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EVALUATION OF ENZYMATIC BROWNING DEGREE OF EDIBLE POTATO TUBERS INDUCED BY HERBICIDES, MECHANICAL DAMAGES AND STORAGE BY MEANS OF INSTRUMENTAL AND SENSORY METHODS

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Key words: cultivar, herbicides, storage, discoloration potential, colorimetrical and visual method

The aim of this study was to compare two research methods (visual and homogenization- colorimetrically) in the evaluation of the degree of discoloration of potato tubers. Three cultivars of potato tubers (Victoria, Asterix, Dorota) were selected for analyses. They originated from a plantation at which the following agricultural practices were applied in the vegetative season: herbicides, constant fertilization (N : P_2O_5 : K_2O - 100 : 100 : 150 kg/ha) and 6-month storage (temperature: 4-6°C, relative air humidity: 90-95%). After the harvest, correlation coefficients for both methods were: -0.84 for non-bruised tubers and -0.69 for bruised ones. However, after the storage period, the correlation coefficients accounted for -0.54 and -0.73, respectively. The high correlation coefficient obtained between the two methods used to evaluate the enzymatic browning indicates that both the methods can be used interchangeably. The period of storage and mechanical damage of potato tubers have contributed to increased blackspot susceptibility of raw parenchyma. In contrast, the application of herbicides significantly differentiated darkening of the parenchyma of raw tubers only in the case of evaluation made with the colorimetric method.

INTRODUCTION

One of important characteristics of potato tubers, determining their functional value for consumption and processing, is enzymatic browning of raw parenchyma tissue. Stresses the potato tubers are exposed to during their vegetative season (drought, excessive precipitation, agrotechnical treatments, pathogens), harvest and transport (mechanical damage, bruising), and long-term storage are likely to deteriorate their quality, *i.e.* increase parenchyma susceptibility to darkening. Tubers tendency to darkening and post-damage spottening is a negative trait that limits or even disqualifies their usability as a raw material. A number of authors, including Rogozińska et al. [1986], Leszczyński [1994], Putz [1995], and Miller [1996], claim that the intensity of enzymatic browning – being to a considerable extent a genetically-modified trait - depends on the content of phenolic compounds and activity of polyphenolic oxidase in tubers. With access of oxygen, that enzyme oxidizes phenolic compounds occurring in a tuber (mainly an aromatic amino acid: tyrosine, and phenolic acids: chlorogenic, caffeic and others). Determination of such a significant characteristics as the susceptibility of tuber parenchyma to enzymatic browning may be carried out with various methods. One of them belong to rapid, cost-effective methods (visual), others require greater financial input and are more labor-consuming (colorimetric). The selection of an appropriate analytical method is crucial since it may determine agricultural product's classification for feeding purposes only. Therefore, the study was aimed at comparing results of the degree of enzymatic browning of potato tubers determined with visual and colorimetric methods, as affected by herbicides applied, mechanical damages and storage.

MATERIALS AND METHODS

Analyses were carried on potato varieties: 'Asterix', 'Dorota', and Victoria', obtained from field experiments carried out with the use of herbicides (Afalon, Stomp, Borax, Sencor+Titus) with constant fertilization (N : P_2O_5 : K_2O_100 : 100 : 150 kg/ha). Samples of potato tubers were collected from each plot in three replications directly after the harvest and after 6-month storage (temp. of 4-6°C, relative air humidity of 90-95%). Potato tubers were also subjected to mechanical damaging according to the method by Rogozińska et al. [1986], using a mechanical barrel. Enzymatic browning of tubers was evaluated with two methods: visual and colorimetric. Evaluation with the visual method proceeded as follows: 10 tubers from each sample were cut alongside into halves and stored for 4 h at a temperature of ca. 22°C. Use was made of a reversed 9-point Danish scale according to Henniger [1968], where: 1 denotes black tubers \rightarrow 9 denotes non-darkening tubers. The sensory analysis was carried out based on Polish Standards which specify conditions of carrying out sensory assessments and requirements for members of a sensory panel [ISO 8589:1998; ISO 6658:1998]. Determination of raw tubers darkening with the colorimetric method was carried out as follows: 6 tubers were collected from

