

Research Article



Biological effects of alginite on tomato plants (Lycopersicon esculentum) and some insects (Leptinotarsa decemlineata, Galleria mellonella and Halyomorpha halys)

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@ • • •	Article Details:					
BY NC	Received:	2021-10-01				
	Accepted:	2021-11-19				
	Available online:	2021-11-30				

DOI: https://doi.org/10.15414/ainhlq.2021.0033

The purpose of our work was to study the effect of Slovak origin alginite on germination of tomato seeds, growth and development of plants, as well as to evaluate biological activity of alginite against to insect pests. Tomato seeds with low germination capacity were treated by immersion in alginite solutions of concentration 0.0001–0.1 %. Alginite solutions with concentrations of 0.1–5.0 % were used for testing against insects. The application of alginite solutions significantly increases the germination of tomato seeds in laboratory conditions by 9.2–13.0 %. There was a significant increase in the length of seedlings and roots by 1.87-2.98 cm and 3.54-4.51 cm. In a greenhouse presowing treatment of tomato seeds with 0.001 % alginite solution significantly increased germination in comparison with the control (by 33.2 %) and 0.01 and 0.1 % alginite solutions (by 22.2 and 25.0 %). Monitoring of plant height showed that a month after the first seedlings emergence the average plant height was significantly higher in the variant 0.001 % alginite solution and reached to 11.41 cm. The yield of fruits in the variants treated with alginite solutions significantly exceeded the control by 316.53–327.71 g per one tomato bush. It was found that alginite solutions at a concentration of 0.1-1.0 % had low ovicidal (2.73-13.19 %) and insecticidal (5.0-33.3 %) effects against insects belonging to different orders - Leptinotarsa decemlineata (Coleoptera), Galleria mellonella (Lepidoptera) and Halyomorpha halys (Hemiptera). Alginite solutions did not have contact insecticidal activity; the death of insects was caused mainly by the consumption of treated feed. At the same time, a high antifeedant effect was revealed from 45.0 to 85.0 %against adults and larvae of 2-3 instars of L. decemlineata and larvae of 2-3 instars of G. mellonella. To conclude, the application of alginite contributed to an increase in seed germination and plant productivity, and it did not reveal significant biological activity against insects.

Keywords: alginite, seeds germination, plant growth, yield, insect biological activity

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Introduction

In modern conditions of climatic anomalies, it is extremely important to achieve stable production of high yields of significant agricultural crops. One of the leading places among vegetables belongs to tomatoes *Lycopersicon esculentum* Mill. Achieving high germination of seeds is possible with the integrated use of all agrotechnical methods, including the use of fertilizers, plant growth and development regulators. Alginite is considered one of the most promising materials with several of useful properties.

Alginite is an organic-bituminous rock, rich in macro- and microelements. Alginite arose as a result of the fossilization of accumulated organic (algae) and inorganic material, especially clay, carbonates, quartz and amorphous modification of silicic acid in the aqueous environment (Gancarčikova et al., 2019; Brindza et al., 2021a). In the Slovak Republic an alginite deposit (maar belonging to the Podrecany Basalt Formation, Pontian in age, approx. 6.5 m.y. B.C.), was discovered near Pinciná, close to the town of Lučenec, the center of the Novohrad region (Motyleva et al., 2014). Despite the fact that alginite contains trace elements, the toxicity of heavy metals is below the toxicity limit. The alginite has high water absorption capacity - up to 110 % and high specific surface (313-654 m² special features of the organic matter (kerogen type II) and by the presence of smectite (Brindza et al., 2021a). According to many authors, alginite, due to its exceptional chemical composition, improves the soil properties, thereby affecting the growth and development of plants, contributes to increase yields (Rauch and Földényi, 2012; Kádár et al., 2015; Benei and Rauch, 2016; Bednárová, 2019; Kropp et al., 2021).

The purpose of our work was to study the effect of Slovak origin alginite on germination of tomato seeds, growth and development of plants, as well as to evaluate biological activity of alginite against to insect pests.

Material and methodology

The experiments were carried out in laboratory conditions and in a greenhouse during 2020.

