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## TRITERPENE SAPONINS AS BITTER COMPONENTS OF BEETROOT

### S u m m a r y

Many varieties of beetroots (*Beta vulgaris* L.) are valued for their productivity as well as their content of nutrients and pigments from a group of betalains that have strong antioxidant properties. On the other hand, the strong bitterness of roots of several beet varieties is a frequent reason for their being not accepted by consumers. The hitherto published studies describe too vaguely the diversity of beetroot varieties in terms of their bitter taste. Up to now, it is still not clear, which of the secondary metabolites that naturally occur in beetroots is responsible for their bitter taste and aftertaste. The objective of this study was to determine the group of compounds that caused that beetroots had a bitter taste and bitter aftertaste of a high intensity level. The first stage in the research study was to select the most bitter beetroot cultivars based on the sensory characteristics of fresh beet roots (of their flesh and skin) of six cultivars ('Nochowski', 'Chrobry', 'Noe 21', 'Rywal', 'Opolski', and 'Wodan'). The sensory profile of the analysed group of beets showed that the flesh and skin of the 'Nochowski', 'Chrobry', and 'Noe 21' cultivars were characterized by the most intense bitterness. The 'Rywal', 'Opolski', and 'Wodan' cultivars were marked by a relatively low intensity level of the bitterness notes. A mixture of triterpene saponins was isolated from a lyophilisate in the roots of the 'Nochowski' cultivar that, according to the sensory evaluation results, was classified into the group with the strongest bitterness traits. The results of sensory analyses of the saponin mixture of the 'Nochowski' cultivar, its concentration being  $C = 1.3515 \text{ g/dm}^3$ , confirmed that the group of those compounds had a strong bitter taste comparable to that of the quinine solution at a  $C_2 = 6.6 \times 10^{-3} \text{ g/dm}^3$  concentration. It was also proved that the beetroot extracts tested were a mixture composed exclusively of saponin compounds, which varied in their chemical structure.

**Key words:** beetroot, triterpene saponins, bitter taste, bitter aftertaste

### Introduction

Beetroot is a raw material considered as a specialty of the Polish agriculture. Bitter cultivars of red beet are characterised by high contents of betalain pigments; thus,

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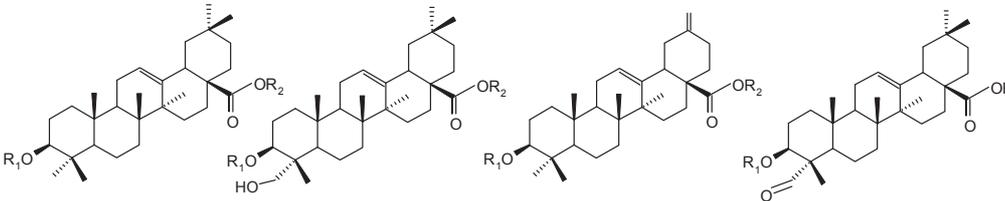
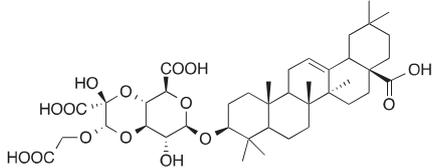
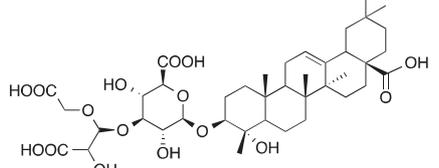
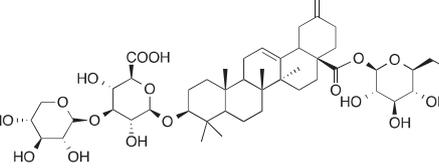
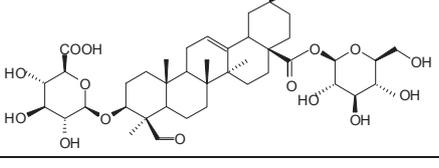
they are desirable cultivars in the food industry. Roots of red beet show a high antioxidant capacity associated with the content of betalain pigments therein. Betalains are effective free radical scavengers and pre-treatment with beetroot juice can counteract xenobiotic-induced oxidative stress in rats [13]. Red beets are typically consumed in a processed form; technological processes (i.e. blanching, drying and heating) enhance the antioxidant potential of that raw material [11]. However, the processing does not considerably enhance the bioavailability of betacyanins contained in red beet roots, which could be caused by partial degradation in the alimentary tract and hindered absorption in the small intestine. The low availability of betacyanins is confirmed by haematological analyses [12].

