratio (Chl a/Chl b). The highest values of chlorophylls and carotenoids were found in the nitrogen range from N-90 to N-130.

Generally, in spring season plants were characterized by higher antioxidant activity and phenolic content compared to autumn. In the case of plants grown in substrate, the highest values of the parameters were obtained. Furthermore, different N levels were required to achieve maximum values phenolic content and the antioxidant activity parameters. In the pot cultivation, the antioxidant activity was the highest at a moderate nitrogen fertilization level (N-90 and/or N-110) taking the mean value from two seasons. The total phenolic content (TPC) was the highest at a moderate nitrogen concentration (N-90), while total flavonoid content peaked at N-70 and N-90. In the hydroponic system, the spring samples showed the highest antioxidant activity values at N nutrition ranging from 110 to 130. For the autumn samples, the highest antioxidant activity was determined at various N levels, specifically at low and medium N nutrition levels (N-50, N-90, and N-110). The phenolic content reached maximum value at N-90 for the spring samples and N-110 for the autumn samples. On the other hand, there is no evident trend in flavonoid content after nitrogen fertilization at different doses.

The results obtained in this study provide useful information on the flowering Chinese cabbage response to various N levels in two different cultivation systems and seasons, indicating that agricultural management gives the possibility to increase the quality of flowering Chinese cabbage from the nutritional and phytochemical point of view. For the mean of two seasons, the yield increased from N-50 to N-90, and N-90 was an optimal level. More intensive N nutrition (N-110 and N-130) did not significantly modify plant yields both in substrate and hydroponic.

3. Publication nr 3:

Liu W., Muzolf-Panek M., Kleiber, T. 2023. Effect of Varied Nitrogen Sources and Type of Cultivation on the Yield and Physicochemical Parameters of flowering Chinese cabbage (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee). *Appl. Sci.*, 13, 5691. https://doi.org/10.3390/app13095691

Punktacja MNiSW$_{2021}$: 100 pkt

IF$_{2021}$: 2,838

The aim of the studies was to investigate the effects of varied nitrogen sources: NH_4NO_3 (N 34%), $Ca(NO_3)_2$ (N 15.5%, Ca 18%), $Mg(NO_3)_2$ (N 11%, Mg 12%), $NaNO_3$ (N 15%, Na 25%) and urea (N 46%, only in pot) and also increasing intensity of N nutrition with that fertilisers (50, 70, and 90 mg N dm^{-3}) on the yield and the physicochemical parameters of flowering Chinese cabbage (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee) grown in two varied soilless systems: pot cultivation (mixture of peat and sand) and hydroponic culture.

In the case of pot cultivation, it was observed that plant yielding increased with increased levels of N nutrition, reaching their maximum at N-90. When different fertilisers were utilized for the cultivation of plants, significant differences in yields were observed at the same N level. Applying $Mg(NO_3)_2$ resulted in the highest average yields, while using $Ca(NO_3)_2$ led to the lowest yields. It is worth noting that despite the high doses of sodium applied (up to 180 mg·dm^{-3}) during the $NaNO_3$ application, there was no decrease in plant yielding compared to other combinations.

In hydroponic cultivation, no significant increase in plant yield was observed between the application of N-70 and N-90, based on the mean values. No statistically significant differences were found among the tested fertilisers when considering the average crop yield. Similar to pot cultivation, no significant reduction in yield was observed for the highest application rate of $NaNO_3$. However, in the case of calcium nitrate, N-90 treatment resulted in a significant increase in plant yield compared to N-70. Compared to pot cultivation, hydroponic cultivation of flowering Chinese cabbage showed higher yields 174.2 g·plant^{-1}, achieved when NH_4NO_3 at N-70 was applied.

