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COMPARISON OF SELECTED METHODS APPLIED FOR THE EVALUATION OF THERMAL PROCESSING EFFICIENCY OF CHICKLING VETCH SEED

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Chickling vetch seeds (raw or autoclaved at 112°C for 10, 20 or 30 min and at 121°C for 10, 20, 30, 60, 90 or 120 min) were applied as an object for comparison of three tests used for the evaluation of seed thermal processing efficiency (i.e. trypsin inhibitor activity – TIA, cresole red index – CRI, and protein dyspersion index – PDI) with a method worked out by the authors and called bromocresole purple index (BCPI). Seed autoclaving caused a decrease in TIA (from 23.13 TUI/mgDM for a raw sample to 2.00 TUI/mgDM for a sample autoclaved at 121°C for 120 min) and in PDI (from 90.46% for a raw sample to 15.63% for a sample autoclaved at 121°C for 120 min), as well as an increase in CRI (from 1.48 mg/gDM for a raw sample up to 4.82 mg/gDM for a sample autoclaved at 121°C for 120 min) and in BCPI (from 15.10 mg/gDM for a raw sample up to 38.09 mg/gDM for a sample autoclaved at 121°C for 120 min). All compared estimation ways appeared to be useful at studied seed evaluation; however, due to great sensibility (c=0.19), detectability (d=327.6s), distinguishability (r=94%) and low time-consumption (h=1.5h), the application of BCPI tests was found the most proper. High correlation coefficients between BCPI and other evaluation methods (BCPI & TIA – r = -0.84; BCPI & CRI – r=0.94; BCPI & PDI – r=-0.75) point out to the possibility of replacing the time- and cost-consuming determinations (e.g. antitrypsin activity – TUI or determination of protein dispersion index – PDI) with a simple, rapid and cheap bromocresole purple index – BCPI.

Symbols and abbreviations: β [%] – maximum relative determination error, **BCPI** – bromocresole purple index, χ – test sensitivity, **CRI** – cresole red index, **CV** – variation coefficient, δ – detectable minimum, η – test time-consumption, **r** – correlation coefficient, **NIR** – least significant differences, **MD** – mean value, **MTN** – the mass of thousand seeds, **N** – the number of compared results, **PDI** – protein dispersibility index, ρ – sample discrimination, **SD** – standard deviation, **TIA** – trypsin inhibitor activity, **TUI** – trypsin inhibitor unit, ω – test detectability.

INTRODUCTION

Chickling vetch, due to its relatively high seed yield with high nutritive value and low soil, weather and agrotechnical requirements, can be an important element in the fight against starvation in many poor Asian and African countries [Dziamba, 1997].

The high nutritive value of these seeds results first of all from relatively high protein content in dry matter (24–36%), advantageous fatty acid composition of the oil (comparable with soybean oil), a low level of crude fiber (with profitable fraction composition), and desirable content of minerals [Grela & Winiarska, 1997; Grela & Skórnicki, 1997; Hanczakowski *et al.*, 1997]. According to Hanczakowski *et al.* [1997], sufficient amounts of amino acids to meet the requirements of the majority of farm animals occur in chickling vetch's seed protein, and high levels of lysine additionally favour the composition of fodder constituents achieved from the seeds in mixtures with cereals. However, technological processes (particularly different heating treatments) that before consumption aim at elevating the availability of nutritional components in the seeds, determine the usability of chickling vetch seeds as a component of foodstuffs or feed [Grela *et al.*, 1997; Zdunek, 1997].

The technological processes, elevating the availability of raw material nutrients, affect the improvement of rheological traits and decrease the antinutritive factor activity. At the same time, they should not trigger any other unfavourable processes, *e.g.* decreasing the digestibility of excessively denatured proteins.

At present, the industrial processing of chickling vetch seeds is frequently performed with HTST-type (high temperature – short time) technological lines with high efficiency, short material retention time and high processing temperature. Therefore, it is necessary to properly choose the parameters of seed thermal processing (type, time, temperature) and to ensure an efficient quality management system of the achieved processed products in order to estimate the semi-products and final products under the simple conditions of an industrial laboratory. Today, evaluation methods consisting in adapting selected methods of antinutritive factor activity estimation (*e.g.* trypsin inhibitor activity TIA [Kakade *et al.*, 1974]) are not useful for current quality management on technological lines, due to the excessive time required, cost and machine-usage as well as complicated analytical procedures. On the other hand, the lack of sensitivity, detectability and method's precision often rules out rapid tests based on utilization of particular reagents (*e.g.* cresole red index – CRI – adapted of soybean seed evaluation methods) [Norm ISO 5514-1979(E)] or on selected properties of seed protein (*e.g.* protein dispersion index – PDI) [Volkert & Klein, 1979].