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| Applied herbicides (I) | Cultivars (II) | After harvest | | | | After storage | | | |
|------------------------------|-------------------|---------------|----------------|------------------|----------------|------------------|----------------|------------------|----------------|
| | | Undamaged | | Damaged | | Undamaged | | Damaged | |
| | | V^1 | C ² | \mathbf{V}^{1} | C ² | \mathbf{V}^{1} | C ² | \mathbf{V}^{1} | C ² |
| Control | Victoria | 8.1 | 0.155 | 7.5 | 0.186 | 7.7 | 0.240 | 7.7 | 0.240 |
| | Asterix | 8.2 | 0.153 | 8.2 | 0.185 | 8.3 | 0.181 | 8.0 | 0.203 |
| | Dorota | 7.7 | 0.208 | 7.6 | 0.205 | 7.5 | 0.306 | 7.1 | 0.378 |
| Mean | | 8.00 | 0.172 | 7.77 | 0.192 | 7.83 | 0.242 | 7.60 | 0.274 |
| Afalon 50WG | Victoria | 8.5 | 0.116 | 7.7 | 0.190 | 7.7 | 0.232 | 7.7 | 0.255 |
| | Asterix | 8.2 | 0.156 | 8.4 | 0.186 | 7.8 | 0.210 | 7.8 | 0.230 |
| | Dorota | 7.7 | 0.263 | 7.4 | 0.313 | 7.9 | 0.296 | 7.2 | 0.366 |
| Mean | | | | | | | | | |
| | Victoria | 8.2 | 0.176 | 7.8 | 0.191 | 7.9 | 0.206 | 7.8 | 0.231 |
| Stomp | Asterix | 8.3 | 0.156 | 8.3 | 0.178 | 7.8 | 0.241 | 7.5 | 0.291 |
| - | Dorota | 7.8 | 0.231 | 7.6 | 0.270 | 7.6 | 0.261 | 7.0 | 0.385 |
| Mean | | | | | | | | | |
| | Victoria | 8.3 | 0.126 | 7.6 | 0.176 | 7.6 | 0.263 | 7.6 | 0.270 |
| Barox | Asterix | 8.0 | 0.176 | 7.3 | 0.253 | 7.9 | 0.251 | 7.5 | 0.288 |
| | Dorota | 8.0 | 0.231 | 7.3 | 0.355 | 7.3 | 0.321 | 7.2 | 0.355 |
| Mean | | | | | | | | | |
| Sencor | Victoria | 8.4 | 0.145 | 7.6 | 0.215 | 7.6 | 0.271 | 7.6 | 0.245 |
| +Titus | Asterix | 8.0 | 0.196 | 7.3 | 0.255 | 7.8 | 0.253 | 7.5 | 0.280 |
| + Inus | Dorota | 7.6 | 0.250 | 7.5 | 0.310 | 7.5 | 0.290 | 7.3 | 0.326 |
| Mean | | | | | | | | | |
| | Victoria | 8.28 | 0.143 | 7.64 | 0.186 | 7.73 | 0.235 | 7.68 | 0.249 |
| Mean | Asterix | 8.18 | 0.160 | 7.90 | 0.201 | 7.95 | 0.221 | 7.66 | 0.253 |
| | Dorota | 7.80 | 0.233 | 7.48 | 0.286 | 7.58 | 0.296 | 7.16 | 0.371 |
| Mean | | | | | | | | | |
| | Ι | n.i. | 0.006 | 0.26 | 0.018 | n.i. | 0.020 | n.i. | 0.031 |
| NIR _{p=0.05} | II | 0.30 | 0.006 | 0.30 | 0.013 | 0.39 | 0.013 | 0.43 | 0.080 |
| P 0.05 | I x II | n.i | n.i. | n.i. | n.i. | n.i. | n.i. | n.i. | n.i. |

TABLE 1. Darkening of raw potato tubers (damaged and undamaged) of the three cultivars examined after harvest and after storage as affected by herbicides applied during the vegetative season.

