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PLANT GROWTH-PROMOTING TRAITS OF ANTARCTIC ENDOPHYTIC BACTERIA

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Successful colonization of Antarctic lands by vascular plants *Deschampsia antarctica* and *Colobanthus quitensis* and their adaptation to stressful environments is associated not only with climate change but also with the functioning of microbial groups of phylo- and endosphere of these plants.

The aim of our study was to screen plant growth-promoting traits in endophytic bacteria of antarctic vascular plants.

Materials and methods. We have studied 8 bacterial cultures isolated from *D. antarctica* collected during the 25th Ukrainian Antarctic Expedition (January-April 2020) along the Western part of the Antarctic Peninsula.

Overnight liquid cultures were obtained on Nutrient Broth medium (HiMedia, Ltd.) in a shaking incubator (26 °C, 160 rpm). Bacterial isolates were grown on Ashby's combined-nitrogen-free medium with sucrose. Drop collapse assay for cyclic lipopeptide production (CLP), motility assay, exoprotease production and phosphate solubilizing ability were performed using generally accepted methods.

Results. All studied isolates have shown plant growth-promoting traits. The most abundant were nitrogen-fixing activity and motility. Both these play important role in plant colonization and promoting the growth of plants in harsh environments. The evidences of CLP were shown by two strains only. There was no notice of phosphate solubilizing ability and exoprotease production.

Conclusions. Endophytic bacteria of antarctic vascular plants could support the growth and nutrition needs of the plants.

Key words: PGPB, antarctic vascular plants, endophytes.

In both managed and natural ecosystems, beneficial plant-associated bacteria play a key role in supporting and/or increasing plant health and growth. Plant growth-promoting bacteria (PGPB) seem to function as a «normoflora» of a plant, can be applied in agricultural production or for the phytoremediation of pollutants [1].

Plant growth-promoting bacteria facilitate plant growth in two ways, either by direct stimulation or by biocontrol (i.e., suppressive activity against soil-borne diseases). The direct stimulation of plant growth may be a consequence of nitrogen fixation, phosphate solubilization, iron sequestration, synthesis of phytohormones (such as auxins, cytokinins, and gibberellins), or modulation of plant ethylene levels [2] helping plants to overcome stress and support cell metabolism. In fact, no single organism has the ability to make use of all the available mechanisms that could be used to promote plant growth. Various PGPB often possess one or more of the above mentioned traits [3].

Successful colonization of Antarctic lands by vascular plants *Deschampsia antarctica* and *Colobanthus quitensis* and their adaptation to stressful environments is associated not only with climate change but also with the functioning of microbial groups of phylo- and endosphere of these plants [4].

The aim of our study was to screen plant growth-promoting traits in endophytic bacteria of antarctic vascular plants.

Materials and Methods

We have studied 8 bacterial cultures isolated from *D. antarctica* collected during the 25th Ukrainian Antarctic Expedition (January-April 2020) along the Western part of the Antarctic Peninsula. Overnight liquid cultures were obtained on Nutrient Broth medium (HiMedia, Ltd.) in a shaking incubator (26 °C, 160 rpm). Nitrogen-fixing activity were checked on Ashby's combined-nitrogen-free medium with sucrose. Bacterial growth was determined by the change of optical density (OD_{600}) and evaluated as + (weak growth), ++ (moderate growth), +++ (abundant growth) [5].

Drop collapse assay for cyclic lipopeptides production (CLPs) was performed onto Parafilm. The reduction of the surface tension and the collapse of the droplet (10 µL aliquots of bacterial overnight culture) indicated the presence of surfactants [6].

Motility assay was performed onto 1/5 Nutrient agar (0.3%). 10 µL aliquots of bacterial overnight culture were spot in medium surface. Colony diameter was measured in 24, 48 and 72 h after inoculation on 0.3% 1/5 NA [7].

Exoprotease production was tested using skim milk agar [8]. A cleared zone surrounding bacterial growth after incubation for 48 and 72 h at 28°C was the evidence of exoprotease production.

Phosphate solubilizing ability was tested on Pikovskaya (PVK) medium [9] incorporated with $Ca_3(PO_4)_2$.

All experiments were performed in triplicates.

Table. **Plant growth-promoting traits of studied isolates***

Isolate No.	CLPs	N_2-fixing activity**	Motility
10.4	+	+++	–
15.6	–	+	+
24.4	+	+++	+
25.2	+	+++	+
26.2	+	+++	+
26.7	+	++	–
39.12	+	–	+
40.1	+	–	–

* Phosphate solubilizing ability and exoprotease production are no shown because it was not detected;

** + weak growth, ++ moderate growth, +++ abundant growth in Ashby's medium.

Results and Discussion

The mentioned set of screened plant growth-promoting characteristics was chosen based on the importance of adequate nutrition in low-temperature environment and defense system against numerous of pathogens [10].

All studied isolates have shown plant growth-promoting traits (Table).

The most abundant were nitrogen-fixing activity, cyclic lipopeptides production and motility. These traits play important role in plant colonization and promoting the growth of plants in harsh environments. Nitrogen is known as one of the limiting factors regarding plant growth [11]. Biological nitrogen fixation plays a great role in subsidizing plants with nitrogen in such limiting or low-mobility environments as Antarctic region. Phosphorus is one of the six elements essential for plant growth. The majority of phosphate solubilizing bacteria affiliates with *Paenibacillus, Bacillus, Pseudomonas, Lactococcus, Enterobacter* and *Alcaligenes* [12]. Although there were *Bacillus* and *Pseudomonas* among studied isolates there was no evidences of phosphate solubilizing ability as well as exoproteases synthesis.

The evidences of CLPs were shown by almost all isolates. Despite the fact that CLPs are known as biocontrol molecules, their role is believed more complicated than this [13]. CLPs exhibit interesting biological activities including interactions with biofilms [14] which affect not pathogens only but could manage colonization activity and the balance among endophytic community itself.

Conclusions

Antarctic endophytic bacteria seem affect plants directly through nutrition facilitating and various colonization mechanisms which needed to be studied deeper.

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