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At harvest prediction of the susceptibility of potato varieties to blackspot after impact over long-term storage

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ABSTRACT

The paper presents a prediction method of the susceptibility blackspot damage after impact of potato varieties for three- and seven-month storage of potato tubers at a temperature of 4 °C based on the geometric parameter of the microstructure of tuber parenchyma determined at harvest. Similar sized and shaped potato tubers of twenty eight cultivars harvested in 2012 and 2013 were analysed. Analyses of the tuber parenchyma microstructure were carried out using an optical confocal microscope for ten tubers of each variety. Cylindrical samples with a thickness of 1 mm and a diameter of 10 mm were collected from the perimedullary zone. Microscopic images of the structure were analysed. The size parameter of the cell cross-section surface area was determined two days after harvest, and the susceptibility of each variety to blackspot after impact was assessed after three and seven months of storage. The mean post-harvest size of the tuber parenchyma cells and the susceptibility to blackspot during storage are related. Tubers of varieties with smaller cells of the perimedullary zone tissue exhibited greater blackspot susceptibility during storage.

1. Introduction

Mechanisation of cultivation processes during the growing period induces external stresses on agricultural commodities, which may lead to damage such as bruises, discolouration, fractures, and acceleration of deterioration processes (Peters, 1996; Malaga-Toboła, 2007; Opara and Pathare, 2014; Stropek and Gołacki, 2016). Despite the considerable research and technological effort undertaken to minimise losses in the processes of production and storage of agricultural products, the losses are still substantial and difficult to eliminate (Smittle et al., 1974; McGarry et al., 1996; Rybiński et al., 2009; Omid et al., 2010; Tomas et al., 2010; Nawrocka et al., 2012; Rusinek et al., 2012; Praeger et al., 2013; Bojanowski et al., 2013; Heltoft et al., 2016; Grudzińska and Barbaś, 2017; Gancarz et al., 2017). The most common external and internal damage is reflected in a considerable decline in the quality of raw materials and the final product, which results in great economic losses (McGarry et al., 1996; Baranowski et al., 2013). The most common internal damage is blackspot, which affects potatoes, apples, pears, apricots, cherries, peaches, onions, garlic, peppers, tomatoes, and cucumbers (Reeve, 1968; Blahovec et al., 1996; Ariana et al., 2006; Arazuri et al., 2010; Grimm et al., 2012; Stropek and Gołacki, 2016). Parenchyma darkening is a consequence of disruption of the cell structure through mechanical injury, physiological stress, or development of diseases and it is known as blackspot. It leads to melanin biosynthesis through oxidation of phenolic compounds catalysed by oxidase enzymes (Reeve, 1968; Zgórska and Frydecka-Mazurczyk, 2000; Storey, 2007; Grimm et al., 2012).

Potatoes are one of the major crops worldwide characterised by multiple applications among others in direct consumption, food industry, and for production of fodder, starch, ethanol, and boxes (Mullins et al., 2006; Andre et al., 2014; Graveland, 2014; Olsen, 2014; Devaux et al., 2014). Blackspot damage to potato tubers is intensified during storage (Blahovec, 2006; Grudzińska and Barbaś, 2017).

Many factors and parameters contributing to development of potato tuber blackspot have been identified; however, many of them are contradictory. Zgórska and Frydecka-Mazurczyk (2000) found that blackspot depends on specific characteristics of a variety. However, in a two-year study of fourteen varieties, Gancarz (2016) demonstrated that cultivars with high susceptibility to blackspot in the first year of the study were characterised by the lowest susceptibility degree in the following year. Mohsenin (1986) showed that the smaller the tuber curvature radius, the higher the susceptibility to external damage. In contrast, investigations reported by McGarry et al. (1996) as well as Baritelle and Hyde (1999) proved that larger tubers, i.e. with a greater curvature radius, were more susceptible to blackspot. Baumgartner et al. (1983) as well as Grudzińska and Barbaś (2017) found that the dry matter content was positively correlated with the development of blackspot. Starch is the major constituent of dry matter and many reports indicate that starch content influences the susceptibility to this damage (Gray and Hughes, 1978; Baritelle and Hyde, 2003). Cell walls

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are an important component of dry matter and may have an impact on the development of blackspot (rke et al., 2000, 2002;).

Although the cell size is not involved in the chemical process of blackspot development, it is regarded as one of the key factors indispensable for initiation of the process. Destruction of the cell membrane leads to release of hydrolytic enzymes, i.e. peroxidase and phenyloxidase, which are catalysts of tissue darkening (Partington et al., 1999; Johnson et al., 2003). Although these biochemical compounds exert an effect on the colour of the spot, they do not have a direct effect on the size of the spot in the tissue, and many studies have found low correlations between tissue susceptibility to blackspot and the activity of polyphenoloxidase PPO or its substrates (McGarry et al., 1996; Stevens and Davelaar, 1996; Lærke et al., 2000).