Preparation of alginite solutions

Alginite solutions at a concentration of 0.0001, 0.001, 0.01, 0.01, 0.1, 1.0 and 5.0 % were prepared using distilled water in calibrated flasks. Alginite powder obtained from Slovak Republic was weighed on an analytical balance, and transferred quantitatively into a calibrated flask. Thereafter, the solution was thoroughly mixed

and used immediately. The solution was thoroughly mixed before each use. Distilled water was used as a control.

Effect of alginite solution on seed germination

The influence of alginite on capacity of seed germination was studied in laboratory conditions using tomato seed with low germination rate. The seeds of tomatoes (*Lycopersicon esculentum* Mill.) were treated by immersion for 15 minutes in alginite solutions of concentration 0.0001, 0.001, 0.01, and 0.1 %. Then the seeds were germinated for 10 days in Petri dishes between moistened filter paper disks in thermostat at temperature of 25 °C (Rao et al., 2006, ISTA, 2017). Each variant consisted of four replicates, 100 seeds per replicate. Seeds treated with water served as a control. The total germination rate, the length of roots and seedlings were determined.

Determination of the effect of alginite solutions on plant growth

To determine the effect of alginite on growth of tomato plants, the tomato seeds were treated presowing by immersion for 24 hours in alginite solutions of concentration (0.1, 0.01 and 0.001 %). Treated seeds were sowed in greenhouse with drip irrigation in holes according to a randomized scheme. Each variant consisted of three replicates, 36 seeds per replicate. The indices of germination, plant height and yield from one tomatoes bush were analyzed.

Determination of the biological activity of alginite solutions in relation to insect test-objects from the orders Coleoptera, Lepidoptera and Hemiptera

The ovicidal, insecticidal and antifeedant properties of 0.1–5.0% alginite solutions were determined according to standard methods in relation to insects belonging to different orders: Leptinotarsa decemlineata Say (Coleoptera), Galleria mellonella L. (Lepidoptera) and Halyomorpha halys Stal (Hemiptera) (Elisovetcaia et al., 2020). The contact effect of the extracts was determined by the method of topical application of 0.6 μ l of alginite solutions to the dorsal surface of insects. The intestinal effect of the extracts was determined by immersion a nutritive substrate (potato leaves) into alginite solutions, as well as by introducing alginite solutions of the appropriate concentration into an artificial nutrient medium (ANM). Variants with treatment using distilled water served as control. The calculation of ovicidal activity was carried out according to the correction for the number of sterile eggs in the control.

Statistical analysis

Data analysis for determination of standard deviation, significant differences and correlation coefficients were performed by Statgraphics Plus 5.0 programme.

Results and discussion

The treatment with alginite solutions significantly increased the germination of tomato seeds in laboratory conditions in comparison with the control by 9.2–13.0 % (LSD $_{\scriptscriptstyle 0.05}$ = 8.82, p ≤0.05) (Table 1). The highest seed germination (81.3 %) was observed in the variant with the alginite concentration of 0.001 %, the lowest (77.5 %) – in the variant with the 0.1 % alginite concentration. There was no significant difference between the variants with alginite solutions treatment of various concentrations, but it was found that with a decrease in concentration from 0.1 to 0.01 and 0.001 %, the germination of tomato seeds insignificantly increases - by 1.0 and 3.8%, respectively. Seed germination in the variant with the 0.0001 % alginite concentration is 1.3-2.3 % less than in the variants 0.001 and 0.01 %, respectively, but at the same time it is 1.5 % higher than in the 0.1 % alginite variant and significantly exceeds the control (by 10.7 %).

Kovár et al. (2021) found that powder, crushed alginite and alginite extracts (sodium solution, potassium solution) have a positive effect on seeds germination of of *Poa pratensis* L., increasing the average germination in comparison with the control by 33.33–334.20 %, as well as alginite and its products increase the germination rate by 0.04–1.52 seeds/day in *P. pratensis*. These data are also confirmed by our results obtained with tomato seeds. Thus, it is obvious that different preparative forms of alginite promote better germination of seeds of various species of cultivated plants.

In our experiments, the treatment of tomato seeds with alginite solutions led to an increase in the length of seedlings and roots in all variants in comparison with the control. At the same time, the most significant increase in the length of seedlings by 18.7 and 29.8 mm $(LSD_{0.05} = 16.1, p \le 0.05)$ was observed in two variants – at a concentration of alginite solutions of 0.01 and 0.1 % (Table 1). A significant increase in root length by 22.2, 35.4 and 45.1 mm was observed in three variants of treatment with alginite solutions – with a concentration of 0.001, 0.01 and 0.1 %, respectively $(LSD_{0.05} = 15.5, p \le 0.05)$ (Table 1).