Roots of red beet cultivars, such as the 'Chrobry' or 'Nochowski' cultivars analysed in this study, with high contents of betalain pigments and, at the same time, characterised by a bitter taste, are used in the food industry to produce, for example, beet crisps (a unique product: crispy slices of beet that are fat-free and with no preservatives added) as well as concentrates and concentrated beet juices. Beetroot concentrate is used as an addition to soups, fruit and vegetable juices, ice-cream, desserts, pasta, and pickles.

Literature sources provide no accurate data to explain which components cause the bitter taste of red beetroots, and to show the variation in bitterness among the cultivars. Most typically, individual cultivars are characterised by their organoleptic attributes, such as flavour, colour, aroma, etc. The variation in terms of bitter taste attributes of different parts of beetroots was described by Biegańska-Marecik et al. [6]. Those authors showed that the skin, root head, and root base were the most bitter parts of the roots, with the centre being the least bitter. However, they did not explain which of the naturally occurring secondary metabolites in red beet roots were responsible for the bitter taste. Also, other studies presented to date did not discuss the occurrence of bitter aftertaste in beetroots [16, 17].

This taste attribute of red beet roots is particularly important for consumers since it eliminates those cultivars that produce a strong bitter aftertaste after consumption.

To the best of our knowledge, no literature sources give any information on saponins found to be bitter substances in beetroot. Many saponins are known to exhibit biological activity, such as antiviral, antidiabetic, cytotoxic, or allelopathic activity, etc., which makes those two substances, and the plants containing them, interesting objects for pharmacological investigations [1, 2, 3, 7, 15, 19].

				
No	R <sub>1</sub>	R <sub>2</sub>	Aglycone	Example of saponin structure
1	Diox -UrA	H	Oleanolic acid	
2	Act - UrA	H	Hederagenin	
3	Pen-UrA	Hex	Akebonoic acid	
4	UrA	Hex	Gypsogenin	

Explanatory notes / objaśnienia:

Act – acetal substituent / podstawnik acetalowy; Diox – dioxolane substituent / podstawnik dioksolanowy; Hex – hexose / heksoza; Pen – pentose / pentoza; UrA – uronic acid / kwas uronowy.

Fig. 1. Chemical structure of saponin aglycones found in roots of beetroot *Beta vulgaris* L. and examples of saponin structures [18]

Rys. 1. Struktury chemiczne aglikonów saponin zawartych w korzeniach buraka ćwikłowego *Beta vulgaris* L. oraz przykłady struktur saponin [18]

As part of our ongoing studies on the isolation and identification of triterpene saponins in the roots of red beet, we found that all the found compounds were glyco-

sides of four different aglycone structures with unique, dioxolane-type and acetal-type substituents (Fig. 1). Based on the high-resolution mass measurements, 44 saponins were detected and identified in the saponin fractions in the roots of beetroot [18]. Those saponins included betavulgarosides I-X identified in the sugar beet [23, 24] and the saponins previously identified in the beetroot [19]. Under the research study, there were analysed the bitter taste and the aftertaste of the identified mixture of saponins in the 'Nochowski' cultivar.

The objective of the research study was to select, on the basis of the results of sensory evaluation (of flesh and skin), the most bitter cultivars of red beet and to determine a group of compounds that cause the high intensity level of bitter taste and aftertaste in beetroot.

### Material and methods

The study was performed using beetroots (*Beta vulgaris* L.) of the 'Nochowski', 'Chrobry', 'Noe 21', 'Rywal', 'Opolski', and 'Wodan' cultivars originating from the Seed Breeding and Horticultural Station at Nochowo (Poland). After harvesting, fresh red beet roots were washed under running water, manually peeled to remove their skins of approx. 0.4 ÷ 0.6 cm in thickness from the flesh of red beetroot and, then, divided into 2 groups: skin and flesh, i.e. the centre of the beets, which were lyophilized and ground. Lyophilisation conditions: freezing of the red beetroot to – 75 °C, drying at 15 °C for 27 h, the temperature of a heating plate in the lyophilisate 20 °C, Lyophilizer Freezezone 6L (Labconco, USA). The intensity of taste attributes of the raw beet root samples was measured using a short 5-point scale [5]. The following taste attributes were distinguished: bitter, bitter aftertaste, sweet, and sour.

