



The absorption of iodine from 5-iodosalicylic acid by hydroponically grown lettuce



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ABSTRACT

The aim of the study was to determine the effect of various doses of 5-iodosalicylic acid (5I-SA; organoiodine compound) on the growth, chemical composition, and efficiency of iodine biofortification of lettuce cultivated in a nutrient film technique hydroponic system. The additional purpose was to compare the impact of 5I-SA with the effect of a selected KIO₃ dose applied separately or together with salicylic acid (SA). The following combinations were tested with the introduction of 5I-SA and KIO₃ (together with SA) into the nutrient solution: 1./Control, 2./KIO₃ (40.0 μM I), 3./KIO₃ + SA (40.0 μM I and 40.0 μM SA), 4./5I-SA (1.6 μM I), 5./5I-SA (8.0 μM I), and 6./5I-SA (40.0 μM I). A strong toxic effect on plants was revealed only in the case of the highest iodine dose applied as 5I-SA (40.0 μM I). In the remaining combinations, lettuce head weight and its phenotype did not differ from the control. However, an unfavourable effect of 40.0 μM I applied as KIO₃ on the chemical composition of plants (the content of sugars, phenolic compounds, and mineral nutrients) was not noted for 5I-SA applied in 1.6 and 8.0 μM I doses. The values of iodine transfer factor for the latter two combinations were higher than after the application of KIO₃ or KIO₃ + SA. Application of KIO₃ + SA increased the uptake and accumulation of iodine in lettuce leaves as compared to the treatment with KIO₃ alone. A higher content of 5I-SA was noted in roots than in leaves. The obtained share of 5I-SA in total iodine content indicates that most of this compound is in plants converted into other speciation forms.

1. Introduction

Iodine is a mineral nutrient necessary for the functioning of human and animal organisms. The widespread problem of the hidden hunger and endemic deficiency of this element results from its low content in some soils. It is also related to the low mobility of iodine within the soil–plant system (White and Broadley, 2009), which is characterised by low values of transfer factor (TF) coefficient, which characterises iodine transport from the nutrient solution to roots and leaves and is expressed as a ratio of I content in plants to I content in soils. For plants cultivated in natural soils, the values of TF do not exceed 1 (Ban-Nai and Muramatsu, 2003; Sheppard and Motycka, 1997). Dai et al. (2004) showed that iodine TF calculated for various vegetable species grown in a pot experiment in a greenhouse ranged from 0.1 to 10 according to the following order: spinach leaves > pakchoi leaves > water

spinach leaves > celery shoots > onion shoots = carrot roots. The lowest TF values (from 0.0005 to 0.02) are noted for cereal grains (Shinonaga et al., 2001). In general, the values of this coefficient are higher when plants are cultivated in hydroponic systems, as no iodine sorption, characteristic for iodine behaviour in soils, occurs (Blasco et al., 2008; Smoleń et al., 2016a).

The development of agrotechnical rules of iodine biofortification (enrichment) of plants is limited because this element is not a plant mineral nutrient. Recent reports indicate that, depending on its dose and chemical form, iodine can be included in the group of ‘beneficial elements’ for selected crop plants (Medrano-Macias et al., 2016). A positive effect of iodine on plants is not demonstrated by a direct increase of yield; however, it affects physiological and biochemical processes in plants. It has been suggested that exogenous application of this element may improve nitrogen use efficiency (Blasco et al., 2012) or

Abbreviations: SA, salicylic acid; 5I-SA, 5-iodosalicylic acid; NFT, nutrient film technique; TF, transfer factor

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selected parameters of the quality of tomato fruits (Kiferle et al., 2013; Smoleń et al., 2015).

It seems necessary to conduct studies focused on determining the preferential form of iodine for plant uptake and accumulation. Another aspect that is just as important is that the process of iodine biofortification of plants should be conducted to improve and in no case lower the nutritional and health-promoting quality of yield. Particular attention should be paid to the vitamin, sugar, phenolic, and mineral contents in crops.

In soil under aerobic conditions, humic acids are involved in the reduction of iodate (IO_3^-) to molecular iodine. The latter can either (I_2) volatilise or interact with soil organic matter (SOM) to form organoiodine compounds (Yamaguchi et al., 2005, 2006). It is worth mentioning that the SOM contains various organic acids, including salicylic acid (SA) and its derivatives (Hue et al., 1986).

Iodate ions (IO_3^-), when applied to the soil in low doses through fertigation, can undergo chemical reduction to I_2 (or I^-) by soil microorganisms and SOM (Yamaguchi et al., 2005) or on the root surface (Kato et al., 2013). As molecular iodine (I_2) is more reactive, it probably binds to the aromatic rings of SOM compounds. It has been suggested that formed organoiodine compounds can be absorbed by plant roots and distributed in plants cultivated in soil and hydroponic more easily than I^- or IO_3^- (Smoleń et al., 2016a).

Yamada et al. (1996) revealed that after being introduced into the soil, mineral iodine undergoes partial conversion to organoiodine compounds. However, their structure and nomenclature were not presented by the authors. Organically bound iodine ranged from 5.6% to 20.3% of water soluble iodine in soil extracts; the remaining forms were I^- and IO_3^- . Some reports indicate that the level of plant uptake of iodoacetic anion is higher or similar to that of I^- and IO_3^- (Umaly and Poel, 1971; Weng et al., 2008). Plant preference towards the uptake of mineral and iodoacetic anions can be ranged as follows: $\text{I}^- > \text{CH}_2\text{ICOOH}^- > \text{IO}_3^- > \text{IO}_4^-$ for barley and pea (Umaly and Poel, 1971) and $\text{CH}_2\text{ICOOH}^- > \text{I}^- > \text{IO}_3^-$ for water spinach (Weng et al., 2008).