Thus, it was found that 0.1, 0.01, and 0.001 % alginite solutions led to both a significant increase in the germination of tomato seeds and an increase in the length of seedlings and roots. Despite the fact that alginite at a concentration of 0.0001 % contributes to a significant increase in seed germination, the difference in the length of roots and seedlings is insignificant in comparison with the control.

Therefore, for the presowing treatment of tomato seeds, it were selected three of the four tested concentrations – 0.1, 0.01 and 0.001 % alginite solutions. The first seedlings emergence was noted on the 11^{th} day after sowing, the last – on the 19^{th} day after sowing.

The total germination of seeds in all variants of greenhouse experiments exceeded the control by 8.3–33.2 %. However, a significant difference (LSD_{0.05} = 20.5, p ≤0.05) was noted only in the variant where the seeds were treated with 0.001 % alginite solution (Figure 1).

According to many authors (Ognjanova-Rumenova and Vaas, 1998; Vass, 1998; Kulich et al., 2001; Pichler et al., 2001; Brinza et al., 2021a), alginite contains a sufficient amount of humic acids, which, as heterogeneous organic compounds, are capable of improving soil fertility. Due to its water-saving properties, alginite is one of the best water-absorbing materials that can regulate the distribution of water towards the plant roots. Probably, these properties also play an important role in seed germination – supplying tomato seeds with nutrients, and retain and distribute moisture, thus creating optimal conditions for swelling and germination of seeds.

Variants	Germination (%)	Seedlings length (mm)	Roots length (mm)		
Control	68.3	15.4	16.9		
Alginite 0.0001 %	79.0	22.9	23.7		
Alginite 0.001 %	81.3	30.8	39.1		
Alginite 0.01 %	80.3	34.1	52.3		
Alginite 0.1 %	77.5	45.2	61.9		
LSD _{0.05}	8.82	16.1	15.5		

 Table 1
 Germination characteristics of tomato seeds treated with different concentrations of alginite

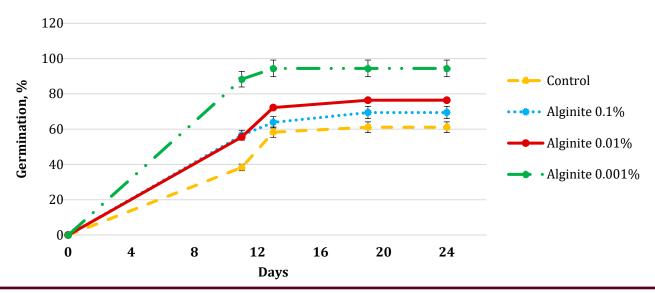
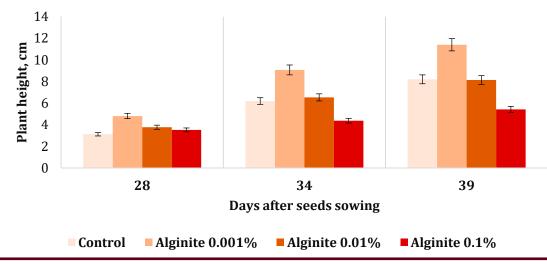


Figure 1 Germination rate of tomato seeds treated with alginite solutions in greenhouse conditions, 2020

Monitoring of plant height showed that a month after the first seedlings emergence the average of plant height was significantly higher in the variant 0.001 % alginite solution and reached to 11.41 cm ($LSD_{0.05} = 2.63$, p ≤ 0.05). The plant heights in other tested variants were at the control level (8.21 cm) (Figure 2).