Interestingly, in pot cultivation, there was a notable increase in the nitrogen level in the aboveground parts of the plants according with an increasing intensity of N nutrition. However, this effect was not observed in hydroponics. In pot cultivation, the nitrogen status of plants was similar for all nitrogen fertilisers type excepting urea. However, in hydroponic cultivation using $NaNO_3$, nitrogen contents were higher than for NH_4NO_3 and $Mg(NO_3)_2$. Changes in nitrogen nutrition did not significantly affect phosphorus levels. The potassium content varied with different nitrogen carriers, and generally, NH_4NO_3 and $Mg(NO_3)_2$ resulted in higher potassium levels in both pot cultivation and hydroponics.

The results from pot cultivation showed that urea fertilization resulted in the highest lightness (L*) values (mean of 46.4), while $Ca(NO_3)_2$ had the lowest L* values (mean of 43.8). The samples treated with urea had the lowest chroma (C) and hue angle (h) values. In hydroponic cultivation, the flowering Chinese cabbage treated with $Mg(NO_3)_2$ at N-90 exhibited the highest L* value (50.77). In pot cultivation, the nutrition source affected carotenoids, Chl a/Chl b, and (Chl a + Chl b)/(x + c), while the N level significantly influenced Chl a, Chl b, carotenoids, and Chl a + Chl b. Hydroponic cultivation revealed significant effects of both N source and N nutrition intensity on chlorophyll and carotenoid contents. Additionally, in flowering Chinese cabbage, Chl a content increased with NH_4NO_3 treatment and decreased with $NaNO_3$ treatment as N levels increased, while Chl b content decreased with $NaNO_3$ treatment at higher N doses.

Both in pot and also hydroponic cultivation systems, the same N levels and N sources had statistically significant effects on all parameters describing the antioxidant activity and phenolic content. In hydroponics, the samples exhibited higher DPPH scavenging activity but lower TEAC, FRAP, TPC, and TFC values compared to samples from pot cultivation. In pot cultivation, regardless of the N dose, the samples treated with $Mg(NO_3)_2$ had the highest values of DPPH, FRAP, TPC, and TFC, while the sample treated with $NaNO_3$ had the highest TEAC value. The highest DPPH scavenging activity (115.8 μmol g^{-1}) was found in the sample treated with $Mg(NO_3)_2$ at N-70, and the highest TEAC value (536.69 μmol g^{-1}) was determined in the sample treated with $NaNO_3$ at N-90. In hydroponic, different conclusions were drawn as the maximum values of DPPH and TEAC scavenging activity were 122.41 μmol g^{-1} DW and 409.51 μmol g^{-1} DW, respectively, both after the NH_4NO_3 treatment. The highest FRAP value (24.12 mmol TE g^{-1}) was achieved for $Ca(NO_3)_2$ at N-90. The maximum TFC value was found in hydroponics for samples fertilized with $NaNO_3$ (1.43 mg g^{-1}).

In conclusion, it was found rather high tolerance of flowering Chinese cabbage to sodium. Increasing levels of N nutrition significantly increased the average yield of plants grown in pots, while in hydroponics, average yields at N-70 were comparable to N-90. The nitrogen source modified plant nutrition with macro- and micronutrients regardless of cultivation type. In case of pot cultivation, the highest pigment content (chlorophylls and carotenoids) was determined in the samples treated with urea at N-90 dose whereas in hydroponic in samples treated with magnesium nitrate (N-70). However, in pot cultivation, pigment content was not correlated to the colour parameters (L*a*b*), whereas in a hydroponic system, Chl *b* was correlated with L* and with b*. The highest antioxidant activity and phenolic content were obtained in the samples from the pot system treated with magnesium nitrate at high N-doses (N-70 and N-90), as well as ammonium and sodium nitrates at the N-90 dose. In hydroponic systems, the highest DPPH activity was observed after treatment with sodium nitrate at N-70 and N-90 doses. In the experiment, except for the sample TEAC activity in the urea treatment, the DPPH and TEAC activities of the samples in pot cultivation increased significantly as the N level increased from N-50 to N-70. In conducted studies, it was confirmed the hypothesis that the N-sources and the intensity of N nutrition modify the yield of plants, both in quantitative and qualitative aspects.