In the sensory analysis on taste quality of fresh roots of red beet, there was applied a sensory panel consisting of 11 selected panellists [4, 9]. The sensory assessment of isolate saponins (extracts) was conducted by a panel of 11 experts (testers and super-testers [10]) whose levels of sensitivity to bitter taste were average and maximum. The experts of the panel [21] were selected and trained specifically with respect to their sensory ability with particular focus on the identification of, mainly, bitter taste and, also, sweet, salty, and sour tastes [14]. The panellists selected to sit on the panel of experts were all trained in testing taste attributes. All the sensory tests were performed in a sensory laboratory under the fully controllable testing conditions [22].

The samples to be sensory evaluated were prepared using the following procedure: from among the randomly selected red beetroots (6 roots of every cultivar), pieces weighing approx. 2 g were taken of their skin and their flesh. The roots of the 'Nochowski' cultivar, characterised by the highest intensity level of bitter taste, were used to isolate saponins. The roots of red beets were washed under running water, their skin was removed (because it was found to be the most bitter part of beetroots) and

lyophilised at 20 °C (in a Labconco Stoppering Freeze Dryer, USA) and, next, comminuted in a mill.

The mixture of saponins was isolated using the following procedure: the first step consisted in extracting the lyophilised plant material (skins of the red beet roots) with ether in a Soxhlet apparatus for 16 h in order to remove fats [8, 20]. The crude extract was re-extracted for 3 h in an ultrasonic bath in 80 % methanol. Three independent extractions were performed for every sample. After filtering the extract, a residue from the filter was extracted twice and evaporated under reduced pressure (Buchi Heating Bath B-480, USA). Subsequently, the methanol extract was suspended in water and loaded onto LiChroprep RP-18 (40-63  $\mu\text{m}$ ; Merck, Germany), 60 mm  $\times$  100 mm column equilibrated with 5 % methanol. The column was first washed with water to remove carbohydrates and betalain pigments and, next, with 40 % methanol to elute the rest of betalain pigments. Finally, the saponin fraction was eluted with 80 % methanol. The evaporation of the solvents yielded a crude saponin fraction (2.3 g). The fraction of saponins was monitored by TLC on silica gel 60 F<sub>254</sub> plates (Merck, Germany) with a chloroform/methanol/water solvent system (7:2:2 v/v/v). The spots were detected through spraying the plates with a Libermann-Burchard reagent and followed by heating at 130 °C.

Subsequently, a solid phase extraction (SPE) was applied to re-purify the isolated compounds and, then, the fraction of saponins (2.3 g) was dissolved in water and deposited on preconditioned 5 g PR-18 Lichrolut cartridges (Merck, Germany). The elutes were monitored on silica gel TLC plates (Merck, Germany) developed in the following system of solvents: chloroform: methanol: water (60:40:10 v/v) [20]. The triterpenoids were visualized by spraying the TLC plates with a Libermann-Burchard reagent. The first fraction (F1) was eluted with water and contained mainly saccharides; the second fraction (F2) was eluted with 40 % methanol and contained, additionally, betalains. The saponins were eluted in the third fraction using 80 % methanol. The methanolic extract (F3) was evaporated under reduced pressure (at a 30 °C vapour temperature in the flask). As a result, 15 mg of saponin mixture was obtained of 1 g of lyophilised plant material. The saponins from this fraction were identified and characterised in our previous study [18].

The fractions of saponins (F3) were dissolved in a distilled water to a concentration of  $C = 5.406 \text{ g/dm}^3$  for a sample of the E<sub>0</sub> saponin mixture and  $C = 1.3515 \text{ g/dm}^3$  for the samples of the E<sub>I</sub> and E<sub>II</sub> saponin mixtures and, subsequently, the sensory analyses were conducted. The type of the taste attribute (bitter, bitter sweet, or sour) yielded by the saponin extract was identified by the sensory panel in the so-called control test (E<sub>0</sub> sample). In the sensory evaluation of relative intensity of bitterness of the saponin extracts, the multiple paired comparison test was applied [5] (referring the samples of E<sub>I</sub> and E<sub>II</sub> extracts to the bitterness standard). In this method, quinine was used

as a reference standard at four ( $W_1$ - $W_4$ ) different concentrations above the stimulus threshold ( $[g/dm^3]$ :  $C_1 = 5.16 \times 10^{-3}$ ;  $C_2 = 6.6 \times 10^{-3}$ ;  $C_3 = 9.54 \times 10^{-3}$ ;  $C_4 = 15.42 \times 10^{-3}$ ). All the analyses were performed in three independent replications; the data were described AS mean values, with standard deviation ( $\pm$  SD). Calculations were made using a Microsoft Excel spreadsheet and a Statistica software version 10. Results were statistically analysed using an analysis of variance (ANOVA of the main effects at  $p = 0.05$ ) with a Statistica software.