In the experiment, ALGEX, 6 was applied by Horčinová Sedláčková et al. (2021) in the form of a watering in two variants with the same concentration of 3 % solution in 2 deciliters of water, but various application in terms of days in the pre-harvest stage of the aboveground plant biomass of 30 individual plants from each species. There are two diametrically opposite trends of ALGEX, 6 application that are manifesting themselves in *Melissa officinalis* and *Malva verticillata* by reducing the root and above-ground part biomass compared to the control variant. The percentage proportionality of root/ above-ground part biomass in *M. officinalis* decreased from 62.48/30.31 % (control), to 45.57/18.85 % (variant 1) and to 36.07/17.27 % (variant 2), as well as in *M. verticillata* the root/above-ground part biomass decreased from 16.03/13.93 % (control) to 14.97/9.42 % (variant 1) and to 11.61/10.14 % (variant 2). In the species *Ocimum* × *citriodorum* Vis. the opposite trend manifested (Horčinová Sedláčková et al., 2021).

In our experiments, it was found that in the variant with the treatment with 0.1 % alginite solution, the tomato plants looked depressed and lagged behind in growth from other variants by 2.51–5.71 cm. Also in this variant, within one and a half months after the appearance of the first seedlings, an average 30 % of plants was died. Therefore, the variant of presowing





seeds treatment with 0.1 % alginite solution was excluded from the experiment.

Cukor et al. (2017) studied growth parameters (height increment, mortality and foliar nutrient content) of Douglas fir (*Pseudotsuga menziesii* (Mirb.) Franco), Scots pine (*Pinus sylvestris* L.) and a mixture of English oak (*Quercus robur* L.), red oak (*Quercus rubra* L.) and Norway maple (*Acer platanoides* L.) seedlings on former agricultural land in central Bohemia, Czech Republic. The results showed that alginite application had greater positive effect on height growth of seedlings than mortality, especially variant C. In most of the cases height increments were significantly positively affected (p <0.05) by both variants of alginite application only in the third year after planting.

Bio-Algeen system (materials based on the marine algae, both of fossil and recent origin) was tested by Kupka et al. (2015) at planting stock of Norway spruce production in a forest nursery. In the plantation experiment, significantly lower mortality was documented in the first year since planting, as well as significantly faster growth for broad-leaved species. In the nursery experiment, considerably more favourable development of the root system was detected as well as better parameters of the above-ground part of the planting stock. As the most effective, the combination of root dipping to Bio-Algeen water solution with granulate application on bed surface and also spraying of aboveground part of seedlings after transplanting were documented. Applications of studied material thus represent important contribution for quality stock production and plantation success on the forested site.

Biochar application to soils is being considered as means to improve fertility while concurrently improving soil functions. Based on results, authors found that biochar increased the microbial biomass values even before the incubation. In single and combined biochar–alginite treatments, more bacterial biomass was adsorbed due to the higher adhesion capability and the increased surface area. The volume of the microbial adsorption is different from species to species and even strains (Kocsis et al., 2020).

The spring and summer seasons of 2020 were characterized by extreme temperatures. Thus, already in the first decade of June, the temperature reached 27–35 °C, and on some days 40-42 °C. In July, the average daily temperatures were even higher, and in August, the mark reached an extreme value 45 °C. In the greenhouse, even with the use of shading green textile and ventilation, the temperature exceeded

35 °C and often reached to 45 °C and more. These conditions significantly affected the ovary of plants. It is known that the pollination of tomato culture occurs mainly during the day, but the ripening of pollen - at night. In this case, the temperature at night should be in the range from +13 to +25 °C. During the day, the temperature indicators should not exceed +35 °C. At temperatures above, tomato pollen becomes sterile, the bush sheds flowers, and ovaries are not formed. Since the temperature conditions in 2020 during the summer season, which saw the flowering of tomatoes, significantly exceeded these indicators, the yield in 2020 was record low. In the control variant, the formation of fruits was sporadic. Plants treated with alginite formed significantly more fruits, which ultimately had a positive effect on the yield. The yield in the variants treated with alginite solutions significantly exceeded the control by 316.53–327.71 g per one tomato bush.

Earlier, Brindza et al. (2021b), investigating the effect of various of alginite preparations (ultrasound-treated alginite), found that they resulted in a reduction in plant weight as well as a reduction in the average fruit weight compared to the control variant. However, our research has shown a significant increase in yield. Probably, the discrepancy between the results can be associated with the use of completely different alginite preparations: we used aqueous solutions, while Brindza et al. (2021b) – solutions from alginite treated with ultrasound.

