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PARAMETERS USED FOR THE EVALUATION OF POTATO (*SOLANUM TUBEROSUM* L.) NITROGEN USE EFFICIENCY: A REVIEW

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Abstract
Potato is one of the important crops worldwide, and cultivation requires a lot of resources and nitrogen (N) to ensure yield. This kind of growing technology can cause environmental pollution. It is necessary to optimize the management and use of potatoes with high nitrogen use efficiency (NUE) varieties as an alternative to the application of large amounts of fertilizers to improve the environmental impact of potato production without affecting yield and quality. The aim of this review is to find various morphological and physiological characteristics of the potato crop that can be used in potato breeding for NUE evaluation and to create new varieties with high NUE. Looking for traits such as yield, protein and starch content, each variety has its limits that can be affected by climatic conditions. Area Under Canopy Cover Progress Curve (AUCCPC) is potentially good for detecting NUE in field conditions. Nitrogen-efficient genotypes tend to have early canopy development but a low amount of N in tubers. Due to different factors that can interact with genotypes under field conditions, the experiments in controlled conditions as *in vitro* system can be used for the investigation of genotypes in a short period. Under *in vitro* conditions, root development can be observed very well, which can usually be impossible in soil experiments.
Key words: morphological parameters, *in vitro*, yield, protein, starch.

Introduction

Potato (*Solanum tuberosum* L.) is known to be the third most important crop in the world by production and the fifth by consumption (Haverkort & Struik, 2015; Devaux, Kromann, & Ortiz, 2014). According to Van Dingenen *et al*. (2019), potato tubers are a great source of starch, antioxidants, vitamins, essential amino acids, and important minerals. Historically, potato production in Latvia has been one of the most important branches of agriculture. Since 2000, the potato growing areas decreased by 33 thousand hectares and potato production has taken a downward turn by 370 thousand tonnes. To ensure potato cultivation at the same level as now, extending the potato growing area or increasing the yield is necessary (Ministry of Agriculture, 2021). The potato requires abundant nitrogen (N) (Haverkort, 2018). Most of the N in the soil is partly available for plants (Zebarth & Rossen 2007). Large N fertilization input, combined with the shallow root system of potatoes (Iwama, 2008), can result in nitrate leaching and subsequent contamination of groundwater (Frink, Waggoner, & Ausubel, 1999; Milburn *et al*., 1990; Sharifi & Zebarth, 2006), and atmosphere through gaseous emissions (Hirel *et al*., 2007). European legislation (Nitrate Directive (91/767/EEC) and Water Framework Directive (2000/60/EC)) has established limits on the N input in crop production in Europe (Vos, 2009). Still, directives have struggled with the low support of farmers as well as various conflicts between environmental and income support objectives (Hasler *et al*., 2022). New potato varieties with improved nitrogen use efficiency (NUE) will reduce the environmental impact of potato production without affecting yield and quality (Vos, 2009), will be acceptable for environmentally friendly farming systems and be beneficial to improve nutrient management. The aim of this review is to find various morphological and physiological traits of the potato crop that can be used in potato breeding for NUE evaluation in order to create new varieties with improved NUE.

Materials and Methods

The semi-systematic monographic method was used to analyse and summarise the main information on potato NUE and traits that could possibly be implemented as an alternative for NUE evaluation. NUE is considered as potatoes' ability to use uptake N in the development of canopy cover, protein and starch content, and yield.

Results and Discussion

Many crops, including potato, have high genetic variation, making breeding of NUE practical (Lammerts van Bueren 2010; Lammerts van Bueren *et al*., 2014). Unfortunately, when it comes to potatoes breeding strategies towards NUE, there is lack of information (Lammerts van Buren & Struik, 2017). Various quality traits such as tuber starch content, tuber protein content, tuber yield, and tuber size distribution are affected by the available N amount (Zebarth *et al*., 2004). Weather conditions need to be considered as well (Vos, 2009). Cohan et al. (2018) explained how the variation of results can be determined by Nitrogen × Environmental interaction.

