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UNIVERSIDADE ESTADUAL PAULISTA (UNESP)
FACULDADE DE CIÊNCIAS AGRÁRIAS E VETERINÁRIAS
CÂMPUS DE JABOTICABAL

PHYSIOLOGICAL AND BIOCHEMICAL ROLE OF SELENIUM IN FLAVONOID SYNTHESIS AND BIOLOGICAL NITROGEN FIXATION IN LEGUMINOUS PLANTS

Matheus Luís Oliveira Cunha
Agronomist engineer

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Matheus Luís Oliveira Cunha

Supervisor: Prof. Dr. André Rodrigues dos Reis

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TÍTULO DA DISSERTAÇÃO: PHYSIOLOGICAL AND BIOCHEMICAL ROLE OF SELENIUM IN FLAVONOID SYNTHESIS AND BIOLOGICAL NITROGEN FIXATION IN LEGUMINOUS PLANTS

AUTOR: MATHEUS LUÍS OLIVEIRA CUNHA
ORIENTADOR: ANDRÉ RODRIGUES DOS REIS

Aprovado como parte das exigências para obtenção do Título de Mestre em Agronomia (Produção Vegetal), pela Comissão Examinadora:

André Rodrigues dos R
Prof. Dr. ANDRÉ RODRIGUES DOS REIS (Participaçao Virtual)
Departamento de Engenharia de Biossistemas / FEI UNESP Tupa

Felipe K.
Prof. Dr. FELIPE KLEIN RICACHENEVSKY (Participaçao Virtual)
Universidade Federal do Rio Grande do Sul / UFRGS Porto Alegre/RS

Prof. Dr. ALCINDO APARECIDO DOS SANTOS (Participaçao Virtual)
Departamento de Química Fundamental / Universidade de São Paulo - USP - São Paulo

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DADOS CURRICULARES DO AUTOR

MATHEUS LUÍS OLIVEIRA CUNHA - Nascido na cidade de Franca no dia 15 de abril de 1997. Em 2020, obteve o título de engenheiro agrônomo pela Faculdade de Ciências Agrárias e Tecnológicas – UNESP Campus de Dracena. Durante a graduação foi bolsista de iniciação cientifica pela FEPAF e FAPESP (Processo n° 2018/19667-9. Como resultado de seu trabalho, teve a oportunidade de apresentar trabalhos científicos em congressos no exterior, receber menção honrosa e publicar diversos artigos em revistas cientificas internacionais e de alto impacto. Em janeiro de 2021 ingressou no Mestrado do Programa de Pós-Graduação em Agronomia (Produção Vegetal), da Universidade Estadual Paulista, Câmpus Jaboticabal, sob a orientação do Prof. Dr. André Rodrigues dos Reis desenvolvendo o trabalho "Papel fisiológico e bioquímico do selênio na síntese de flavonoides e fixação biológica do nitrogênio em plantas leguminosas" sendo bolsista FAPESP (Processo n° 2020/10053-8). Ainda no mestrado, foi aprovado no processo seletivo do doutorado no CENA/USP com bolsa de doutorado também aprovada pela FAPESP.

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A todos os pesquisadores que trabalham arduamente para melhorar a vida das pesquisas e que em diversas vezes não são valorizados e são reconhecidos por “Ah, então você não trabalha, só estuda”.

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PAPEL FISIOLÓGICO E BIOQUÍMICO DO SELÊNIO NA SÍNTESE DE FLAVONÓIDES E FIXAÇÃO BIOLÓGICA DO NITROGÊNIO EM PLANTAS LEGUMINOSAS