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The study was targeted to compare several selected methods for the evaluation of chickling vetch seed thermal processing (trypsin inhibitor activity – TIA, cresole red index – CRI, and protein dispersibility index – PDI) with a test elaborated by the authors and referred to as bromocresole purple index – BCPI.

MATERIAL AND METHODS

The study was performed using chickling vetch seeds classified as fine (1000-kernel weight of 92 g) [Dziamba, 1997]. The seeds contained 87.32% of dry matter, 26.47% DM of crude protein, 1.22% DM of crude fat, 5.42% DM of crude fiber and 3.00% of ash. Measurements of 1000-kernel weight, crude protein, crude fat, crude fiber and ash as well as seed's humidity were carried out according to obligatory norms and by conducting three independent replications for every trait measured.

A part of the seeds obtained were not subjected to thermal treatment, they were further referred to as "crude seeds". Other seeds were divided into 9 samples (50 g each), transferred into glass cylinders of 250 mL volume and covered with Petri dishes, in order to separately place each of them in the geometrical center of ASVE-type vertical laboratory sterilizer. They were then subjected to autoclaving under conditions of one of the variants of thermal processing intensity (*i.e.* at 112°C for 10, 20 or 30 min, or at 121°C for 10, 20, 30, 60, 90 or 120 min).

For crude seeds, every sample was subjected to autoclaving (in 5 replications) in order to evaluate the influence of thermal processing on some seed protein traits: trypsin inhibitor activity – TIA [Kakade *et al.*, 1974], protein dispersion index – PDI [Norm AACC 46-24 1968], cresole red index – CRI [Norm ISO 5514 1979E], and bromocresole purple index – BCPI [Szmigielski, 1999; 2002].

Measurement of trypsin inhibitory activity (TIA) according to the method of Kakade *et al.* [1974] is based on the evaluation of a part of trypsin activity that is blocked due to the reaction with buffered extract containing the enzyme inhibitors achieved from the seed sample. This requires the application of synthetic BAPA substrate (*i.e* N-α-benzoilo–DL–arginino–p–nitroanilide) [Kakade *et al.*, 1974].

As a result of the trypsin-BAPA reaction, yellow p-nitroanilin is formed (the absorbance measured at a wavelength of 410 nm is proportional to its concentration).

The protein dispersion index (PDI) is one of the methods for evaluating the nutritional usefulness of foodstuff or fodder components subjected to heating (*e.g.* leguminous plant seeds), in which a post-denaturation decrease of protein solubility is given as an intensity indicator. The PDI measurement according to AACC norm [AAC:46-24, 1968] is based on the estimation of the percentage of sample protein that under experimental conditions (*i.e.* 10-min homogenization at 25°C and 8500 rad/min centrifuging as well as 10-min centrifugation at 36.3xg) is dissolved or remains as a dispersed, stable, water emulsion in a solution. Protein content in the sample before homogenization and the amount remaining in the dispersion was estimated using the Kjeldahl method.

Cresole Red Index (CRI) is a physicochemical method for the evaluation of heating intensity based on the application of acid-base indicator (3',3"-dimethylo-phenolosulphophthalein – commonly called cresole red) as an active substrate that is sorbed on the surface of properly ground product sample (*e.g.* seeds) from a working solution containing 0.10 mg/mL of the reagent. The CRI measurement according to ISO norm [ISO: 5514, 1979E] consists in determination of the difference between contents of the active substance before and after contact with the sample.

The essence of the BCPI method is to measure the amount of acid-base indicator (5',5"–dibromo–3',3"–dimethylophenolosulphophthalein – commonly called bromocresole purple) that is bound by 1 g of properly ground seeds subjected to reaction with the working solution containing the active substance.

A working solution at a concentration of 0.13 mg/mL (applied for BCPI determinations) was prepared in a measuring flask (1 L capacity) by dissolving the 130 mg bromocresole purple (Merck 1992/93 No 3025) in 40 mL of 0.1 mol/L NaOH, adjusted to 1 L with 0.1 mol/L HCl and stirred until a homogenous concentration was achieved.