### Results and discussion

The main stage of all the analyses comprised a sensory evaluation of raw roots of 6 beetroot cultivars ('Nochowski', 'Chrobry', 'Noe 21', 'Rywal', 'Opolski', and 'Wodan'). The sensory evaluation was conducted on the samples of both the skin and the flesh of roots of the abovementioned beetroot cultivars. Tab. 1 shows the results of the sensory analyses of beetroot cultivars and beet root parts.

Table 1. Results of sensory evaluation of taste of various parts of beet roots

Tabela 1. Wyniki oceny sensorycznej smaku różnych części korzeni buraków ćwikłowych

Cultivar of beet-root Odmiana buraka ćwikłowego	Part of root Część korzenia	Taste characteristics / Wyróżniki smaku			
		bitter gorzki	bitter aftertaste posmak gorzki	sweet słodki	sour kwaśny
'Nochowski'	flesh	1.9 ± 0.1	2.4 ± 0.2	4.1 ± 0.1	0.7 ± 0.5
	skin	3.7 ± 0.2	3.3 ± 0.3	1.1 ± 0.1	1.0 ± 0.1
'Chrobry'	flesh	1.8 ± 0.3	2.2 ± 0.1	3.9 ± 0.3	0.3 ± 0.1
	skin	3.5 ± 0.1	3.2 ± 0.4	1.4 ± 0.4	0.5 ± 0.5
'Noe 21'	flesh	2.3 ± 0.1	2.4 ± 0.3	4.0 ± 0.2	0.6 ± 0.4
	skin	3.6 ± 0.2	3.5 ± 0.3	1.2 ± 0.3	0.9 ± 0.3
'Rywal'	flesh	0.5 ± 0.3	0.2 ± 0.4	3.5 ± 0.5	0.4 ± 0.5
	skin	0.6 ± 0.2	0.4 ± 0.2	3.9 ± 0.5	0.5 ± 0.1
'Opolski'	flesh	0.5 ± 0.4	0.9 ± 0.3	4.2 ± 0.6	0.7 ± 0.3
	skin	0.9 ± 0.2	1.0 ± 0.3	4.8 ± 0.4	0.2 ± 0.4
'Wodan'	flesh	0.7 ± 0.5	0.4 ± 0.5	4.6 ± 0.5	0.4 ± 0.5
	skin	1.1 ± 0.2	1.4 ± 0.3	4.2 ± 0.1	0.8 ± 0.1

Table 1 shows mean values  $\pm$  standard deviations. / W tabeli przedstawiono wartości średnie  $\pm$  odchylenia standardowe.

The score rating of the bitter taste intensity of the above mentioned red beet cultivars ranged from 0.5 to 3.7 points, while the intensity scores of the bitter aftertaste ranged from 0.4 to 3.5 on a 5-point scale. The bitter taste and aftertaste of the skin of red beet roots had a higher intensity level than that of the flesh. As for the skin, the

score rating of the intense bitter taste thereof ranged from 0.6 to 3.7, while that of the flesh ranged from 0.5 to 2.3 depending on the cultivar. The highest intensity level of bitter taste and aftertaste was recorded for the roots of the ‘Nochowski’, ‘Chrobry’, and ‘Noe 21’ cultivars. The score rating of the bitter aftertaste of the skin of the ‘Nochowski’, ‘Chrobry’, and ‘Noe 21’ cultivars was between 1.8 and 3.7.

It was also found that the cultivars with a high intensity level of bitter taste had, at the same time, a lower intensity level of the sweet taste (‘Nochowski’, ‘Chrobry’, and ‘Noe 21’ cultivars). As for the other cultivars, a high intensity level was recorded only for the sweet taste. As regards all the cultivars evaluated, the intensity of their sour taste was evaluated as low; its score rating ranged from 0.2 to 1.0.