RESUMO - O selênio (Se) é essencial para humanos e animais e considerado benéfico para plantas em baixas concentrações. Pouco se sabe sobre o papel do Se na nodulação de plantas leguminosas. O objetivo desse trabalho foi avaliar o efeito do Se (i) sobre o perfil de flavonoides nas raízes; (ii) sobre o metabolismo do nitrogênio e do sistema antioxidante e (iii) no perfil de nutrientes, carboidratos, pigmentos fotossintéticos e produtividade de grãos das plantas de soja, amendoim e feijão-caupi. Para isso, foram conduzidos 2 experimentos para cada cultura, sendo um conduzido em solo e o outro em hidroponia. Os tratamentos dos experimentos realizados em solo foram compostos por 5 doses de Se (0, 7,5, 15, 30 e 45 µg kg^{-1}) enquanto que nos experimentos realizados em hidroponia, os tratamentos foram compostos por 4 concentrações de Se (0, 5, 10 e 15 µmol L^{-1}) aplicados via solo e solução nutritiva respectivamente, no estágio fenológico V_2 das plantas, na forma de selenato de sódio. Nossos resultados mostraram que a suplementação com Se aumenta a síntese de flavonóides (daidzeína, genisteína, kaempferol, quercetina, rutina e naringerina) nas raízes, as quais podem ter papel na sinalização química para aumentar a infestação das raízes por rizóbio, consequentemente aumentando o fluxo de ureídeos e metabolismo antioxidativo nas plantas de soja, feijão-caupi e amendoim, tornando essas plantas mais tolerantes a estresses abióticos e gerando aumento de produtividade. Além disso, o Se pode reduzir o estresse oxidativo e aumentar a atividade das enzimas antioxidantes, eficiência do metabolismo do nitrogênio e a concentração de pigmentos fotossintéticos nas folhas.

Palavras-chave: biofortificação agronômica, compostos nitrogenados, estresse oxidativo, ureídeos, fixação biológica de nitrogênio.

PHYSIOLOGICAL AND BIOCHEMICAL ROLE OF SELENIUM IN FLAVONOIDS SYNTHESIS AND BIOLOGICAL NITROGEN FIXATION IN LEGUMINOUS PLANTS

ABSTRACT - Selenium (Se) is essential for humans and animals and considered beneficial to plants in low concentrations. Little is known about the role of Se in the nodulation of leguminous plants. This study aimed to: (i) to evaluate the effect of Se on the flavonoid profile in the roots; (ii) nitrogen metabolism and the antioxidant system and (iii) evaluate the effect of Se nutrient profile, carbohydrates, photosynthetic pigments and grain yield of soybean, peanut and cowpea plants. For this, 2 experiments were conducted for each crop, one conducted in soil and another in hydroponics. The treatments carried out in soil were composed of 5 Se doses (0, 7.5, 15, 30 and 45 µg kg-^{1}) while in hydroponics the treatments were composed of 4 Se concentrations (0, 5, 10 and 15 µmol L^{-1}) applied via soil and nutrient solution, respectively at the V_2 phenological stage of plants in the form of sodium selenate. Our results showed that Se supply increases the synthesis of flavonoids (daidzein, genistein, kaempferol, quercetin, rutin and naringerin) in roots which can have a role as chemical signals to increase root infestation by rhizobia, consequently increasing the flow of ureides and antioxidant metabolism in soybean, cowpea and peanut plants, making these plants more tolerant to abiotic stresses and increasing yield. In addition, Se can reduce oxidative stress and increase the activity of antioxidant enzymes, efficiency of nitrogen metabolism and concentration of photosynthetic pigments in leaves.

Keywords: agronomic biofortification, nitrogen compounds, oxidative stress, ureides, biological nitrogen fixation.

CHAPTER 1 - General considerations

Matheus Luís Oliveira Cunha[a], Maria Gabriela Dantas Bereta Lanza[a], André Rodrigues dos Reis[b,*]

[a] Universidade Estadual Paulista “Júlio de Mesquita Filho” (UNESP), Via de Acesso Prof. Paulo Donato Castellane s/n, Postal Code 14884-900, Jaboticabal, SP, Brazil.

[b] Universidade Estadual Paulista “Júlio de Mesquita Filho” (UNESP), Rua Domingos da Costa Lopes 780, Postal Code 17602-496 Tupã, SP, Brazil.