Seed samples were ground, sieved through a 0.2-mm mesh and 100-mg aliquots were placed in conical flasks (100 mL capacity), 50 mL of bromocresole purple solution was added (0.13 mg/mL concentration) and stirred for 30 min using a magnetic stirrer. Following this, the solution was centrifuged for 5 min at 44.8xg and 1 mL of extract was placed in a tube containing 20 mL of 0.02 mol/L NaOH. The solution was stirred and after 10 min absorbance was measured at 589 nm wavelength against distilled water as a reference (A_b – equation 1):

$$BCPI = (A_0 - A_b) \times C \times V / A_0 \times M \quad (1)$$

where: BCPI – amount of absorbed dye [mg/g], A_0 – absorbance of the mixture achieved due to mixing 1 mL of working solution with 20 mL of 0.02 mol/L NaOH; A_b – absorbance of the mixture achieved due to mixing 1 mL of extract (after centrifuging) with 20 mL of 0.02 mol/L NaOH; C – dye solution concentration [mg/mL] (0.13 mg/mL); V – volume of the dye solution added to the sample [mL] (50 mL); and M – weight of the sample subjected to tests [g] (0.1 g).

Absorbance of the working solution was measured analogously (A_0 – equation 1), but instead of the extract (after centrifuging), 1 mL of working solution (0.13 mg/mL concentration) was taken.

The amount of adsorbed active substance (bromocresole purple) was calculated using Equation 1 as the difference between its content in the solution before and after the contact with ground seed sample.

The following items were accepted as the criteria for comparison of the methods used: sensitivity (χ), detectability (ω), discrimination (ρ), and time-consumption (η) of tests (BCPI, CRI, PDI, TIA). Correlation coefficients (r) between results were also calculated [Oktaba, 1986]. Sensitivity of tests (χ) was estimated as the absolute value of slope of a straight line in linear regression equation of test's value as a seed heating time function (taking values for raw seeds and seeds autoclaved at 121°C and for 120 min as the basis of correlation) [Gawęcki & Wagner, 1982; Szmigielski *et al.*, 2001]. Test detectability (ω) was expressed with detectable minimum (δ) and calculated as the shortest time

of thermal processing required for the change of measured value by a unit equaled to maximum relative error of measurement [Szmigielski *et al.*, 2001]. The maximum relative error of measurement (β) was calculated by means of differential calculus, and maximum test's result was accepted as the calculation basis [Sielanko & Sowa, 1994; Szmigielski *et al.*, 2001]. Sample discrimination (ρ) was calculated as the significance of differences between test results for samples differing in autoclaving time at the same thermal processing temperature and is expressed as the percentage of significant relations in reference to all checked relations. The significance of differences for the results obtained was estimated with an analysis of variance (5% significance level) and the least significant differences (NIR) – with Tukey's (NIR) [Oktaba, 1986].

Test time-consumption (η) was expressed as the time necessary for making one replication of a test according to analytical procedures for a single seed sample.

RESULTS AND DISCUSSION

Autoclaving of chickling vetch seeds at different times and temperatures affected the significance of differentiation of the samples achieved.

The trypsin inhibitor activity (TIA) of seed samples was highly dependent on time and temperature of autoclaving. The level of the anti-nutrient factor for crude seeds (23.13 TUI/mg_{DM}, Table 1) was relatively low and similar to the results achieved by Grela *et al.* [1997], Hanczakowski *et al.* [1997] as well as Dziamba *et al.* [1996]. It should be noted that studies carried out so far have also reported on great differentiation of seeds in this respect. For instance, Troszyńska *et al.* [1997] achieved results higher (than those of the presented study) by about 35% (*i.e.* 31.20 TUI/mg_{DM}) for Derek cv.

The extreme results (achieved due to investigation of samples autoclaved at 121°C for 120 min and raw ones) differed over 11-fold in TUI level (Table 1).

Despite such great dispersion of results, the reduced usefulness of the method when evaluating the samples of high and excessively high heating intensity (autoclaved at 121°C for 30, 60, 90 and 120 min is remarkable, which contributes to a decrease in their discrimination. The great dispersion of TIA results combined with their low value for samples favours lower precision (measured with variability coefficient – CV) (Table 1). Analogous results, *i.e.* a decrease in TIA determination and a decrease in sample distinguishing for intensive autoclaving (121°C for 30, 60, 90 and 120 min) were also observed by Szmigielski [2002], as well as Szmigielski and Matyka [2002] in similar studies on soybean and bean seeds, respectively.