Although the problem of potato blackspot and many determinants of this type of damage have been known for several decades, postharvest determination of the susceptibility of a given variety to the damage in a given year is still impossible with the current knowledge and available technology (Baritelle and Hyde, 1999; Zgórska and Frydecka-Mazurczyk, 2000; Blahovec, 2006). Blackspot is caused by tissue damage involving destruction of the cellular structure, which determines the quality and performance traits, in particular those associated with mechanical properties (Aguilera, 2005; Devaux et al., 2008; Zdunek et al., 2008; Singh et al., 2014). As shown by studies conducted so far, the microstructure of plant tissue depends on the variety, tissue type (location in the tuber), fertilisation, environment, harvest date, and storage conditions and undergoes changes via mechanical and thermal interactions, which makes it an element that combines many others parameters affecting blackspot (Konstankiewicz et al., 2002; Sun and Li, 2003; Gancarz and Konstankiewicz, 2007a; Chassagne-Berces et al., 2009; Gancarz et al., 2014; Winisdorffer et al., 2015). The results of investigations on the impact of parenchyma cell size in the potato tuber on the blackspot damage are inconclusive, although a majority of studies confirm the influence of this parameter on disorder development (Hudson, 1975; Gancarz, 2016). Geometric characteristics of potato cells such as the size and shape are possible to determine in each phase of tuber development; they are also relatively predictable and should be used to predict tuber susceptibility to blackspot darkening, which is important from a practical point of view (Gancarz et al., 2007; Gancarz and Konstankiewicz, 2007a; Gancarz et al., 2014; Gancarz, 2016). Based on the specific quantitative correlation between the cell size and development of blackspot caused by mechanical damage, the aim of the paper was to develop a method for prediction of the susceptibility to blackspot damage of potato tissue after impact for three and seven months of storage of tubers based on the tissue cell sizes measured at harvest.

2. Materials and methods

2.1. Materials

Fourteen varieties of potatoes (*Solanum tuberosum* L.) per year: Asterix, Andromeda, Bartek, Czapla, Denar, Fresco, Irga, Irys, Pasat, Roxana, Syrena, Tajfun, Victoria, and Vineta, cultivated in 2012 and 2013, were used. It gives twenty eight cultivars in research. All varieties were obtained from Nowosiółki in Telatyn commune, Poland. The varieties had different levels of blackspot damage susceptibility, according to the information provided by the PBAI (2010) in the characteristics of the national register of the potato varieties. All varieties were cultivated on black earth. A green fertilizer (white mustard $3\,kg\,m^{-2}$) and a mineral fertilizer N: P: K in an amount 9: 8: 12 (x $10^{-3}\,kg\,m^{-2}$) were used. The tubers were hand collected in their full maturity state. For the first two weeks, the tubers were maintained at 15 °C and ca. 95% humidity. The temperature in the chamber was then gradually lowered over two weeks to reach 4 °C (95 % RH). Tubers were stored for three and seven months, and then subjected to a reconditioning treatment at two weeks, at 20 °C (95% RH) to investigate their susceptibility to blackspot. The tubers were sampled for the mechanical analyses used to determine of their susceptibility to blackspot during the selected storage periods and year of cultivar.

2.2. Methods

2.2.1. Tuber selection

The microscopic post-harvest analysis and the determination of tuber susceptibility to after-impact blackspot during the storage periods were conducted on 10 tubers of each variety with a similar size and shape. Computer image analysis was employed as described in Gancarz and Konstankiewicz (2007b). Briefly, the method consists in taking images of several externally intact tubers of each variety. Tuber size and shape was defined based on the surface and elongation of the flat tuber image. The surface area of the tubers used in the investigations ranged from 12.56 cm^2 to 28.26 cm^2 and the elongation was in the range of 0 – 0.2 (Gancarz et al., 2007; Gancarz and Konstankiewicz, 2007b). These parameters closely correspond to the 4–6 − cm tuber sizes, which are most desirable in industry.

2.2.2. Sample tissue preparation

The laboratory analyses of the microstructure were conducted two days after harvest at constant room temperature (22 °C) and at 45 – 60% relative air humidity. Ten tubers for each year of harvest and variety were used for microscopy. A cylindrical sample with 1-mm thickness and a diameter of 10 mm was taken from perimedullary zone tissue of each research tuber (Gancarz, 2016).

2.2.3. Confocal microscopy

Analyses of the microstructure of the tuber perimedullary zone were performed using an optical confocal microscope “CONFOCAL 2002” (JZD Komorno, Czech Republic), which facilitates observation of the tissue microstructure without prior preparation (Konstankiewicz et al., 2002). The samples were placed on a table moving precisely in a perpendicular plane towards the optical axis of the microscope with an accuracy of 1 μm, to facilitate taking a sequence of images, which can be combined and reveal a larger surface area of the analysed structure (Gancarz et al., 2003, 2007). A sequence of 25 microscopic images of the structure was taken over approximately sixty seconds. An additional advantage of this method is the fact that the sample does not dry up and no microstructure changes take place. The measurements were made only for the perimedullary zone of the tuber. This area was selected, as it is the largest tissue and where blackspot develops most frequently. In the analyses of the sample microstructure performed with the optical confocal microscope, only the surface oriented perpendicular to the stolon-apex axis was observed. This direction of the observations was selected, since previous investigations conducted by Gancarz et al. (2014) did not show anisotropy of the size and shape of the tuber perimedullary zone relative to the sampling direction, i.e. perpendicular or parallel to the stolon-apex axis.

2.2.4. Microscopy image analysis

Quantitative analysis of cell structure parameters for the microscopic images was performed in accordance with a previously developed methodology using the Aphelion program, ADCIS, France (Gancarz et al., 2007). The semi-automatic analysis of the images obtained with the optical confocal microscope was carried out, because a fully automatic analysis overestimates the cell size parameter and simultaneously underestimates the number of cells obtained in the analyses (Gancarz et al., 2003, 2007). Parameters and their distributions were obtained for the flat cross-sections of the cell structure. The distributions associated with the cell size, i.e. the surface area of the cell flat cross-section A_0 was determined. From all varieties analysis involved seven thousand confocal microscopy images, which provided parameters of the cell flat cross-section and their distributions for approx. 250,000 cells. Size parameters were determined for each slice for