

Ice nucleation bacteria- a challenge for agriculture

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ABSTRACT

Ice nucleating bacteria can harm plants by stimulating the formation of ice in their tissues. On the other hand they contribute to cloud formation and rain precipitation on earth. It looks like they can play a role to local weather formation. In the same time the ice nucleating bacteria, has proteins which can bind ice crystal and contribute to the crystals formation. They can be dangerous during late frosts in the spring in orchards, affecting the blossoms, and producing damages and crop losses. They can be used in different purposes, in order to obtain ice in different conditions. The damage in orchards is highlighted. Some bacterial products were issued in order to fight with this kind of frost and to protect plants.

Keywords—ice nucleating bacteria, plant protection, *Pseudomonas*, *INA* genes

I. INTRODUCTION

The bacteria stimulating ice formation are known in agriculture and in food industry and environmental microbiology too. For example, some bacteria from milk, can determinate an abnormal freezing of this product. Strains of *Pseudomonas fluorescens* were found responsible for it. The temperature for ice nucleation in milk, was of -1, 7°C, for the culture of bacteria, and -1, 4°C for a bunch of bacterial concentrated cells [1]. In the same time, in dilution without any cell, did not appear ice crystals. The bio aerosols are [2] pollen, fungal spores, bacteria, seeds and other bio particles, which serve to dissemination of organisms, and can serve as nucleation centers for clouds droplets, ice crystals and precipitation. Distribution in air and in rain water of such microorganisms, show a percent of 2.2 to 19, 4% in rain water and 1, 8-9, 5 % in the air samples [3]. They are able to speed up the ice formation at higher temperature [4], many of them, strains from *Pseudomonas* genus (*P. syringe*, *P. poae*, *P. fluorescens*, *P. graminis*, and *P. viridiflava*.)

II. POTENTIAL OF ICE NUCLEATING BACTERIA

A. Generalities

Some bacteria, can influence the climate by determination of precipitations and even participation in regulate weather at secondary, marginal level [5]. Bacteria are found in atmosphere, as bio-aerosols, and they are very diverse *Proteobacteria*, *Actinobacteria*, *Firmicutes*, *Acidobacteria*, *Bacteroidetes*, *Firmicutes* phyla [4]. Some bacteria are active ice nucleators [6], in special *Pseudomonas* strains bearing *INA* genes, and even some fragments of bacterial cells in quantity of 199-482/mL, and about 12% of bacteria bear the *INA* genes, and excreted membrane vesicles. In special *Pseudomonas* strains have this capacity of catalyze ice formation and have impact on precipitations formation? An older work, showed that plants pathogenic bacteria, like *Xanthomonas* and *Pseudomonas*, can participate to the condense formation in clouds [7]. The mechanism of ice nucleation is the hydrogen bonding at the contact point water –bacteria, imposing a structural order in the molecules network [8]. In fact, *Pseudomonas syringe* life cycle is linked with the water cycles in nature [9]. Ice nucleating activity has the strain *Pseudomonas antarctica* IN- 74, isolated from Ross Island in Antarctica [10]. The experiments shown that the ice nucleation activity remains even after cell death [11]. The responsible for the effect are special proteins, ice nucleation protein, determined

by *INA* genes, which allow ice crystals formation at temperature near melting point [12]. The protein, located in outer membrane, is monomer having about 118 amino acids and a GM of 118kDal in three domains [13] can control the ice crystals formation, and the heterogeneous water solution containing the nuclei for crystals formation, freeze at temperature higher than normal freezing temperature, at about -2 C, producing in function of freezing rate, damage of plants cells [14]. Even in snow were isolated bacteria with variable ice nucleation activity [15]. The ice nucleation activity decrease in presence of UV rays, NO₂ and ozone [16].

B. Damages in cultures

The same activity and phenomenon, happens in plants which determinate the plants frost damage in spring. First time this was reported in England [17]. The spring frost occurs and coincides with the presence of *Pseudomonas* strains in trees phyllosphere (*P. syringae*, *P. agglomerans* and *P. fluorescens*). This kind of bacteria was discovered even in subtropical area most of them being gram negative from *Pseudomonas* and *Sphingomonas* genera [18]. In wheat cultures, of various types, *Pseudomonas syringae* are the most frequent INA bacteria, and its can be propagated with seeds [19]. This was associated with diseases and destruction by late frost of stone fruits from orchards for the first time in Romania [20]. Infected flowers became brown and faded destroyed by the frost and ice [21], producing in the same time, the so called apoplexy of stone fruit trees, determining damages for farmers.

C. Possible applications

Some products were issued in Europe to fight the ice formation in plants tissues and limitation of the bacteria, in order to limits the phenomenon, and main were prevention with bacteria strains which do not nucleate ice, in order to avoid perish and destruction of flowers (Frostban (*P. syringae* 742 RS) Frostban B (*P. fluorescens* A506) and others. In Belgium, some researches [22] studied the pathogenity and aggressiveness of *Pseudomonas syringae* populations in orchards and concluded that there are no chemicals that prevent the evolution of bacterial diseases, which require finding alternative methods to prevent infection by using resistant varieties and biological control methods. Other researches focus on obtaining non-ice nucleating mutants of *Erwinia herbicola* and tested it on cultivated sunflower seedlings to avoid ice formation [23]. The same was obtained with *P. syringae*. Some authors [13] consider that's not good, because the wild strain can assure the formation of rain, an important element for the crops too. In the same time, some authors thing that the snow additive with bacterial content can be dangerous because they are phytopathogens, can change the species composition in mountains area [24]. The ice nucleating bacteria can be inhibited by use of copper sulphate [25]. Applications in plant protection, killing parasitic nematods using Snowmax –INA agent was designed [26]. Snowmax was created to supplement snow for skiing in ski resorts. The ice nucleating bacteria can be usefor destroying pest during winter [27].

The use of ice nucleation bacteria in food industry as texturing freezing food and in seeding the clouds for rain, are some possible applications [28]. Use of this kind of bacteria for rain making is still in study, being involved in so called bio-precipitation [13]. The ice nucleating bacteria, like bacteria from the genera *Pseudomonas*, *Xanthomonas* and *Erwinia*, can reduce super-cooling, freezing time and costs [29] and are in generally safe for humans having only a low risk [30].

III. CONCLUSIONS

The ice nucleation active bacteria, can have –together with other active particles in the atmosphere and influence on local weather, on precipitations, but can also contribute to the damage of cultures in special in early spring. Some characteristics of these microorganisms can be used to improve weather-were needed- to protect plants against pest and pathogens, or to contribute to preserve food. In order to protect the cultures from frost, and ice formation, we must find new tools, and the answer can be new researches to identify the best measure to be taken for preservation of cultures. There are still possible applications of this kind of bacteria that we can discovered in the near future.

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