Nitrogen Use Efficiency

Potato varieties with improved NUE will perform well under high and low N availability (Ospina *et al*., 2014). Various definitions of NUE have been described, but it may differ in various crops (Fageria

& Baligar, 2005). Moll, Kamprath, & Jackson (1982) defined NUE as the yield per unit of available N in the soil, including the residuals N present in the soil and the fertilizer. Mostly, potato NUE is explained as the production of dry matter per unit of N in soil (Getahun *et al*., 2020). In Meise et al. (2019) study, potato NUE has been calculated as fresh tuber yield produced per unit of N in the soil. There have been cases where potato NUE is defined as tuber dry matter (kg) per total N available in the soil (kg), also known as agronomic NUE (Tiemens-Hulscher, Lammerts van Bueren, & Struik, 2014). Hawkesford & Griffith's (2019) study established NUE based on cereal crop yield per unit of total available nitrogen in the soil. The value obtained is equivalent to potato dry matter yield per available N in the soil like mentioned in Tiemens-Hulscher, Lammerts van Bueren, & Struik (2014). It is suggested that NUE differs and depends on the potato genotype and maturity type (Milroy, Wang, & Sandras, 2019). According to Hirel et al. (2007), NUE can be divided into nitrogen uptake efficiency (NUpE) and nitrogen utilization efficiency (NUtE). NUpE describes the plant's ability to capture N from the soil and can be defined as the N content in the plant per available N (Meise *et al*., 2019; Shum & Jansen, 2014). The total N uptake reflects in the biomass (Ierna & Mauromicale, 2019) and canopy development (Tiemens-Hulscher, Lammerts van Bueren, & Struik, 2014). The NUpE can differ between years, N fertilizer amount and variety. NUtE is plants' ability to produce yield or quality parameters and can be defined as dry matter yield per available N, resulting not only in tuber yield but also in starch and protein content (Ospina *et al*., 2014; Tiwari *et al*., 2020). In potatoes with good NUE, NUtE will increase as crop available N decreases (Bohman, Rosen, & Mulla, 2021), because an increasing amount of N has a negative effect on NUE and NUtE (Mirosavljević *et al*., 2019). This is the reason why NUE is better detected in cases with a smaller amount of N in the growing media (Hawkesford & Griffiths, 2019).

Canopy Development

The potato crop is very sensitive to the amount of N fertilizer. Depending on a higher N availability, plants tend to have longer life cycles and even have larger leaves (Haris, 1992). Oliveira (2000) mentioned that more N can increase the number of photosynthetically active leaves as well as induce leaf appearance and branching upper part of the plant. On the contrary, Biemond & Vos (1992) described that leaf appearance on a branch is not affected by available N. There are several empirical models with a focus on potato canopy development (MacKerron & Waister, 1985). Canopy cover can be used in the vegetation season for the evaluation of NUE (Gastal *et al*., 2015; Tiemens-Hulscher, Lammerts van Bueren, & Struik, 2014). The Area Under Canopy Cover Progress Curve (AUCCPC) model developed by Khan (2012) uses three basic equations to describe canopy expansion, maintenance, and senescence. This AUCCPC model needs Canopy ground cover (%) (Ospina *et al*., 2014) and temperature, calculated in thermal time (Khan, 2012). According to Yin et al. (2003), instead of thermal time, beta thermal time can be used. The descriptive AUCCPC can be used to determine the types of potato maturity (Khan, van Eck, & Struik, 2013). Potato crops can adapt leaf development to limit N, while still maintaining productivity per leaf area by changing individual leaf size and branching (Vos, 2009). AUCCPC model can be used to describe the interaction between genotype and available N in soil and strongly correlate with NUE (Tiemens-Hulscher, Lammerts van Bueren, & Struik, 2014). The accumulation and production of dry matter in the potato crop are related to canopy development and leaf area index (Haverkort *et al*., 1991). The leaf area index was correlated with yield and yield parameters by increasing the proportion of large tubers (Van Oijen, 1991). These parameters can be increased by the amount of N available (Kleinkopf, Westermann, & Dvelle, 1981). Ospina et al. (2014) reported that nitrogen significantly affected the thermal time needed to reach maximum soil cover, which was higher in lover available N compared to high available nitrogen. Ospina (2014) also mentioned how NUE for genotypes that performed well under a rich N showed the same tendency at low N. AUCCPC is highly correlated with yield (Ospina *et al*., 2014). Tiemens-Hulscher, Lammerts van Bueren, & Struik (2014) under organic conditions observed that cultivars that rapidly established a high maximum soil cover can maintain this maximum for long and slowly leaf senescence potentially sustaining high yields. The study explained that nitrogen-efficient genotypes suitable for organic farming must have early canopy development with high agronomic NUE and NUtE, but a low amount of N in tubers.