* Corresponding author at: andre.reis@unesp.br (A.R.R)

ABSTRACT

Selenium (Se) is an essential element for humans and animals and considered beneficial element for plants. In humans, Se deficiency is related to heart disease, and pregnancy complications. Agronomic biofortification of crops is a safe and efficient strategy to combat Se deficiency in humans. Se can benefit the growth and survival of plants under abiotic stresses by mitigation of oxidative stress and increasing the concentration of proteins, sugars, and amino acids in plant cells. Several studies have shown that Se application at low concentrations improved the defense mechanism of plants by increasing the antioxidant enzymatic activity to eliminate ROS and protect against cellular oxidation. Selenium can boost nitrogen (N) metabolism by activating N assimilation enzymes such as nitrate reductase, nitrite reductase, glutamine synthetase and glutamate synthase, which results in greater production of amino acids, proteins and availability of organic N and consequently greater crop yield. Se application at low concentration mimics sulfur deficiency in plants increasing the amino acids cysteine, methionine, selenocysteine and selenomethionine in plants. This literature review aimed to present a compilation of main findings on how Se can be the secret weapon in agriculture to increase the tolerance of plants to abiotic stresses by increasing the antioxidant metabolism, photosynthetic and nutritional status of plants, increasing yield as well as the quality of agricultural products for mitigation of hidden hunger.

Keywords: Abiotic stress, Antioxidant metabolism, Biofortification, Nitrogen metabolism, Sulfur.

1. INTRODUCTION

Selenium (Se) concentration in soils is generally very low, with an average of 0.4 mg kg-1 worldwide (Fordyce, 2013; Reis et al., 2017). There is great variation in Se concentration between soils around the world. In China (Enshi area) the Se concentration in the soil range from 3.76 to 79.08 mg kg^{-1} (Yuan et al., 2012), 8.2 mg kg^{-1} in the United States of America (Pine Ridge Fort Collins, CO) (Yasin et al., 2015) and 0.024-3.06 mg kg^{-1} in India (Punjab area) (Dhillon and Dhillon, 2016).

Se is an essential trace element for animals, including humans, and although not essential, it is considered a beneficial element for plants. In humans, Se deficiency is related to heart disease (Oropeza-Moe et al., 2015), and pregnancy complications (Hofstee et al., 2020). Recent studies have shown that Se deficiency in humans was associated with mortality risk from COVID-19 (Moghaddam et al., 2020). Se deficiency in China has been marked by causing Keshan disease, endemic heart disease with high lethality (Chen, 2012).

Malnutrition is growing and the estimative of world population will be 9 billion people in 2030, and developing countries are the ones that will suffer most from malnourishment problem (Khush et al., 2012). One strategy to reduce malnutrition is through agronomic biofortification, which is a technique of fertilizer application to edible crop plants. Agronomic biofortification aims to increase the concentration of micronutrients in the edible parts of crops (Khush et al., 2012; White, 2018; Reis et al., 2020; Lanza and Reis, 2021). In addition to improving the nutritional quality of foods, biofortification is used to combat hidden hunger mainly in developing countries (De Valença et al., 2017; Reis et al., 2017).

Se can benefit plant growth under abiotic stresses by mitigation of reactive oxygen species (ROS) formation. However, Se application at high concentration can cause toxicity in plants (Sun et al., 2010; Malik et al., 2011; Silva et al., 2018; White, 2018). Se can mitigate oxidative stress by regulating ROS through enzymatic and non-enzymatic antioxidant systems, and regulation of the photosynthetic apparatus (Chauhan et al., 2019; Silva et al., 2020). In addition, several studies have shown a positive effect of Se on the enzymatic antioxidant metabolism by increasing the activity of superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX), and glutathione peroxidase (GPx) in plants (Silva et al., 2018; Chauhan et al., 2019; Silva et al., 2020; Lanza and Reis, 2021). Se increases photosynthetic pigments such as chlorophylls and stimulate carotenoids biosynthesis, as well as stomatal conductance and internal CO_2 concentration (Figure 1) (Bian et al., 2020; Silva et al., 2020; Zhang et al., 2020; Lanza and Reis, 2021).

Application of high Se concentration results in an antagonistic effect with sulfur (S) affecting plant metabolism (White et al., 2004; Silva et al., 2022). Se and S compete for metabolic pathways, where Se affects the expression of key genes governing S transporters and enzymes responsible for S assimilation in plants (White, 2018). The accumulation of Se in plant tissues can influence physiological functions of S metabolites, which can affect the development, growth, and response of plants to abiotic and biotic stresses (White, 2018).