PDI values achieved for the chickling vetch seeds tested indicate the great dependence of the test results on sample heating intensity. The difference between extreme PDI values (achieved for raw samples and samples autoclaved for 120 min at 121°C) was five times higher than the lowest result (Table 2). However, despite such great dispersion of results, the disproportion in PDI changes appearing through indiscrimination and low measurement precision for samples subjected to intensive thermal processing (autoclaved for 60, 90 and 120 min at 121°C), are quite obvious.

A decrease in sample identification and PDI determination precision in respect to autoclaved seeds was also achieved in analogous studies performed for soybean [Szmigielski, 2002] and bean [Szmigielski & Matyka, 2002].

Autoclaving of chickling vetch seeds (under different time and temperature conditions) also affected significant changes in cresole red index (CRI).

Seeds autoclaved at 121°C for 120 min were characterized by three times higher CRI values (as compared to the raw ones), and the difference between these results is over 200% of the lower result (achieved for the raw sample) (Table 3).

The CRI measurements were characterized by their relatively high precision and their variability coefficient CV was about twice as high for results of the samples at low heating

TABLE 1. Trypsin inhibitor activity (TIA) [TUI/mg d.m] for chickling vetch seed samples.

Type of processing	Processing time (min)					
	10	20	30	60	90	120
Autoclaving 112°C	18.69 ± 2.15^{a}	12.51 ± 2.12^{b}	5.15 ± 2.00^{c}			
	(11.50%)	(16.95%)	(38.83%)			
Autoclaving 121°C	14.41 ± 2.12^{a}	10.59 ± 2.00^{b}	3.10 ± 1.00^{c}	$2.60\pm1.32^{c,d}$	$2.10\pm1.25^{c,d,e}$	$2.00\pm1.05^{c,d,e,f}$
	(14.71%)	(18.89%)	(32.26%)	(50.77%)	(59.52%)	(52.50%)
Control	23.13±2.25 (9.72%)					

(N = 45, NIR =3.8, $NIR_{Autoclav.\ 121°C}$ = 3.07). The results whose differences are not significant (Tukey's least significant differences – NIR) are denoted with identical letters in superscripts.

TABLE 2. Protein dispersibility index (PDI) [%] for chickling vetch seed samples.

Type of processing	Processing time (min)					
	10	20	30	60	90	120
Autoclaving 112°C	86.19 ± 5.05^{a}	75.15 ± 4.15^{b}	59.83 ± 4.02^{c}			
	(5.86%)	(5.52%)	(6.72%)			
Autoclaving 121°C	85.14 ± 3.86^{a}	65.32 ± 4.00^{b}	42.63 ± 4.23^{c}	22.15 ± 4.16^{d}	$18.28\pm4.00^{d,e}$	$15.63\pm3.45^{d,e,f}$
	(4.53%)	(6.12%)	(9.92%)	(18.78%)	(21.88%)	(22.07%)
Control	90.46±5.25 (5.80%)					

(N = 45, NIR = 8.28, $NIR_{Autoclav.121°C}$= 7.38) The results whose differences are not significant (Tukey's least significant differences – NIR) are denoted with identical letters in superscripts.

intensity than for those which were strongly heated (autoclaved for 60, 90 and 120 min at 121°C) (Table 3). It indicates the greater usefulness of the CRI test (as compared to TIA and PDI methods) for the evaluation of intensively heated samples. Statistical estimation of the significance of differences betweenthe CRI results points to the indiscrimination of variants of similar heating both with low and high intensity (Table 3). Szmigielski [2002] as well as Szmigielski and Matyka [2002] drawn similar conclusions on the basis of their earlier studies on soybean and bean seeds.

Results obtained by means of the bromocresole purple index (BCPI) for the chickling vetch seeds examined depended greatly on heating intensity. Extreme BCPI values (minimum – achieved for raw samples; maximum – for samples autoclaved at 121°C for 120 min) differed 2.5-fold, and the difference between them was *ca*. 150% of the minimum (Table 4).

The BCPI test was characterized with very high measurement precision both in relation to samples with high and low heating intensity, which obviously affected the discrimination of almost every heating variant tested (only differences between the results achieved for samples autoclaved at 112°C for 10 and 20 min were insignificant – discrimination 94%) (Table 5). In similar studies carried out by Szmigielski and Matyka [2002] on bean seeds, identical result of BCPI test identification was achieved (94%), and in analogous experiments performed by Szmigielski [2002] on soybean seeds, the criterion was at an 82% level.