4. Publication nr 4:

Liu W., Muzolf-Panek M., Kleiber T. 2022. The Effect of Various Foliar Treatments and Nitrogen Nutrition Levels on the Yield and Physicochemical Parameters of flowering Chinese cabbage.
Agronomy, *12*, 737. https://doi.org/10.3390/agronomy12030737

Punktacja MNiSW$_{2021}$: 100 pkt
IF$_{2021}$: 3,949

This study investigated the effect of nitrogen nutrition levels (70 and 90 mg N per dm^3) and various foliar treatments (Se, Si, Li, V, and SA—salicylic acid) on the quantity of the yield and physicochemical parameters of flowering Chinese cabbage grown in two varied soilless cultivation systems: pot cultivation (mixture of peat and sand) and hydroponic. There are no practical recommendations for foliar spraying for the cultivation of this plant species.

The research hypothesis set out that the level of nitrogen nutrition and the application of foliar spraying modify the yielding of plants both in quantitative and qualitative aspects.

The yield of flowering Chinese cabbage, cultivated in both pots and hydroponic systems, was significantly influenced by various foliar spray treatments and nitrogen nutrition. Generally, the plants grown hydroponically showed higher yields compared to those grown in pot. Increasing nitrogen nutrition improved the yielding of plants. The influence of foliar spraying on yielding varied depending on the nitrogen level and cultivation method. The highest yield (182.0 g·plant^{-1}) in pot cultivation was found at N-90 with simultaneous Li spray treatment, which was about 17.5% higher than the yield of control plants (for N-90). The highest yield in the hydroponic system was found in the control at N-90. However, there were no statistically significant differences between the control and the rest of combinations (Si, Se, Li, and SA). In contrast to pot cultivation, V treatment significantly decreased the yield by 25.2% compared to the control combination. For the lower nitrogen nutrition level, the lowest yield (139.6 g) was found under Si treatment (reduced about 30.5% comparing to the control).

The effect of the studied factors on the plant nutrient status was varied. Increasing nitrogen nutrition of plants grown in pots improved nitrogen status of plants, but did not affect other macroelements (P, K, Ca, and Mg) content. The combinations treated with SA characterized the highest N content but the lowest yield. The influence of SA treatment on Mg status depended on the N nutrition level. Higher N nutrition reduced P content under Se spray treatment. At N-70, SA treatment decreased Ca content.

Increased N nutrition improved Fe, Mn, Zn, and Cu status. Se, Li, V, and SA treatments decreased Fe content, while Si and SA treatments improved it. Li treatment decreased P but increased Na content. The influence of Si treatment on Mg content varied with N level. Increased N nutrition improved mean Fe, Mn, and Cu content. Si and Se treatments increased Fe content, while all foliar treatments reduced Mn content. At N-90, Si and Se treatments increased Zn content, while Li, V, and SA treatments decreased it. SA treatment increased Cu content, while Li and V treatments decreased it.

Studied factors influenced on the colour parameters. SA-treated samples had the lowest lightness, while V-treated (N-70) and Se-treated (N-70) had the highest lightness. In pot cultivation, Li and SA treatments reduced greenness, and the highest yellowness was observed in control (N-70) and SA-treated (N-90) samples. In hydroponic SA treatment resulted in the highest greenness and yellowness, while V and Si treatments decreased greenness. In the substrate cultivation, Chl a, Chl b, and carotenoid content increased with the increase of N levels, except in the Li-treated sample. Foliar spray treatments (Se, Si, Li, SA) at N-70 increased Chl a content compared to the control. V foliar spray treatments had various effects on chlorophyll content.

In pot cultivation, N nutrition did not affect TPC but influenced TFC. Higher N levels resulted in lower TFC values. The SA treatment at N-70 had the highest TPC (26.30% higher than the control), while the Si treatment at N-90 had the lowest TPC. The lowest TFC was observed after SA treatment at N-90, and the highest was after Se treatment at N-70. TPC and TFC were significantly correlated ($r=0.48$) in pot cultivation. In the hydroponic, N level did not influence TFC, but higher N levels decreased TPC. The highest TPC was after SA treatment at N-70, and the lowest was after Li treatment at N-90. Si treatment increased TFC at N-90, while other foliar treatments did not differ from the control. There was no correlation between TPC and TFC in the hydroponic experiment.