Thus, the results of our studies showed that alginite solutions at a concentration of 0.0001-0.1 % increase the germination of tomato seeds, promote an increase in seedlings and roots, and significantly increase the growth rate and productivity of plants. However, we did not add any form of alginite to the soil, but we believe that such research is worth doing in the future. It is also worth studying the effect of foliar treatments on plant growth and development. At the same time, it was reported (Gömöryová et al., 2009; Rauch and Földényi, 2012; Oravcová et al., 2018) that the introduction of alginite into the soil improves the condition of both the soil and plants. Therefore, the future research of alginite effects on the growth and development of plants by foliar treatments will be relevant. However, the treatment of plants during the growing season can affect the arthropod complex in the agrocenosis of tomatoes. This is due to the fact that, both in open field conditions and greenhouses, alginite solutions not only directly fall on insects in the agrocenosis of crops when processing plants, but can also dry out and remain on the surface of the leaf apparatus, getting inside phytophagous insects with food. Therefore,

it is advisable to study the effect of alginite solutions on some common insects and phytophagous. Among pests at the moment in Europe, including the Republic of Moldova and the Slovak Republic, on Solanaceae crops such species as Leptinotarsa decemlineata Say (Coleoptera) and Helicoverpa armigera Hbn. (Lepidoptera: Noctuidae) are especially relevant (Elisovetcaia, 2010; Elisovetskaya and Nastas, 2012). In addition to these species, which have been known for a long time, a special role is played by the recently acclimatized, but already established themselves as extremely harmful species – the bugs Nezara viridula L. and Halyomorpha halys Stal. (Hemiptera) (Ivanova et al., 2020). Therefore, it was decided to select insect species belonging to these orders - Coleoptera, Lepidoptera, and Hemiptera – as test objects. Instead of the Helicoverpa armigera Hbn., the Galleria mellonella was selected. G. mellonella has a number of advantages over the cotton scoop: cultivation under laboratory conditions is more economical and less laborious, the degree of cannibalism in caterpillars of G. mellonella is significantly less pronounced than in caterpillars of cotton scoop. The marble bug was also selected based on the convenience of laboratory breeding.

It was revealed that alginite solutions at a concentration of 0.1-1.0 % had low ovicidal properties against *Halyomorpha halys* – 2.73 and 13.19 % in terms of the percentage of sterile eggs in the control (Table 2). The ovicidal effect is most likely due not to the toxicity of alginite itself, but to its physicochemical properties. Since alginite contains a clay fraction and is able to retain water, after processing the egg-laying, it covers the ovipositions like a slime film. After drying, such a film probably prevents both free access of air and the hatching of larvae from eggs.

The insecticidal activity of alginite solutions is low and is due to the intestinal action of the drug. Therefore, when treatment the feed, the death of imago and larvae of the Colorado potato beetle (L. decemlineata) was 10.0-20.0 and 20.0-30.0 %, respectively, and when feeding larvae of 2–3 ages *G. mellonella* with ANM with the addition of alginite solutions, the death of insects did not exceed 5.0-10.0 % (Table2). Probably, the insecticidal effect is due to the ingress of clay particles of alginite into the intestines of insects with feed, which subsequently leads to disruption of digestion processes and, as a consequence, to the death of insects. It was found that alginite solutions did not have contact activity with respect to insects - no death was observed with topical application of the solutions to the dorsal surface of insects (Table 2).

Despite the low insecticidal properties, alginite solutions against all species of insects showed a rather pronounced antifeedant effect (Table 2). Especially pronounced antifeedant properties were shown by 1.0-5.0 % solutions against imagoes *L. decemlineata* – 80.0-85.0 %. This is due to the fact that imagoes, unlike larvae, are able to tolerate longer hunger strikes. Therefore, the consumption of processed feed for imagoes decreased significantly during the experiment without significant damage to their survival. It was noted that, in comparison with the control, larvae of *G. mellonella* also significantly reduced feed consumption – by 51.24–68.92 %. We have previously