High discrimination of samples achieved using the BCPI method is confirmed by the best detectability (among compared methods) and the relatively high sensitivity of measurements – BCPI is only worse than PDI, although it is about three times less sensitive (Table 5). It should be noted that in the research by Szmigielski and Matyka [2002], as well as Szmigielski [2002], BCPI was characterized with the highest or one of the highest distinguishing, although its sensitivity was (similar to this study) several times lower (2.5–4.0) than for the PDI method.

Nevertheless, the high sensitivity of the PDI method does not guarantee good discrimination of samples. It could be due to the low detectability resulting from the high maximum relative error (β), low determination precision and disproportionate test value changes in relation to the intensity of the heating factor (observed for samples with high heating intensity).

Szmigielski [2002], Szmigielski and Matyka [2002], as well as Szmigielski *et al.* [2001] achieved low PDI test detectability (high δ) along with a relatively high maximum relative error (β) of the determination.

The cresole red index (CRI) is characterized with comparable discrimination to the protein dispersion index (PDI), though the CRI method is about 20-fold less sensitive compared to PDI (Table 5).

The proportionality of CRI test's changes to the changes in seed heating intensity as well as the relatively high detectability of the method determines its unexpectedly great discrimination (Table 5). Similar generalizations are included in papers by Szmigielski [2002], as well as Szmigielski and Matyka [2002].

Slight sample discrimination, caused mainly by low usefulness of the test for samples with high heating intensity and low detectability (ω) resulting from high β, are the disadvantages of the TIA method (Table 5). Differences of ω

TABLE 5. Detectability, sensitivity, discrimination time-consumption and maximum relative determination error of tests in the evaluation of efficiency of chickling vetch seed thermal processing.

Criteria for test comparison	Type of test			
	BCPI	CRI	PDI	TIA
Detectability (δ) [s]	327.60	511.20	703.80	646.20
Sensitivity (χ)	0.19	0.03	0.62	0.18
Time-consumption (η) [h]	1.50	1.50	4.50	2.50
Discrimination (ρ) [%]	94	83	83	67
Maximum relative determination error β [%]	2.43	4.92	8.09	8.20

TABLE 3. Cresole red index (CRI) [mg/g_{DM}] for chickling vetch seed samples.

Type of processing	Processing time (min)					
	10	20	30	60	90	120
Autoclaving 112°C	1.90 ± 0.24^{c}	$2.42\pm0.20^{a,b}$	2.76 ± 0.25^{a}			
	(12.63%)	(8.26%)	(9.06%)			
Autoclaving 121°C	$2.69\pm0.26^{d,e,f}$	$2.79\pm0.25^{d,e}$	2.93 ± 0.20^{d}	3.66 ± 0.31^{c}	$4.53\pm0.30^{a,b}$	4.82 ± 0.30^{a}
	(9.67%)	(8.96%)	(6.83%)	(8.47%)	(6.62%)	(6.22%)
Control	48±0.22 (14.86%)					

(N = 45, NIR = 0.46, $NIR_{Autoclav.\ 121°C}$= 0.53) The results whose differences are not significant (Tukey's least significant differences – NIR) are denoted with identical letters in superscripts.

TABLE 4. Bromocresole purple index (BCPI) [mg/g_{DM}] for chickling vetch seed samples.

Type of processing	Processing time (min)					
	10	20	30	60	90	120
Autoclaving 112°C	$15.43\pm0.80^{b,c}$	16.70 ± 0.75^{b}	18.41 ± 0.76^{a}			
	(5.18%)	(4.49%)	(4.13%)			
Autoclaving 121°C	18.20 ± 0.82^{f}	19.86 ± 0.83^{e}	21.48 ± 0.80^{d}	27.08 ± 0.82^{c}	31.88 ± 0.92^{b}	38.09 ± 0.90^{a}
	(4.51%)	(4.18%)	(3.72%)	(3.02%)	(2.89%)	(2.36%)
Control	5.10±0.72 (4.77%)					

(N = 45, NIR = 1.55, $NIR_{Autoclav.\ 121°C}$= 1.65) The results, whose differences are not significant (Tukey's least significant differences – NIR) are denoted with identical letters in superscripts.

TABLE 6. Correlations between the traits studied (significance level $p<0.01$).