Tian et al. (2017) observed broccoli plants growing in hydroponics with 40 µM of Se showed increased expression of S transporter (SULTR 1;1) In addition, these authors found that Se increased the activity of ATP-sulfurylase and adenylyl-sulfate (APS) reductase, which are two key enzymes in the S/Se assimilation pathway. Boldrin et al. (2016) reported that application of 10 µM of Se as sodium selenate increased the transcription of SULTR1; 1, SULTR1; 3 and SULTR4; 1 in wheat roots, suggesting that supplementation with Se mimics the deficiency of S pellet in the expression of S-transporting genes and does not increase the accumulation of S caused by the presence of Se.

Se can enhance nitrogen (N) metabolism by the indirect effect on the activity of enzymes that act on N metabolism (Prasad and Chetty, 2008; Lei et al., 2018; Bian et al., 2020). Se enhances the activity of nitrate reductase (NR), glutamine synthetase (GS), and glutamate synthase (GOGAT) due to coordination between N and sulfur S metabolism (Rios et al., 2010b). Fertilization of plants with Se induces the assimilation of S and Se, stimulating N metabolism to maintain the proper balance between nitrogenous organic compounds and sulfates (Rios et al., 2010b). Furthermore, Se can increase NR activity by increasing the absorption of molybdenum, which is cofactor for NR activity in plant cell (Figure 2).

Se fertilization increase total soluble solids, which play an important role in photosynthesis and carbohydrate production (Mimmo et al., 2017; Golubkina et al., 2018; Gouveia et al., 2020). In addition, Se increases the concentration of organic acids, phenolic compounds (Mimmo et al., 2017), photosynthetic pigments (Silva et al., 2020), amino acids (Jezek et al., 2011), and proteins (Silva et al., 2020) which are compounds responsible for growth and yield of crops.

This literature review aimed to present a compilation of main findings on how Se can be a secret weapon in agriculture to increase the tolerance of plants to abiotic stresses by increasing the antioxidant and sugar metabolism, photosynthetic and nutritional status of plants, increasing yield as well as the quality of agricultural products which might benefit human health.

CONCLUSION

Mechanisms of soybean response to Se were related to increased photosynthetic pigments as well as enhanced efficiency of enzymatic antioxidant metabolism by increasing the activity of CAT and APX enzymes. Se also increased the sugars concentration in the leaves and influenced the metabolism of N by increasing the activity of nitrate reductase and average nodule fresh weight, which increased the synthesis of ureides and the bioavailability of amino acids and proteins in the leaves, resulting in higher root dry weight. However, the responses were dose-dependent, and the form of dose response varied among traits.

Our results revealed hormetic-like responses of soybean plants exposed to Se at both molecular and whole-plant levels. Hormetic-like U or inverse U shape responses were revealed for photosynthetic pigments, CAT, nitrate reductase, nodules per pot, nodule fresh weight, and amino acids. However, the concentration of flavonoids in the soybean roots showed dynamic, multiphasic responses to Se. New studies are needed to reveal the biochemistry behind these hormetic-like responses.

This study presents new insights into the effects of Se on N metabolism, enzymatic antioxidant metabolism, photosynthetic pigments, and nodulation in leguminous plants that may be important for increasing soybean yield.

REFERENCES

Agathokleous E, Feng Z, Peñuelas J (2020) Chlorophyll hormesis: are chlorophylls major components of stress biology in higher plants?. **Science of Total Environment** 726: 138637. https://doi.org/10.1016/j.scitotenv.2020.138637

Agathokleous E (2021) The rise and fall of photosynthesis: hormetic dose response in plants. **Journal of Forest Research** 32: 889-898. https://doi.org/10.1007/s11676-020-01252-1

Agathokleous E, Kitao M, Koike T (2022) Testing phaeophytinization as an index of ozone stress in trees. **Journal of Forest Research** 1-8 https://doi.org/10.1007/s11676-022-01556-4

Alexieva V, Sergiev I, Mapelli S, Karanov E (2001) The effect of drought and ultraviolet radiation on growth and stress markers in pea and wheat. **Plant Cell & Environment** 24: 1337-1344. https://doi.org/10.1046/j.1365-3040.2001.00778.x