Table 2	Biological effects of alginite solutions on insects
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Variants	Haliomorpha halis				Le	Leptinotarsa decemlineata			Galleria mellonella	
	ovicidal activity (0A) (%)		insecticidal activity** (%) antifeedant activity (%)	insecticidal activity*** (%)		antifeedant activity (%)		insecticidal activity*** (%)	antifeedant activity (%)	
	proportion of non-hatching eggs	Ovicidal activity given sterile eggs in control	larvae 2 instars	larvae 2 instars	imago	larvae 2-3 instars	imago	larvae 2-3 instars	larvae 2-3 instars	larvae 2-3 instars
Control	8.3*		0	0	0	0	0	0	0	0
Alginite 0.1%	11.1	2.7	0	20.0	10.0	20.0	75.0	45.0	5.0	51.24
Alginite 1.0%	21.5	13.2	0	33.3	20.0	25.0	80.0	60.0	10.0	57.45
Alginite 5.0%	-	-	-	-	20.0	30.0	85.0	60.0	10.0	68.92

Notes: * proportion of sterile eggs, ** contact insecticidal activity, *** intestinal insecticidal activity

© Slovak University of Agriculture in Nitra www.uniag.sk found that extracts from plants *Juniperus sabina* L., Cupressaceae and *Pinus sylvestris* L., Pinaceae are able to reduce the feed intake of *G. mellonella* by 47.4–84.5 % (Elisovetcaia and Brindza, 2018; Elisovetcaia et al., 2019). The data obtained by us on the effect of 0.1–5.0 % alginite solutions on insect nutrition revealed a sufficiently high level of antifeedant activity, comparable to the effect of plant extracts (Elisovetcaia et al., 2020).

Alginite as a bituminous rock contains a high content of silicon (Vass et al., 1997). The role of silicon (Si) on plant health has been tested under open field conditions, hydroponic cultures, and under greenhouse/glasshouse environment (Luyckx et al., 2017). Still, presently there are a limited number of studies, which demonstrate there are advantages of Si application for greenhouse crops.

Meeting the growing demand for vegetables under situations of biotic and abiotic stresses is a big challenge. Si application is considered as an ecofriendly approach for crop production; therefore, Si application is commonly recommended under package and practices for cereals. Likewise, in vegetables, Si application has been documented to reduce the attack of diseases (Bakhat et al., 2018). For example, potassium silicate treatment of pea seedlings was observed to increase chitinase and β -1,3-glucanase activity against the fungal pathogen Mycosphaerella pinodes and it is the causes of blight disease in pea (Dann et al., 2002). Similarly, Si application has considerably reduced the root rot and powdery mildew disease in cucumber and the rust disease of cowpea (Heath and Stumpf, 1986; Chérif et al., 1994; Liang et al., 2005). Moreover, nano-silicon application can prevent postharvest diseases of vegetables (James and Zikankuba, 2017; Barman et al., 2018). In this direction, studies have also demonstrated that higher Si content in plant tissues reduced the incidence of several insect pests (Reynolds et al., 2009). Correa et al. (2005) reported that soil or as a foliar spray of Si as calcium silicate to cucumber plants increases the mortality of the nymphs of Bemisia tabaci.

In our experiment, we did not evaluate all the effects of alginite on tomatoes presented by Brindza et al. (2021b). However, it can be assumed that in addition to better seed germination, plant growth and other effects that we have determined when applying alginite to tomato plants, other effects can be determined. Therefore, it is useful to continue the experiments with the application of alginite to tomatoes.

Conclusions

It was found that 0.0001–0.1 % alginite solutions contribute to a significant increase in the germination of tomato seeds, as well as an increase in length of seedlings and roots in comparison with the control by 1.4–3.7 times. In a greenhouse, the treatment with 0.01–0.001 % alginite solutions not only increased seedlings emergence and plant height, but also contributed to a significant increase in yield compared to control. Possessing low insecticidal properties against insects, alginite solutions, at the same time lead to a decrease in damage to treated plants due to a high antifeedant effect, thereby preserving quality of above-ground part of plant and yield.

Conflict of interests

The authors declare that they have no conflict interests.

Ethical statement

This article complies with all ethical standards.

Funding

The research was carried out in frame of bilateral scholarship program between the Republic of Moldova and the Slovak Republic for 2019–2020 "Biopesticides for Pests' Management in Organic Farming" supported by grant of the Ministry of Education, Science, Research and Sport of the Slovak Republic.

Acknowledgements

The authors Elisovetcaia Dina, Ph.D. and Ivanova Raisa, Ph.D. are grateful to the Ministry of Education, Science, Research and Sport of the Slovak Republic for supporting researcher's mobility within the bilateral scholarship programs for 2019–2020.

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