Based on the statistical analysis, it was confirmed that the individual red beet cultivars significantly differed in the intensity levels of their bitter taste and bitter aftertaste; further, there were found noticeable differences in the bitter taste and bitter aftertaste depending on the part of beet roots (Tab. 2). Therefore, the null hypothesis ( $H_0$ ), i.e. the lack of differences in the perception of bitter taste and bitter aftertaste ( $F = 9.8995$ ) among the evaluated red beet cultivars ( $F = 9.6776$ ), was rejected. The probability of committing an error by rejecting the null hypothesis is 1 % ( $p = 0.013146$ ) for the rejection of  $H_0$  because of the lack of differences in the perception of the bitter taste among the cultivars. In turn, the probability of committing an error when rejecting  $H_0$  because of the lack of differences between the bitter taste and the bitter aftertaste of different cultivars is 2 % ( $p = 0.025484$ ).

Table 2. Results of statistical analysis (ANOVA of main effects at  $p = 0.05$ ) to confirm significant differences in the perception of bitter taste and bitter aftertaste of 6 red beet cultivars evaluated

Tabela 2. Wyniki analizy statystycznej (ANOVA efektów głównych, na poziomie istotności  $p = 0,05$ ), potwierdzające istotne różnice w odczuwaniu smaku i posmaku gorzkiego w 6 ocenianych odmianach buraka ćwikłowego

Effect / Efekt	Univariate significance tests for results of sensory evaluation of bitter taste and bitter aftertaste / Jednowymiarowe testy istotności wyników oceny sensorycznej smaku i posmaku gorzkiego				
	SS	Df	MS	F <sup>b</sup>	p <sup>c</sup>
Free term	37.10083 <sup>a</sup>	1 <sup>a</sup>	37.10083 <sup>a</sup>	135.6520 <sup>a</sup>	0.000082 <sup>a</sup>
Red beet cultivars	13.23417 <sup>a</sup>	5 <sup>a</sup>	2.64683 <sup>a</sup>	9.6776 <sup>a</sup>	0.013146 <sup>a</sup>
Bitter taste and aftertaste	2.70750 <sup>a</sup>	1 <sup>a</sup>	2.70750 <sup>a</sup>	9.8995 <sup>a</sup>	0.025484 <sup>a</sup>
Error	1.36750	5	0.27350	-	-

Explanatory notes / Objaśnienia:

SS – sum of squares / suma kwadratów; Df – degrees of freedom / stopnie swobody; MS – mean square / średnia kwadratów; a – values highlighted in red / wartości podświetlone na czerwono; b – statystyka F służy do weryfikacji hipotezy istotności całego modelu / F statistic is used to verify the hypothesis on significance of the whole model; c – p-level of probability / p poziom prawdopodobieństwa

The results of sensory analyses of the individual cultivars and red beet parts confirmed that the roots of all the cultivars tested had a bitter taste, which was perceptible at varying intensity levels.

The bitter taste was defined as a predominant taste attribute of three out of six red beet cultivars, while in the case of all the cultivars tested, the intensity of the bitter taste (aftertaste) of the skin was stronger than that of the flesh of roots (Fig. 2, 3).

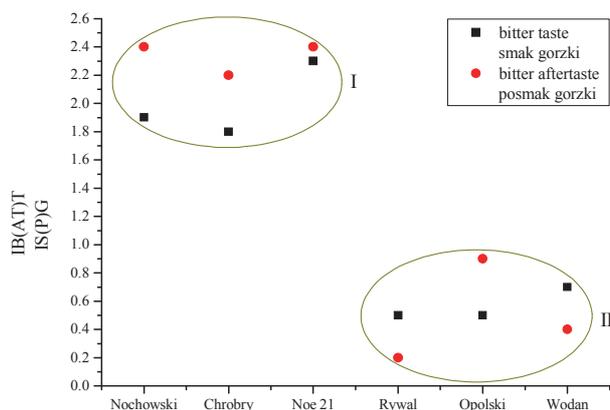


Fig. 2. Red beet cultivars (root flesh) grouped according to bitter taste and aftertaste intensity level BT(AT)IL

Rys. 2. Odmiany buraka ćwikłowego (miąższ) pogrupowane pod względem intensywności smaku i posmaku gorzkiego IS(P)G