Andrade FR, da Silva GN, Guimarães KC, Barreto HBF, de Souza KRD, Guilherme L RG, Dos Reis AR (2018) Selenium protects rice plants from water deficit stress. **Ecotoxicology and Environmental Safety** 164: 562-570. https://doi.org/10.1016/j.ecoenv.2018.08.022

Alves LR, Prado ER, de Oliveira R, Santos EF, Lemos de Souza I, Dos Reis AR, Gratão PL (2020) Mechanisms of cadmium-stress avoidance by selenium in tomato plants. **Ecotoxicology** 29: 594-606. https://doi.org/10.1007/s10646-020-02208-1

Azevedo RAD, Alas RM, Smith RJ, Lea PJ (1998) Response of antioxidant enzymes to transfer from elevated carbon dioxide to air and ozone fumigation, in the leaves and roots of wild-type and a catalase-deficient mutant of barley. **Physiologia Plantarum** 104: 280-292. https://doi.org/10.1034/j.1399-3054.1998.1040217.x

Baral B, da Silva JAT, Izaguirre-Mayoral ML (2016) Early signaling, synthesis, transport and metabolism of ureides. **Journal of Plant Physiology** 193: 97-109. https://doi.org/10.1016/j.jplph.2016.01.013

Bhadwal S, Sharma S (2022) Selenium alleviates physiological traits, nutrient uptake and nitrogen metabolism in rice under arsenate stress. **Environmental Science and Pollution Research** 1-20. https://doi.org/10.1007/s11356-022-20762-5

Bo LEI, Bian ZH, Yang QC, Jun WANG, Cheng RF, Kun LI, Tong YX (2018) The positive function of selenium supplementation on reducing nitrate accumulation in hydroponic lettuce (*Lactuca sativ*a L.). **Journal of Integrative Agriculture** 17(4): 837-846.https://doi.org/10.1016/S2095-3119(17)61759-3

Bradford MM (1976) A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. **Analytical Biochemistry** 72: 248-254. https://doi.org/10.1016/0003-2697(76)90527-3

Bian ZH, Bo LEI, Cheng RF, Yu WANG, Tao LI, Yang QC (2020) Selenium distribution and nitrate metabolism in hydroponic lettuce (*Lactuca sativa* L.): Effects of selenium forms and light spectra. **Journal of Integrative Agriculture** 19: 133-144.https://doi.org/10.1016/S2095-3119(19)62775-

Bieleski RL, Turner NA (1966) Separation and estimation of amino acids in crude plant extracts by thin-layer electrophoresis and chromatography. **Analytical Biochemistry** 17: 278-293. https://doi.org/10.1016/0003-2697(66)90206-5

Bo LEI, Bian ZH, Yang QC, Jun WANG, Cheng RF, Kun LI, Tong YX (2018) The positive function of selenium supplementation on reducing nitrate accumulation in hydroponic lettuce (*Lactuca sativa* L.). **Journal of Integrative Agriculture** 17: 837-846.https://doi.org/10.1016/S2095-3119(17)61759-3

Bocchini M, D'Amato R, Ciancaleoni S, Fontanella MC, Palmerini CA, Beone GM, Businelli D (2018) Soil selenium (Se) biofortification changes the physiological, biochemical and epigenetic responses to water stress in *Zea mays* L. by inducing a higher drought tolerance. **Frontiers in Plant Science** 9: 389.https://doi.org/10.3389/fpls.2018.00389

Bosse MA, da Silva MB, de Oliveira NGRM, de Araujo MA, Rodrigues C, de Azevedo JP, Dos Reis AR (2021) Physiological impact of flavonoids on nodulation and ureide metabolism in legume plants. **Plant Physiology and Biochemistry** 166: 512-521.