¹visual method,²colorimetric method

each sample and ground, then 25 g of the mash were homogenized by *ca*. 30 sec in 0.05 mol/L phosphate buffer with pH 6.5, and left for 24 h at a temperature of 22-24°C. Next, the filtrate obtained was centrifuged for 20 min at 12,000 rpm. At that rate, the acceleration acting on the sample is by *ca*. 60,000 times higher than the gravitational one. The centrifuged filtrate was diluted (1:3) and darkening was determined with a spectrophotometer at a wavelength of 475 nm [Dean *et al.*, 1993].

In the homogenization method, Dean *et al.* [1993] determined 5 categories of raw tubers parenchyma darkening, *i.e.*: 0.0-0.2 – resistant to darkening; 0.21-0.4 moderately resistant to darkening; 0.41-0.6 – moderately susceptible to darkening; 0.61-0.8 – susceptible to darkening; and >0.8 – very susceptible to darkening.

Results obtained were elaborated statistically by means of a two-way analysis of variance. The least significant difference (NIR_{0.05}) was calculated using a Tukey's test. Pearson's correlation coefficients were calculated additionally for a comparison of the analytical methods applied in the study.

RESULTS AND DISCUSSION

Results of ample research indicate that application of herbicides and their mixtures in potato cultivation does not exert any significant impact on increased enzymatic browning of tuber parenchyma after harvest, once determined visually [Ceglarek et al., 1990; Zarzecka et al., 1997; Zarzecka, 1998], which has been confirmed in the reported study. In contrast, a significant increase of darkening was observed in herbicide-treated tubers evaluated with the colorimetric method (Table 1). Tendencies for enhanced browning and changes inside parenchyma are secondary phenomena occurring often after disruption of cell structure due to mechanical damage, physiological stress or infection with pathogens [Rogozińska et al., 1986; Zgórska, 1989; Miller, 1996; Pawelzik & Delgado, 1999]. In the presented study, a significant increase was observed in darkening of mechanically-damaged tubers evaluated with the colorimetric method as affected by the application of herbicides. In turn, the visual assessment of tubers did not demonstrate any significant differences (Table 1). The effect of genetic determinants on parenchyma darkening has been described by a number of authors, including Zgórska [1989], Ceglarek et al. [1990], Dean et al. [1993], Leszczyński [2000], and Delgado et al. [2001]. Results of a study by Zgórska [1989] show that the highest susceptibility to darkening was observed for starch-rich varieties, and similar findings were reported in the presented study where var. Dorota characterized by the highest of content of starch appeared to be the most susceptible to darkening. The effect

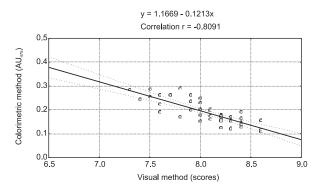


FIGURE 1. Correlation coefficient between the visual and colorimetric methods in determination of the oxidative potential of undamaged potato tubers after harvest.

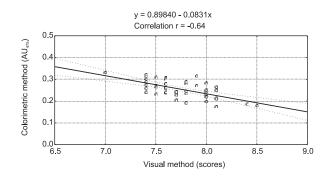


FIGURE 2. Correlation coefficient between the visual and colorimetric methods in determination of the oxidative potential of damaged potato tubers after harvest.

of starch-rich varieties susceptibility to darkening results from the fact that during that process a dark pigment is absorbed by starch which instead of being glossy becomes grey and mat [Styszko, 2002]. An increase in darkening intensity after longterm storage is a common regularity widely discussed by a number of authors [Sowa, 1987; Zgórska, 1989; Delgado et al., 2001]. It has also been confirmed in the reported experiment. A correlation calculated for the results of darkening degree obtained with the use of visual and colorimetric method was significantly positive, irrespective of experimental factors and period of assessment (Figures 1-4). The correlation coefficients calculated for both these methods after harvest reached: -0.84 for non-bruised tubers, -0.69 for bruised tubers, whereas after storage they accounted for -0.54 and -0.73, respectively. Similar results were obtained by Delgado et al. [2001] and Wichrowska [2004], although they referred to different varieties. Once obtaining a high correlation between the methods, Delgado et al. [2001] assigned a 9-point Danish scale of Henniger [1968] to a 5-point scale by Dean et al. [1993].