Yield

The increase of N availability in the soil will increase potato yield. However, each variety has its limits (Fontes *et al*., 2010). Yield is the result of genotype, environment and genotype × environment interaction (Steyn *et al*., 2016). Higher N availability will induce leaf development, consequently inducing tuber bulking (Goffart, Olivier, & Frankinet, 2008). The results presented by Kasal et al. (2011) showed a significant relationship between NUE and the amount of rainfall and temperature levels in the vegetation season towards yield building. Crop's ability to uptake N and show its NUE can be affected by soil composition and microbial processes in the soil (Burger & Jackson, 2004; Walley *et al*., 2002). According to Milroy, Wang,

& Sadras (2019), yields increased with an increase in N availability during vegetation, especially when the genotype has a later maturity type. Still, the effects of nitrogen and maturity type depend on meteorological conditions in each location. In the case of the potato crop, Skrabule, Vaivode, & Ruža (2012) concluded that increasing the amount of N above 120 kg ha^{-1} will not result in a significant increase in yield, which can coincide with the work by Mőller et al. (2007), who indicates that 48% of the variation in yield can be determined by differences in nitrogen availability. Using a smaller amount of N, the potato will use more efficiently N compounds that are available in the soil (Skrabule, Vaivode, & Ruža, 2012). Plant density is also an important factor for establishing uniformity and ensuring high yields (Masarirambi *et al*., 2012). Nitrogen availability affects numerous traits and yield components, such as the final number of the tubers and harvest index (Biemond & Vos, 1992). Nitrogen also affects different quality aspects, such as tuber size distribution (Zebarth *et al*., 2004). Hawkesford (2014) conclude with Zebarth et al. (2004), that increasing the yield without additional N will result in better NUE and an effective limitation of N in an environment without the expense of quality.

Protein

According to Haile, Nigussie & Ayana (2012), protein content in wheat (*Triticum aestivum*) grains can be strongly influenced by the amount of available N, time of application, and genotype. The results between yield and protein, indicate an inverse yield-protein relationship. The amino acids originating from nitrogen uptake are also used for the synthesis of enzymes and proteins that build the architecture of plants. Protein content in plants rises after the flowering stage when a large amount of uptaken N is used for protein synthesis by realising free amino acids from protein hydrolysis that are exported to reproductive storage organs (Masclaux *et al*., 2001; Masclaux-Daubresse *et al*., 2010). The main potato protein patatin makes about 40% of the potato-soluble protein (Camire, Kubow, & Donnelly, 2009). This kind of protein can be found in tubers and stolons, and its quality aspects have better properties than soy (*Glycine max*) proteins (Waglay & Karboune, 2016). Bartá & Bártová (2008) and Bártová et al. (2013) have characterized patatin as a storage protein. The best connection between NUE and patatin is described in Lehesranta et al. (2007), where authors explain, that patatin and other storage proteins are negatively affected by lower available N in the soil.