Antioxidant activity of plants was assessed using three methods: TEAC, DPPH, and FRAP. In the pot experiment, the intensity of N nutrition and foliar treatment significantly affected the antioxidant activity. SA treatment at N-70 showed the highest TEAC values, while Li and V treatments decreased TEAC values. The lowest activity measured by the DPPH assay was found in the samples treated with after SA and V (N-70), and the highest activity after Li treatment (N-90). The highest chelating activity (FRAP) of flowering Chinese cabbage was noted after Se treatment (N-90). In the pot cultivation DPPH activity was correlated with TPC ($r=0.42$), and FRAP activity correlated with carotenoid content ($r=0.75$). In the hydroponic, Se treatment induced the highest antioxidant activity, while SA treated plants showed the lowest activity. Correlation coefficients were high among the different antioxidant activity assays.

Antioxidant activity did not correlate with phenolic or carotenoid content in the hydroponic experiment.

In this study, it was confirmed that the intensity of nitrogen nutrition and the application of foliar spraying modify the yield of plants, both the quantity and the quality aspects. The factors under analysis had a diversified and multidirectional influence on the yield, growth, and quality of the plants.

CHAPTER VI Conclusion

The studies carried out as part of the dissertation led to the following conclusions:

1. Increasing nitrogen nutrition had a significant influence on:
   a) Plant yielding. Increasing nitrogen nutrition positively affected yields in substrate and hydroponic cultivation systems. In pot cultivation, the range N level from N-50 to N-90 increased the yield both in spring and autumn. In hydroponic cultivation, there was no significant difference between different N level in spring. But in autumn season, yielding were increased from N-50 to N-90. Generally, the optimal nitrogen level to achieve maximum yields was found for N-90. Higher nitrogen doses (>N-90) did not significantly enhance yields. In general, the plants grown in hydroponic systems were characterised by higher yields than those grown in pots.
   b) Plant nutrient status. In pot cultivation, the N content of the plant increased with the increasing nitrogen nutrition. In hydroponic, the N content of plants was stable with the increase of nutrient level. Ca and Mg contents in plants were stable in the case of hydroponics compared to cultivation in the substrate.
   c) The colour of leaves. In both the spring and autumn seasons in the pot experiment, the greenness of the plants increased with an increase in the nitrogen level (from N-50 to N-110 in spring and to N-90 in autumn).
   d) Pigments. The highest content of chlorophylls and carotenoids were generally determined at the N-90 to N-130.
   e) Antioxidant activity. In general, the spring samples were characterised by higher values of the antioxidant activity and phenolic content values than the autumn one; the highest values were determined for the pot cultivation.

2. Planting season had a significant influence on:
   a) During autumn cultivation in the substrate the N content was approximately 60% higher than in spring season. The plants grown in spring obtained higher biomass comparing with autumn.
   b) The plant nutrient status: in the substrate cultivation, the content of N, K, Ca, Fe, Mn, and Na in leaves was higher in autumn compared to spring; the same tendencies were found in the cases of P, K, Ca, Fe, Zn, Cu, Na

in the hydroponic system. The content of Fe was higher in the autumn period (for both types of cultivation).

c) Colour parameters of plants . The greenness of flowering Chinese cabbage in spring is higher than autumn, especially in the substrate.