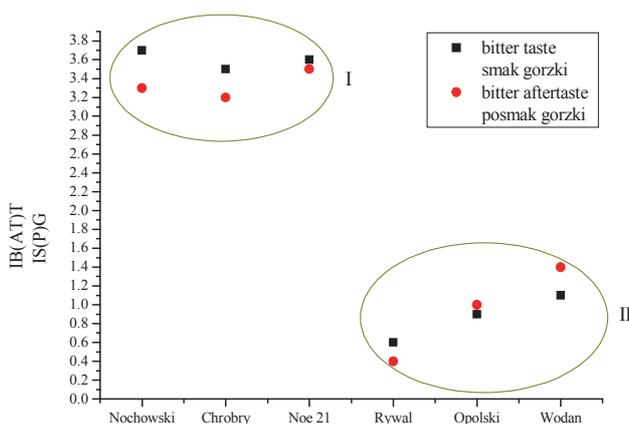


Fig. 3. Red beet cultivars (root skin) grouped according to bitter taste and aftertaste intensity level BT(AT)IL

Rys. 3. Odmiany buraka ćwikłowego (skórka) pogrupowane pod względem intensywności smaku i posmaku gorzkiego IS(P)G

The flesh and skins of the ‘Nochowski’, ‘Chrobry’, and ‘Noe 21’ cultivars (Group I) manifested the strongest bitterness notes, whereas the roots of red beet of the ‘Wodan’, ‘Opolski’, and ‘Rywal’ cultivars (Group II) had a strongly intense sweet taste and, at the same time, the intensity level of their bitter taste was rated very low.

The sensory evaluation of the flesh and skins of red beet roots made it possible to distinguish a group of the most bitter cultivars; this group consisted of the ‘Nochowski’, ‘Chrobry’, and ‘Noe 21’ cultivars. The cultivars in this group had the highest intensity level of their bitter taste and aftertaste. The red beet cultivars evaluated can be divided into two groups: in Group I, there are the beet root cultivars having a strong level of bitter taste (aftertaste) and in Group II, there are those showing a minimal intensity level of bitter taste (aftertaste). The mean score rating for the perception of the bitter taste of the ‘Nochowski’, ‘Chrobry’, and ‘Noe 21’ cultivars was 1.9 as for the flesh and 3.6 as for the skin.

The other red beet cultivars (Group II) had low scores for the intensity level of their bitter taste ranging from 0.5 to 0.7 as for the flesh and from 0.6 to 1.1 as for the skin. Based on the univariate analysis of variance ( $p = 0.05$ ), statistically significant differences were found between the bitter taste and the bitter aftertaste of the ‘Nochowski’, ‘Chrobry’, and ‘Noe 21’ cultivars compared to the ‘Rywal’, ‘Opolski’, and ‘Wodan’ cultivars; the latter cultivars showed a minimal intensity level of their bitter taste (aftertaste).

Another interesting outcome is linked with mutual relationships between the sweet taste and the bitter taste (aftertaste) of red beet cultivars in Group I (strongly bitter) and Group II (slightly bitter) (Fig. 4, 5). Those Figures point to a predominance of the bitterness attributes over the sweet taste attributes of the skins of the cultivars in Group I, whereas the cultivars in Group II show a reverse relationship.

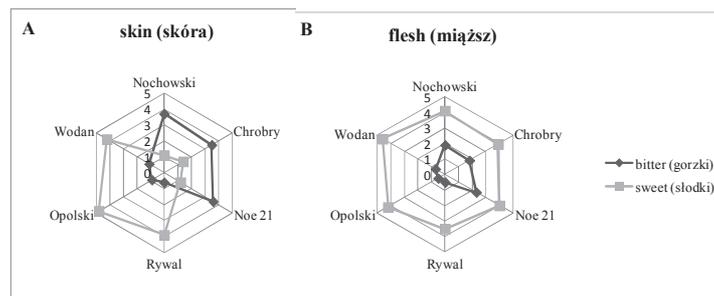


Fig. 4. Relationship between sweet and bitter taste of skins (A) and flesh (B) of evaluated red beet cultivars

Rys. 4. Relacja pomiędzy smakiem słodkim i gorzkim w skórkach (A) i mięszu (B) badanych odmian buraka ćwikłowego