Starch

The starch content and starch yield are significant parameters for potato starch processing. Varieties with higher starch content can help in cases when environmental conditions weren't so favourable for yield (Ruža, Skrabule, & Vaivode, 2013). The available N influences the starch content and starch yield by positively affecting canopy development (Koch *et al*., 2020). In most cases, increasing available N has a negative effect on starch content in tubers (Bachmann-Pfabe & Dehmer, 2020; Öztürk *et al*., 2010). Kumar et al. (2007) indicate that tuber quality is determined by genotype. However, the results of Ruža, Skrabule, & Vaivode (2013) showed that the starch content tends to decrease with a higher fertilization level. N deficiency results in the accumulation of carbohydrates such as sugars and starch (Remans *et al*., 2006; Scheible *et al*., 2004). Tuber yield negatively correlated with tuber starch content (Schönhals, 2014). However, Bombik, Rymuza, & Olszewski (2019) found no significant correlation between tuber yield and starch content. Any physiological and morphological changes are not only controlled by the environment but also genetically, to develop varieties with good NUE, knowledge about the genetics of the NUE and NUE-related traits, such as starch, is vital (Getahun, 2017). According to Zhang et al. (2020) experiment, N metabolism showed a close correlation with NUtE, which indicates different mechanisms of how potatoes respond to N deficiency.

NUE evaluation under controlled conditions

It may not be possible to fully assess the entire genotypes for NUE under field conditions, but it's possible to evaluate most of them by improving existing and evolving new phenotyping methods that are based on different growing conditions (Cohan *et al*., 2018). Field testing of potatoes allows only one or two trials per year, depending on the climate zone, while the use of *in vitro* culture systems allows investigating many plants under highly controlled conditions, excluding different pathogen effects in a short period (Schum & Jansen, 2014; Schum *et al*., 2017). *In vitro* culture can be a great opportunity to monitor root growth in different N supplies (Shum *et al*., 2017). Increased root growth under environmental stress conditions can be an advantage because it allows improved nutrient and water uptake by using resources from the remote soil zone (Ghanem *et al*., 2011). Such morphological characteristics as the number and length of roots depend on plant species, genotype and water availability (Malamy, 2005; Christensen *et al*., 2017). Changes in the soil can induce the adaptive response to development (Ghanem *et al*., 2011). N deficiency induces an increased outgrowth of lateral roots (Hawkesford, 2014). During the vegetative phase, the roots of plants behave as sink organs for the assimilation of inorganic N and the synthesis of amino acids originating from uptake and reduction of the nitrate assimilatory pathway (Hirel & Lea, 2001). Unfortunately, potatoes are known to have a shallow root system with genetically determined abundant root development, but roots for genotypes with higher NUE (Sharifi, Zebarth, & Coleman, 2007) can develop better. According to Hajari, Snyman, & Watt (2014; 2015), experiments on NUE related parameters *in vitro* are rare. While Meise et al. (2018, 2019) mentioned pot trials as a great alternative to field trials to assess NUE and assess N deficiency. The same approach was used by Pourazari, Anderson, & Weih (2018) conducting experiments in the greenhouse to evaluate tuber yield associated with plants available N, as well as Kollaricsné Horváth et al. (2019), explored the genetic expression of NUE genes potato genotypes using pot trial. Xie et al. (2018) used a hydroponic approach to evaluate the shortage and availability of N in potatoes. According to Shum et al. (2017), the stability of plant performance under different amounts of N in the *in vitro* system can correlate to the yield stability of cultivars in pot trials and field trials not only for prescreening of germplasm and identification of different genotypes but also for the investigation of different NUE parameters.

Conclusions

It is necessary to optimize the management and use of high-NUE potato varieties as an alternative to the existing large amounts of fertilizer application existing to improve the environmental impact of potato production without affecting yield and quality standards. Varieties with high NUE will help increase potato yield and reduce the cost of production and the environmental impact. NUE can be calculated with different methods or detected by various traits of the potato crop. A very good predictor of NUE in field trials can be AUCCPC and yield. AUCCPC and yield are highly correlated between themselves and depend on the available N from the soil. Protein and starch content are also good traits that show how well the potato genotype interacts with the amount of N. Still, field trials are exposed to different factors and can take a lot of years and resources to conduct results, and NUE assessment in controlled conditions in greenhouses and laboratories are needed. The *in vitro* system allows the investigation of many plants under controlled conditions in a short period. Under *in vitro* conditions, root development can be observed very well, which can usually be impossible in soil experiments.

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