Cataldo DA, Maroon M, Schrader LE, Youngs VL (1975) Rapid colorimetric determination of nitrate in plant tissue by nitration of salicylic acid. **Communications in Soil Science and Plant Analysis** 6: 71-80. https://doi.org/10.1080/00103627509366547

Campos JVL, Luís Oliveira Cunha M, do Nascimento V, de Figureueiredo PAM, Ferrari S (2022) Can foliar application of nutrients increase the productive potential of peanuts?. **Gesunde Pflanzen** 74: 1-5. https://doi.org/10.1007/s10343-022-00739-7

Campbell WH (1988) Nitrate reductase and its role in nitrate assimilation in plants. **Physiologia Plantarum** 74: 214-219. https://doi.org/10.1111/j.1399-3054.1988.tb04965.x

Castillo-Godina RG, Foroughbakhch-Pournavab R, Benavides-Mendoza A (2016) Effect of selenium on elemental concentration and antioxidant enzymatic activity of tomato plants. J Agric Sci Technol 18(1): 233-244.

Chakrabarti M, Mukherjee A (2021) Metallo-adaptive response: a unique survival strategy of plants under genotoxic stress. **Nucleus**, 1-8. https://doi.org/10.1007/s13237-021-00360-5

Cheng B, Lian H, Liu Y, Yu X, Sun Y, Sun X, Liu S (2016) Effects of selenium and sulfur on antioxidants and physiological parameters of garlic plants during senescence. **Journal of Integrative Agriculture** 15: 566–572. doi: http://doi.org/10.1016/s2095-3119(15)61201-1

Cunha MLO, de Oliveira LCA, Silva VM, Montanha GS, Dos Reis AR (2022) Selenium increases photosynthetic capacity, daidzein biosynthesis, nodulation and yield of peanuts plants (*Arachis hypogaea* L.). **Plant Physiology and Biochemistry** 190: 231-239. https://doi.org/10.1016/j.plaphy.2022.08.006

Carara GDL, Cunha MLO, do Nascimento V, dos Santos Batista Bonini C, Pereira Prado E, Ferrari S (2022) Effects of Application of Silicon Doses and Irrigation On the Photosynthetic Parameters of Cotton. **Gesunde Pflanzen** 74: 1-6. https://doi.org/10.1007/s10343-022-00699-y

De Brito Mateus MP, Tavanti RFR, Galindo FS, da Rocha Silva AC, Gouveia GCC, Aparecido CFF, Dos Reis AR (2020) *Coffea arabica* seedlings genotypes are tolerant to high induced selenium stress: Evidence from physiological plant responses and antioxidative performance. **Ecotoxicology and Environmental Safety** 203: 111016. https://doi.org/10.1016/j.ecoenv.2020.111016

De Brito Mateus MP, Tavanti RFR, Tavanti TR, Santos EF, Jalal A Dos Reis AR (2021) Selenium biofortification enhances ROS scavenge system increasing yield of coffee plants. **Ecotoxicology and Environmental Safety** 209: 111772. https://doi.org/10.1016/j.ecoenv.2020.111772

Dubois M, Gilles KA, Hamilton JK, Rebers PT, Smith F (1956) Colorimetric method for determination of sugars and related substances. **Analytical Chemistry** 28: 350-356

Ende WVD, Peshev D (2013) Sugars as antioxidants in plants. In **Crop improvement under adverse conditions** (pp. 285-307). Springer, New York, NY.

Ferrari S, Marsala L, Oliveira Cunha ML, dos Santos Cordeiro LF, Tropaldi L, de Mattos Barretto VC, Alves de Oliveira LC (2021a) Can the application of low doses of glyphosate induce the hormesis effect in upland rice?. **Journal of Environment Science and Health Part B** 56: 814-820. https://doi.org/10.1080/03601234.2021.1957372

Ferrari S, Marsala L, Luis Oliveira Cunha M (2021b) Effects of Low Doses of Glyphosate on Agronomic Traits of Upland Rice. **Gesunde Pflanzen** 73: 533-539. https://doi.org/10.1007/s10343-021-00573-3

Gouveia GCC, Galindo FS, Lanza MGDB, da Rocha Silva AC, de Brito Mateus MP, da Silva MS, Dos Reis AR (2020) Selenium toxicity stress-induced phenotypical, biochemical and physiological responses in rice plants: Characterization of symptoms and plant metabolic adjustment. **Ecotoxicology and Environmental Safety** 202: 110916. https://doi.org/10.1016/j.ecoenv.2020.110916