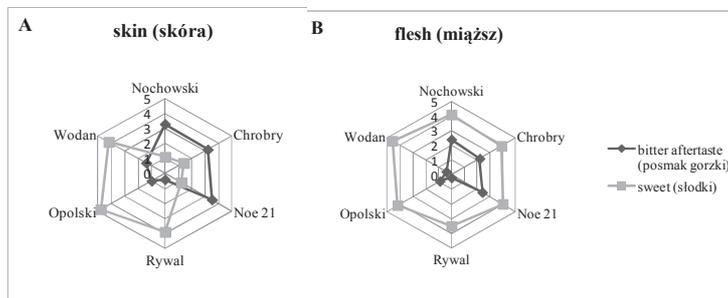


Fig. 5. Relationship between sweet and bitter aftertaste of skins (A) and flesh (B) of evaluated red beet cultivars

Rys. 5. Relacja pomiędzy smakiem słodkim i posmakiem gorzkim w skórkach (A) i mięszu (B) badanych odmian buraka ćwikłowego

In the flesh of all the cultivars evaluated, the sweet taste predominates over the bitterness taste (Fig. 4B, 5B). However, it is worth stressing that the flesh of the ‘Nochowski’, ‘Chrobry’, and ‘Noe 21’ cultivars has a moderately intense level of bitterness and the bitterness attributes of the ‘Wodan’, ‘Opolski’, and ‘Rywal’ cultivars are practically imperceptible.

A strong accumulation of bitterness in the ‘Nochowski’, ‘Chrobry’, and ‘Noe 21’ cultivars was the basis for selecting one of them for the purpose of identifying and separating potentially bitter taste components, i.e. triterpene saponins. The saponin extracts were separated from the lyophilisates in the whole roots (skin and flesh together) of the ‘Nochowski’ cv. 100 g of lyophilisate was got out of 1 kg of fresh red beet roots, which was equivalent to 10 % lyophilisation efficiency.

After removing fats, the bitter skins of beet roots were dried and extracted with methanol; next, they were evaporated and, subsequently, a fraction containing the saponin mixture was suspended in water, loaded onto LiChroprep RP 18 (40-63 $\mu$ m, Merck, Germany) and washed with 80 % methanol. A solid phase extraction (SPE) was applied to re-purify the compounds isolated. The lyophilised plant material (1 g) yielded 15 mg of fraction of the saponin mixture. The content of saponins in 1 kg of fresh material was 0.15 % in terms of fresh weight of the roots of red beet of the ‘Nochowski’ cv.

Structural analyses of saponins present in crude extracts from red beet roots (cv. ‘Nochowski’ cv.) were previously reported [6]. All the above described procedures accomplished resulted in detecting 41 saponins, which were pre-identified. The pre-identified saponins were glycosides of four different sapogenins, oleanolic acid, hederagenin, akebonoic acid, and gypsogenin (Fig. 1.). The aglycones in the triterpene saponins were substituted at C-3 or C-28 with one and two sugar chains; this resulted in a complex multi-component mixture derived from the roots of red beet (*Beta vulgaris*

L.). The compounds isolated had molecular ions with  $m/z$  ranging from 763 to 1117 [M-H]. The majority of the saponins detected in the mixture of triterpene saponins were derivatives of the oleanolic acid found in 27 pre-identified saponins, whereas the akebonoic acid was found in 10 compounds, the hederagenin in 6 saponins, and the gypsogenin only in one saponin [6].

After the group of bitter compounds mixture was isolated, a subsequent sensory evaluation was performed with the use of the same analyses already applied to identify the group of compounds responsible for a high intensity level of bitter taste and bitter aftertaste of beetroot. For this purpose, the members of the sensory panel tested three independent SPE-purified samples ( $E_0$ ,  $E_I$ , and  $E_{II}$ ), which were evaporated to dryness and which contained the mixtures of triterpene saponins obtained from the roots of the 'Nochowski' cv.

The results of the analyses confirmed that all the tested samples of saponin extracts had a strong bitter taste. While analysing a relative intensity level of bitterness of the saponin extracts, the  $E_I$  extract was classified, in terms of the intensity of bitter taste (ISG), as being comparable to the quinine solutions at concentrations of  $C_2 = 6.6 \times 10^{-3} \text{ g/dm}^3$  and  $C_3 = 9.54 \times 10^{-3} \text{ g/dm}^3$ . The ISG of the  $E_I$  extract was, as a rule, identical to the quinine solution at  $C_2 = 6.6 \times 10^{-3} \text{ g/dm}^3$ .

The analyses conducted proved that the factors responsible for the bitter taste of red beet might be highly complex and, among them, the triterpene saponins were the key components. Further studies on the bitter taste of red beets should be focused on the explanation which of the triterpene saponins, and to what extent, are responsible for the bitterness attributes of red beets. The performed spectral analysis of the saponin extracts confirmed that there were saponin compounds therein [18].