	TIA	CRI	BCPI	PDI
TIA	–			
CRI	- 0.96	–		
BCPI	- 0.84	0.94	–	
PDI	0.98	- 0.92	- 0.75	–

and β values in combination with disproportionate TIA changes for intensively heated samples cause the similar – in terms of sensitivity – BCPI and TIA tests to differ widely in sample's discrimination (Table 5). Szmigielski [2002], Szmigielski and Matyka [2002], as well as Szmigielski *et al.* [2001] in similar studies on soybean and bean seed samples also achieved relatively low TIA method detectability and discrimination.

Similarly, great differences referring to time-consumption are found between the BCPI and TIA methods (BCPI and CRI, as compared to TIA, require only half the time), although PDI (with three times higher time-consumption than BCPI and CRI) is characterized by extremely high value.

High correlation coefficients achieved for the results of the tests compared (Table 6) point to the interdependence between the compared traits and indicate the possibility of replacing them with one another when studying the properties of autoclaved chickling vetch seeds and calculating the achieved results. Masłowski *et al.* [2001], Szmigielski [2002], as well as Szmigielski and Matyka [2002] also achieved high correlation coefficients between BCPI, CRI, PDI and TIA traits.

The possibility of replacing the time-consuming, expensive and complicated analytical methods (PDI, TIA) by fast, cheap and simple BCPI and CRI ones is particularly important.

Due to higher sensitivity, detectability and discrimination, the BCPI test should be preferred when studying the heating intensity of chickling vetch seed.

CONCLUSIONS

1. Autoclaving of chickling vetch seeds caused changes in the properties of the proteins depending on heating intensity.

2. Protein dispersion index (PDI), trypsin inhibitor activity (TIA), cresole red index (CRI) and bromocresole purple index (BCPI) are useful for the evaluation of the autoclaving intensity of chickling vetch seeds.

3. High sensitivity, detectability and discrimination of BCPI test, as well as its low time-consumption facilitate precise control over technological process parameters.

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PORÓWNANIE WYBRANYCH METOD STOSOWANYCH DO OCENY SKUTECZNOŚCI OBRÓBKI TERMICZNEJ NASION LĘDŹWIANU SIEWNEGO

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Nasiona lędźwianu siewnego (surowe lub autoklawowane w temperaturze 112°C przez 10, 20 lub 30 minut i w temperaturze 121°C przez 10, 20, 30, 60, 90 lub 120 minut) zastosowano jako obiekt porównania trzech testów stosowanych do oceny skuteczności obróbki termicznej nasion (tj. aktywności inhibitora trypsyny – TIA, wskaźnika czerwieni krezolowej – CRI i współczynnika dyspersji białka – PDI) oraz metody opracowanej przez autorów i nazwanej wskaźnikiem purpury bromokrezolowej (BCPI). Autoklawowanie nasion spowodowało obniżenie TIA (z 23,13 TUI/ $mg_{s.m.}$ – dla próby surowej do 2,00 TUI/ $mg_{s.m.}$ – dla próby autoklawowanej w 121°C przez 120 minut) i PDI (z 90,46% – dla próby surowej do 15,63% – dla próby autoklawowanej w 121°C przez 120 minut) oraz wzrost CRI (z 1,48 mg/$g_{.S.M.}$ – dla próby surowej do 4,82 mg/$g_{S.M}$ dla próby autoklawowanej w 121°C przez 120 minut) i BCPI (z 15,10 mg/$g_{S.M.}$ – dla próby surowej do 38,09 mg/$g_{S.M}$ dla próby autoklawowanej w 121°C przez 120 minut). Wszystkie porównywane sposoby okazały się przydatne do oceny badanych nasion, jednak ze względu na dużą czułość (χ=0,19), wykrywalność (β=2,43%), rozróżnialność (ρ=94%) oraz małą czasochłonność (η=1,5h), zastosowanie testu BCPI okazało się najwłaściwsze.

Wysokie współczynniki korelacji pomiędzy BCPI i pozostałymi metodami oceny (BCPI i TIA – r = -0,84; BCPI i CRI – r = 0,94; BCPI i PDI – r = -0,75) wskazują na możliwość zastąpienia czasochłonnych i kosztownych oznaczeń (np. oceny aktywności antytrypsynowej - TUI lub oznaczenia współczynnika dyspersji białka – PDI) przez prosty, szybki i tani wskaźnik purpury bromokrezolowej – BCPI.