Giannopolitis CN, Ries SK (1977) Superoxide dismutases: I. Occurrence in higher plants. **Plant Physiology** 59: 309-314. https://doi.org/10.1104/pp.59.2.309

Gomes-Junior RA, Gratão PL, Gaziola SA, Mazzafera P, Lea PJ, Azevedo RA (2007) Selenium-induced oxidative stress in coffee cell suspension cultures. **Functional Plant Biology** 34(5): 449-456. https://doi.org/10.1071/FP07010

Hasanuzzaman M, Bhuyan MB, Zulfiqar F, Raza A, Mohsin SM, Mahmud JA, Fotopoulos V (2020) Reactive oxygen species and antioxidant defense in plants under abiotic stress: Revisiting the crucial role of a universal defense regulator. **Antioxidants** 9(8): 681. https://doi.org/10.3390/antiox9080681

Heath RL, Packer L (1968) Photoperoxidation in isolated chloroplasts: I. Kinetics and stoichiometry of fatty acid peroxidation. **Archives of Biochemistry and Biophysics** 125(1): 189-198. https://doi.org/10.1016/0003-9861(68)90654-1

Ikram M, Raja NI, Javed B, Hussain M, Hussain M, Ehsan M, Akram A (2020) Foliar applications of bio-fabricated selenium nanoparticles to improve the growth of wheat plants under drought stress. **Green Processing Synthesis** 9: 706-714.https://doi.org/10.1515/gps-2020-0067
Jalal A, de Oliveira Junior JC, Ribeiro JS, Fernandes GC, Mariano GG, Trindade VD R, Dos Reis AR (2021) Hormesis in plants: Physiological and biochemical responses. **Ecotoxicology and Environmental Safety** 207: 111225. https://doi.org/10.1016/j.ecoenv.2020.111225

Kapoor D, Singh S, Kumar V, Romero R, Prasad R, Singh J (2019) Antioxidant enzymes regulation in plants in reference to reactive oxygen species (ROS) and reactive nitrogen species (RNS). **Plant Gene** 19: 100182. https://doi.org/10.1016/j.plgene.2019.100182

Khan N, Ali S, Zandi P, Mehmood A, Ullah S, Ikram M, Babar MA (2020) Role of sugars, amino acids and organic acids in improving plant abiotic stress tolerance. **Pakistan Journal of Botany** 52: 355-363.

Łabanowska M, Filek M, Kościelniak J, Kurdziel M, Kuliś E, Hartikainen H (2012) The effects of short-term selenium stress on Polish and Finnish wheat seedlings—EPR, enzymatic and fluorescence studies. **Journal of Plant Physiology** 169: 275-284. https://doi.org/10.1016/j.jplph.2011.10.012

Lanza MGDB, Silva VM, Montanha GS, Lavres J, de Carvalho HWP, Dos Reis AR (2021) Assessment of selenium spatial distribution using µ-XFR in cowpea (*Vigna unguiculata* (L.) Walp.) plants: Integration of physiological and biochemical responses. **Ecotoxicology and Environmental Safety** 207: 111216. https://doi.org/10.1016/j.ecoenv.2020.111216

Lara TS, de Lima Lessa JH, de Souza KRD, Corguinha APB, Martins FAD, Lopes G, Guilherme LRG (2019) Selenium biofortification of wheat grain via foliar application and its effect on plant metabolism. **Journal of Food Composition and Analysis** 81: 10-18.https://doi.org/10.1016/j.jfca.2019.05.002

Lichtenthaler HK (1987) Chlorophylls and carotenoids: pigments of photosynthetic biomembranes. **Methods in Enzymology** 148: 350-382. https://doi.org/10.1016/0076-6879(87)48036-1

Marchica A, Ascrizzi R, Flamini G, Cotrozzi L, Tonelli M, Lorenzini G, Pellegrini E (2021) Ozone as eustress for enhancing secondary metabolites and bioactive properties in *Salvia officinalis*. **Industrial Crops Products** 170: 113730. https://doi.org/10.1016/j.indcrop.2021.113730