The character and contents of those compounds constitute an important criterion when assessing the quality of beetroot products.

## Conclusions

1. The red beet cultivars: 'Nochowski', 'Chrobry', 'Noe 21', 'Rywal', 'Opolski', and 'Wodan' as well as their individual parts showed statistically significant differences in their bitter tastes, which was perceptible at various intensity levels depending on the cultivar and root parts.
2. Statistically significant differences were found among the bitter tastes and bitter aftertastes of the 'Nochowski', 'Chrobry', and 'Noe 21' cultivars compared to the 'Rywal', 'Opolski', and 'Wodan' cultivars. The first three cultivars showed a high intensity level of the bitter taste and aftertaste and the other three cultivars had a minimal intensity level of those attributes.

3. The bitter 'Nochowski', 'Chrobry', and 'Noe 21' red beet cultivars were characterised by a strong sensation of bitter aftertaste; the bitter aftertaste of the skin of their roots had a higher intensity level compared to that of their flesh.
4. The cultivars with a high strong intensity level of their bitter taste had, at the same time, a lower intensity level of the sweet taste. As for the other cultivars, a strong intensity level of the sweet taste was reported.
5. The strongest bitterness attributes were found in the case of the roots of the 'Nochowski' cv. and the saponin mixture isolated from this specific cultivar at a concentration of  $C = 1.3515 \text{ g/dm}^3$  showed the intensity level of the bitter taste that was comparable to that of the quinine solutions at concentrations of  $C_2 = 6.6 \times 10^{-3} \text{ g/dm}^3$  and  $C_3 = 9.54 \times 10^{-3} \text{ g/dm}^3$ .

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## SAPONINY TRITERPENOWE JAKO GORZKIE SKŁADNIKI BURAKA ĆWIKŁOWEGO

### Streszczenie

Wiele odmian buraka ćwikłowego (*Beta vulgaris* L.) jest cenionych ze względu na plenność, zawartość składników odżywczych oraz barwników z grupy betalain, wykazujących silne właściwości przeciwutleniające. Z drugiej strony silna goryczka korzeni szeregu odmian buraków ćwikłowych jest częstą

przyczyną ich nieakceptowania przez konsumentów. Dotychczas przedstawiane prace zbyt ogólnie charakteryzują zróżnicowanie odmian buraka ćwikłowego pod względem cech goryczkowych. Obecnie niewiadome jest również, które z naturalnie występujących metabolitów wtórnych korzeni buraka ćwikłowego odpowiadają za jego smak i posmak gorzki. Celem pracy było określenie grupy związków powodujących wysoką intensywność smaku i posmaku gorzkiego buraka ćwikłowego. Pierwszy etap badań dotyczył selekcji najbardziej gorzkich odmian buraków na podstawie ich cech smakowych (miąższu i skórek sześciu odmian ('Nochowski', 'Chrobry', 'Noe 21', 'Rywal', 'Opolski' i 'Wodan') świeżych korzeni buraka ćwikłowego. Charakterystyka sensoryczna badanej grupy buraków ćwikłowych wykazała, że najbardziej intensywną goryczką zarówno w miąższu, jak i w skórce odznaczają się odmiany 'Nochowski', 'Chrobry' i 'Noe 21'. Z kolei odmiany takie, jak: 'Rywal', 'Opolski' i 'Wodan' charakteryzowały się stosunkowo niską intensywnością cech goryczkowych. Mieszaninę triterpenowych saponin wyizolowano z liofilizatu korzeni odmiany 'Nochowski', zaklasyfikowanej wg ocen sensorycznych do grupy o najsilniejszych cechach goryczkowych. Wyniki badań sensorycznych wyizolowanej mieszaniny saponin o stężeniu  $C = 1,3515 \text{ g/dm}^3$  z odmiany 'Nochowski' potwierdziły, że grupa tych związków wykazuje silny smak gorzki, porównywalny do roztworu chininy o stężeniach  $C_2 = 6,6 \times 10^{-3} \text{ g/dm}^3$ . Dowiedziono również, że badane ekstrakty buraka ćwikłowego stanowią mieszaninę złożoną wyłącznie ze związków saponinowych o zróżnicowanej strukturze chemicznej.

**Słowa kluczowe:** burak ćwikłowy, saponiny triterpenowe, smak gorzki, posmak gorzki ☒