Owing to the fact that not the entire range of the 9-point Danish scale is usually exploited, since points $1 \rightarrow 5$ denoting black tubers are the so-called "dead scale", we suggest resigning from those points. We also postulate further modification of the Danish scale assignment to the scale by Dean *et al.* [1993], (Table 2). In addition, to facilitate interpretation of results, it seems advisable to apply a scale that would not be

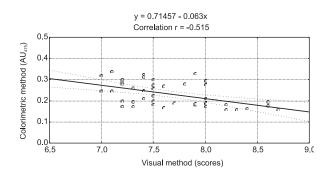


FIGURE 3. Correlation coefficient between the visual and colorimetric methods in determination of the oxidative potential of undamaged potato tubers after storage.

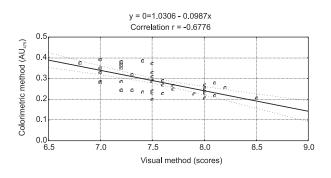


FIGURE 4. Correlation coefficient between the visual and colorimetric methods in determination of the oxidative potential of damaged potato tubers after storage.

TABLE 2. Susceptibility categories of discoloration potential determined with the homogenization and visual methods .

| | Susceptibility categories | Colorimetric method (AU ₄₇₅) | Visual method (scores) |
|---|---------------------------|--|------------------------------|
| 1 | Resistant | 0.0-0.2 | 8.1-9.0 |
| 2 | Moderately resistant | 0.21-0.4 | 7.1-8.0 |
| 3 | Moderately susceptible | 0.41-0.6 | 6.1-7.0 |
| 4 | Susceptible | 0.61-0.8 | 5.1-6.0 |
| 5 | Very susceptible | >0.8 | 1.0-5.0 |

revered, *i.e.*: 1- would indicate tubers resistant to darkening and 5 – tubers highly susceptible to darkening.

CONCLUSIONS

1. In evaluation of the degree of darkening with the two methods applied in the experiment, the instrumental one turned out to be more reliable.

2. A high correlation between the methods: visual and homogenization ones indicates that they can be used interchangeably.

3. Damage and storage of potato tubers caused an increase in the intensity of darkening of raw tubers parenchyma.

4. The applied herbicides contributed significantly to the increased darkening of parenchyma, determined with the colorimetric method. The visual methods not always confirmed such a dependency.

5. The commonly applied reversed Danish scale used in the sensory evaluation requires modification.

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OCENA STOPNIA CIEMNIENIA ENZYMATYCZNEGO BULW ZIEMNIAKÓW JADALNYCH WYWOŁANEGO WPŁYWEM HERBICYDÓW, USZKODZEŃ MECHANICZNYCH I PRZECHOWYWANIA METODĄ INSTRUMENTALNĄ ORAZ ORGANOLEPTYCZNĄ

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Cel badań stanowiło porównanie dwóch metod badawczych (wizualna i kolorymetryczna - spektrofotometryczna). Do badań wybrano bulwy trzech odmian ziemniaków (Victoria, Asterix, Dorota) uzyskane z plantacji, na której w okresie wegetacji zastosowano herbicydy, na tle stałego nawożenie N : P_2O_5 : K_2O (100 : 100 : 150 kg/ha) oraz przechowywane przez okres 6 m-cy (temp. 4-6°C, wilgotność względna powietrza 90-95%). Współczynniki korelacji dla obu metod po zbiorach wynosiły: dla bulw nie obitych (-0,84), obitych (-0,69), natomiast po przechowywaniu wartość ta wynosiła odpowiednio (-0,54) i (-0,73). Uzyskany wysoki stopień korelacji pomiędzy metodami dla oceny stopnia ciemnienia enzymatycznego wskazuje, iż metody mogą być stosowane zamiennie. Uszkadzanie mechaniczne jak i przechowywanie bulw ziemniaków przyczyniło się do wzrostu intensywności ciemnienia miąższu surowego. Natomiast stosowanie herbicydów różnicowało istotnie ciemnienie miąższu bulw surowych, tylko w przypadku zastosowania do oceny metody kolorymetrycznej.