Marsala L, Oliveira Cunha ML, do Nascimento V, Pereira Prado E, da Silva Viana R, Ferrari S (2022) Can 2, 4-D promote the hormesis effect in upland rice?. **Journal of Environmental Science Health Part B** 57: 680-685.https://doi.org/10.1080/03601234.2022.2099687

McCullough H (1967) The determination of ammonia in whole blood by a direct colorimetric method. **Clinica Chimica Acta** 17: 297-304. https://doi.org/10.1016/0009-8981(67)90133-7

Moldes CA, Medici LO, Abrahao OS, Tsai SM, Azevedo RA (2008) Biochemical responses of glyphosate resistant and susceptible soybean plants exposed to glyphosate. **Acta Physiologia Plantarum** 30: 469-479. https://doi.org/10.1007/s11738-008-0144-8

Peng WT, Zhang LD, Zhou Z, Fu C, Chen ZC, Liao H (2018) Magnesium promotes root nodulation through facilitation of carbohydrate allocation in soybean. **Physiologia Plantarum** 163: 372-385. https://doi.org/10.1111/ppl.12730

Reis HPG, Barcelos JPQ, Junior EF, Santos EF, Silva VM, Moraes MF, Reis AR (2018) Agronomic biofortification of upland rice with selenium and nitrogen and its relation to grain quality. **Journal of Cereal Science** 79: 508–515. doi: http://doi.org/10.1016/j.jcs.2018.01.004

Saddhe AA, Manuka R, Penna S (2021) Plant sugars: Homeostasis and transport under abiotic stress in plants. **Physiologia Plantarum** 171: 739-755. **https://doi.org/10.1111/ppl.13283**

Sali A, Zeka D, Fetahu S, Rusinovci I, Kaul HP (2018) Selenium supply affects chlorophyll concentration and biomass production of maize (L.). **Journal of Land**

Management, Food and Environment 69: 249-255. https://doi.org/10.2478/boku-2018-0021

Shahid MA, Balal RM, Khan N, Zotarelli L, Liu GD, Sarkhosh A, Garcia-Sanchez F (2019) Selenium impedes cadmium and arsenic toxicity in potato by modulating carbohydrate and nitrogen metabolism **Ecotoxicology and Environmental Safety** 180: 588-599.https://doi.org/10.1016/j.ecoenv.2019.05.037

Silva VM, Boleta EHM, Lanza MGDB, Lavres J, Martins JT, Santos EF, dos Reis AR (2018) Physiological, biochemical, and ultrastructural characterization of selenium toxicity in cowpea plants. **Environmental and Experimental Botany** 150: 172-182. https://doi.org/10.1016/j.envexpbot.2018.03.020

Silva VM, Tavanti RFR, Gratão PL, Alcock TD, Dos Reis AR (2020) Selenate and selenite affect photosynthetic pigments and ROS scavenging through distinct mechanisms in cowpea (*Vigna unguiculata* (L.) walp) plants. **Ecotoxicology and Environmental Safety** 201: 110777. https://doi.org/10.1016/j.ecoenv.2020.110777

Smoleń S, Skoczylas Ł, Rakoczy R, Ledwożyw-Smoleń I, Liszka-Skoczylas M, Kopeć A, Sady W (2015) Selected aspects of nitrogen metabolism and quality of field-grown lettuce (*Lactuca sativa* L.) depending on the diversified fertilization with iodine and selenium compounds. **Acta Scientiarum Polonorum Hortorum Cultus Ogrod** 14: 159-175.
Vogels GD, Van der Drift C (1970) Differential analyses of glyoxylate derivatives. **Analytical Biochemistry** 33: 143-157. https://doi.org/10.1016/0003-2697(70)90448-3

Zhang M, Tang S, Huang X, Zhang F, Pang Y, Huang Q, Yi Q (2014) Selenium uptake, dynamic changes in selenium content and its influence on photosynthesis and chlorophyll fluorescence in rice (*Oryza sativa* L.). **Environmental and Experimental Botany** 107: 39-45. https://doi.org/10.1016/j.envexpbot.2014.05.005

Yemm EW, Cocking EC, Ricketts RE (1955) The determination of amino-acids with ninhydrin. **Analyst** 80: 209-214. https://doi.org/10.1039/AN9558000209