No clear information has been found on the possibility of root uptake of organoiodine compounds with iodine bound to the phenolic group or its subsequent transport into the upper parts of the plant. What is known is that in soil and soil solution various reactions of mineral iodine take place that lead to the formation of organic forms of this element (Medrano-Macías et al., 2016). In soilless (hydroponics) systems, these processes occur much slower due to the lower bioactivity of the environment.

The physiological role of SA in plants is variously described in the literature. Some reports indicate that SA functions as a plant phytohormone (Gust and Nürnberger, 2012). According to others (Fariduddin et al., 2003; Hayat et al., 2010), SA can be defined as a phytohormone-like substance. It results from the fact that this compound takes part in the regulation of growth, development, and other physiological and biochemical processes (Fariduddin et al., 2003; Hayat et al., 2010).

SA is involved in, for example, plant protection/adaptation to biotic and abiotic stress factors (Hayat et al., 2010). Such processes in plants may be regulated by the conversion and volatilisation of SA to a respective ester, methyl salicylate (MeSA). The latter compound is a signalling molecule that triggers stress responses in other parts of the same or nearby plants. SA degradation in plants takes place through its volatilisation and formation of sugar conjugates with SA (Zhang et al., 2013).

The efficiency of plant adaptation to stress conditions in the presence of exogenous SA depends on its concentration and the duration of plant treatment with this compound. In hydroponic systems, introduction of SA into the nutrient solution may limit pathogen development as plants develop higher resistance (Mandal et al., 2009; Spletzer and Enyedi, 1999). A positive influence of SA was also revealed on the increase of plant resistance/tolerance to salinity (Tari et al., 2002), *Fusarium oxysporum* f. sp. *lycopersici* (Mandal et al., 2009), or *Alternaria*

solani (Spletzer and Enyedi, 1999). Excessive doses of SA ($> 50 \mu\text{M}$ SA) applied for too long, however, may be harmful to plants, as was described for tomato (Jung et al., 2004). Introduction of SA into the nutrient solution (in a dose of 1 mg SA dm^{-3} ; i.e. $7.24 \mu\text{M}$ SA) improved the efficiency of iodine biofortification of tomato, particularly when applied together with KIO_3 rather than potassium iodide (KI) (Smoleń et al., 2015).

The research hypothesis of the current study stated that low-molecule aromatic organoiodine compounds (such as 5-iodosalicylic acid; 5I-SA) in which iodine is bound to a phenolic ring can be absorbed from the nutrient solution by plants grown in a hydroponic system. It was also assumed that the influence of this organic form of iodine on plants varies from the effect exerted by its mineral form (IO_3^-). The impact of organoiodine compounds containing an aromatic ring on plants is yet to be revealed. Currently, no information has been presented on the processes of their conversion in plants, including the possible secondary iodine metabolites to be formed in plants after the application of low-molecule aromatic organoiodine compounds. Therefore, it cannot be recommended to use such compounds for biofortification purposes. Based on the current state of knowledge, however, it can be advised to conduct iodine enrichment of crop plants with the use of mineral forms of iodine, such as KI or KIO_3 (Blasco et al., 2008, 2012). Plant biofortification with iodine should be conducted to a level safe for consumers — people and farm animals. This results from the need to properly balance the share of iodine from biofortified vegetables in the daily intake. Studies on the application of KI or KIO_3 so far allow the determination of the optimal dose of I for lettuce cultivation so that the consumption of enriched crop is safe for consumers (Lawson et al., 2015, 2016).

Tonacchera et al. (2013) have demonstrated an increased level of urinary iodine excretion (UIE) measured in healthy volunteers after the consumption of vegetables biofortified with the mineral form of I (potatoes, cherry tomatoes, carrots, and green salad). UIE is one of the major indicators of I levels in the human population. Kopeć et al. (2015) recorded higher iodine assimilability and improved I metabolism in rats fed lettuce biofortified with iodine (KI) in relation to control lettuce using a diet including synthetic KI. In the research conducted by Koronowicz et al. (2016), inhibition of Caco-2 (cancer human) cell proliferation as well as mediated induction of mitochondrial apoptosis and/or cell differentiation were demonstrated to occur after treatment with KI-biofortified lettuce extract, but not after treatment with KI. This study showed 1326 differently expressed Caco-2 transcripts after treatment with iodine-biofortified and non-fortified lettuce extract; these transcripts were related to gonadotropin receptors, apoptosis signal, as well as Huntington's, Alzheimer's, and Parkinson's diseases.

The aim of this study was to determine the effect of various doses of 5I-SA on the growth, development, mineral composition, and efficiency of iodine enrichment of lettuce grown in a hydroponic nutrient film technique (NFT) system. The study also focused on comparing the application of a selected dose of 5I-SA with the introduction of equimolar amounts of KIO_3 and SA into the nutrient solution.

2. Materials and methods

2.1. Plant material and treatments

A two-year study (2014–2015) was conducted with lettuce (*Lactuca sativa* L. var. *capitata* 'Melodion' c.v.) cultivation in an NFT system without medium disinfection in the high experimental plastic tunnel of the Faculty of Horticulture of the University of Agriculture in Kraków. The tunnel was equipped with six individual NFT sets with 200 dm^3 medium containers, allowing to test six various nutrient solutions. Each NFT set contained beds for plant cultivation in four repetitions.

Each year seeds were sown into rockwool plugs in the first half of March (11.03.14 and 11.03.15). On 10.04.14 and 15.04.15, two-leaf seedlings were placed in the holes (spaced 25 cm apart) of styrofoam

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