3. Different nitrogen source had a significant influence on:
   a) Plant yielding. Mean the highest yields of plants grown in hydroponic were found under the application of $Mg(NO_3)_2$, the lowest—when $Ca(NO_3)_2$ was used. Compared to pot cultivation, the yields of plants in hydroponics were higher than pot. The highest yielding of plants (174.2 $g \cdot plant^{-1}$) were obtained at N-70 with NH_4NO_3 in hydroponic. Increasing levels of N nutrition significantly increased the average yield of the plants grown in pots, while in hydroponics, the average yields at N-70 were comparable to those at N-90.
   b) Nutrient content. The nitrogen source modified plant nutrition in terms of N, P, K, Ca, Mg, Na, Fe, Mn, Zn and Cu.
   c) Pigment content. For the pot cultivation system, the highest pigment contents (chlorophylls and carotenoids) were found in the samples treated with urea at the N-90. In the hydroponic system, the highest pigment contents were determined in the plants treated grown at N-70 (magnesium nitrate).
   d) Antioxidant activity. In the hydroponic systems, the highest DPPH activity was found after treatment with sodium nitrate at the N-70 and N-90 doses. The DPPH and TEAC scavenging activities of the samples in pot cultivation increased significantly as the N level increased from N-50 to N-70 after the urea treatment.

4. Foliar treatment had a significant influence on:
   a) Plants yielding. The highest yield (182.0 $g \cdot plant^{-1}$) in the pot cultivation was found at N-90 with the simultaneous Li spray treatment. In comparison with the control combination, the Se, Si, and SA treatments reduced the yield by about 21.8%, 17.1%, and 25.7%, respectively.
   b) N content. The highest N content was found in the combinations treated with SA (both at N-70 and N-90), but these combinations were characterized by the lowest yielding.
   c) Pigment content and the antioxidant activity. The samples sprayed with Se and Si were characterized by the high content of Chl a, Chl b, and

carotenoids in the pot experiment and high antioxidant activity (FRAP in the pot experiment and TEAC, DPPH, and FRAP in the hydroponic experiment).

The SA foliar spray treatment significantly influenced the plants' quality and could be classified properly according to the antioxidant activity, chlorophyll content, carotenoid content, lightness, yellowness, and the N level. In the pot experiment, SA-treated plants showed high radical scavenging activity (the TEAC values at N-70) and contained high levels of N, chlorophylls, and carotenoids. In the hydroponic experiment, the SA-sprayed sample was characterized by low antioxidant activity and low content of chlorophylls and lightness but high yellowness.

5. The suitability of plants for soilless cultivation was proved, both in the substrate (mixture mineral soil and sand) and in rockwool (cultivation with recirculation of the nutrient solution).

In conclusion, the results confirmed the hypothesis. After a comprehensive analysis of the extensive dataset accumulated during this rigorous research project, definitive conclusions can be drawn with confidence and conviction. The empirical evidence and statistical analyses indicate a strong correlation between the projected theoretical framework and the actual findings, supporting the validity of the initial hypothesis. These conclusive results serve to validate the original research supposition and provide compelling evidence that reinforces the scholarly foundation upon which this study is built. The outcomes of this research underscore the significance and reliability of the findings, contributing to the existing body of knowledge in the field of horticulture.

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Abbreviations:
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1. **Liu W**., Liu Y., Kleiber T. 2021. A review of progress in current research on Flowering Chinese Cabbage (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee). *J. Elem.*, 26(1): 149-162. DOI: 10.5601/jelem.2020.25.4.20

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2. **Liu, W**.; Muzolf-Panek, M.; Kleiber, T. Effect of Nitrogen Nutrition and Planting Date on the Yield and Physicochemical Parameters of Flowering Chinese Cabbage. *Agronomy* 2022, *12*, 2869. https://doi.org/10.3390/agronomy1211286

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3. **Liu, W**.; Muzolf-Panek, M.; Kleiber, T. Effect of Varied Nitrogen Sources and Type of Cultivation on the Yield and Physicochemical Parameters of Flowering Chinese Cabbage (*Brassica campestris* L. ssp. *chinensis* var. *utilis* Tsen et Lee). *Appl. Sci.* 2023, 13, 5691. https://doi.org/10.3390/app13095691

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4. **Liu, W**.; Muzolf-Panek, M.; Kleiber, T. The Effect of Various Foliar Treatments and Nitrogen Nutrition Levels on the Yield and Physicochemical Parameters of Flowering Chinese Cabbage. *Agronomy* 2022, *12*, 737.
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