Bacterial microflora isolated from the bark surface of poplars growing in areas where air pollution is very high

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ABSTRACT

In the autumn of 1976 bacteria of the genera Bacillus, Pseudomonas, Flavobacterium, Erwinia and Cellulomonas were isolated from the bark surface of poplars growing in protective belts around several industrial plants. It was found that the qualitative and quantitative composition of the surface bacterial microflora changes in dependence on the degree of resistance of the poplars to the action of the dust emitted by the industrial establishment and containing high amounts of heavy metals.

Two international conferences have been devoted to problems of the ecology of microorganisms on the surface of plants (Preenece and Dickinson, 1971; Dickinson and Preenece, 1976).

At the last of these conferences organized in 1975 by the University of Leeds (England), Smith (1976) discussed the results of investigations on the influence of air pollution on the structure and function of microbiological ecosystems developing on the surface of plants. Surface microorganisms are capable of absorbing inorganic substances, and in this way they exert an influence on the metabolic processes of their host plant. The development of diseases in plants is particularly dependent on the influence of industrial pollution on pathogens and the saprophytic microflora.

For investigating the surface bacterial flora poplars were chosen, because, under conditions of industrial pollution, they grow much more vigorously than do other trees and shrubs. Some selected poplar varieties tolerate a high SO₂ concentration and dust containing large amounts of heavy metals.

The aim of the study was to gain a better knowledge of the bacterial microflora isolated from the surface of poplar bark from healthy trees

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and those with symptoms of fungal diseases, and more or less resistant to the action of dust containing high concentrations of heavy metals.

MATERIAL and METHODS

Bacteria were isolated in the autumn of 1976 from the bark surface of poplars growing in protective belts around several industrial plants. A list of the emission sources, and the approximate composition of the emitted gases and dusts as well as the poplar varieties from which the bacteria were isolated is given in Table 1. In the last column of this table information is included on the occurrence on these poplars of symptoms of fungal leaf and bark diseases.

Isolation and classification of the isolated bacteria

Fragments of bark were collected from all poplar varieties mentioned in Table 1, with careful maintenance of sterile conditions, and always from the side of the stem exposed to the source of emission. Bark was collected at two heights (1 and 2 m). On account of the varying age of the trees (4—10 years) the bark structure differed widely. The next day after sampling the pieces of bark were divided in the laboratory into smaller fragments and placed as inoculum on Petri dishes with YPG medium, pH 7.0—7.2 and nutrient agar Difco, pH 6.9. For isolation of pure cultures the method of seeding on Petri dishes was applied, and then the morphological and biochemical properties of the isolated bacterial strains were investigated (Bergey, 1974).

RESULTS

On the basis of the results of studies by Białobok and Rachwał (1974) and the author’s own observations, the particular poplar varieties from which the surface bacterial microflora was isolated were classified to separate groups according to their higher or lower resistance to the local air pollution conditions (Table 2). This table also gives the number of bacterial strains isolated from the bark of the poplar varieties examined.

Bacterial strains isolated: genus Bacillus — bacteria of this genus were isolated from the bark surface of all the examined poplar varieties;

Genus Pseudomonas — microorganisms of this genus were not isolated from the surface of the resistant poplar P. ‘Marilandica’ growing in the neighbourhood of the metallurgic Works “Legnica” and “Lenin” or on the moderately sensitive P. × canescens susceptible to the polluting emission of the “Legnica” Works.

Genus Flavobacterium — the presence of these bacteria was noted on the bark of the resistant P. × candidans and the moderately sus-
Table 1

Characteristic of emission sources and poplar varieties from which bacteria were isolated

<table>
<thead>
<tr>
<th>Emission source</th>
<th>Approximate chemical composition of gases and dust emitted</th>
<th>Poplar varieties</th>
<th>Disease symptoms on poplars</th>
</tr>
</thead>
</table>
| 1. Zinc Works "Miasteczko Śląskie" in Miasteczko Śląskie | SO₂ dust with high Pb, Zn, Cu, Fe, As, Mg, Al, Si, Sn, Ti, Cr, Ca, Ni, Mn, Mo | P. ‘Marilandica’  
P. ‘Hybrida 194’  
(P’NE 49’) | no disease symptoms |
| 2. Zinc Works "Wełnowiec" in Katowice-Wełnowiec | as above | P. × candidans  
P. ‘Marilandica’ | some trees with symptoms of fungal diseases |
| 3. Zinc Works "Orzel Bialy" in Brzeziny Śląskie | as above | P. alba  
P. × candidans | no disease symptoms |
| 4. Zinc Works “Szopienice” in Katowice-Szopienice | as above | P. × berolinensis  
P. × candidans | no disease symptoms |
| 5. Copper Works in Legnica | as above | P. × berolinensis  
P. × candidans  
P. × canescens | shoots damaged by Valsia cordida  
no disease symptoms  
no disease symptoms  
leaves damaged by Venturia populina  
no disease symptoms |
| 6. Iron and Steel Works “Lenin” in Nowa Huta | SO₂ and dust | P. × candidans  
P. nigra ‘Fastigiata’ | shoots damaged by Cryptodiaporthe populnea  
no disease symptoms  
shoots damaged by Cryptodiaporthe populnea |
| 7. “Czarna Huta” near Tarnowskie Góry | Large quantities of Cl, H₂S, nitrogen oxides and a number of organic substances | P. ‘Gertica’  
P. sp. | no disease symptoms |

ceptible P. ‘Marilandica’ growing in the environs of the Zinc Works “Wełnowiec” and on two moderately susceptible poplar varieties and two varieties sensitive to dust and gas emitted by the Copper Works in Legnica.
Table 2
Number of bacteria isolated in dependence on the degree of resistance of the poplars to industrial pollution from various sources

<table>
<thead>
<tr>
<th>Emission source</th>
<th>Poplar varieties</th>
<th>Degree of resistance</th>
<th>Genera of bacteria</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Bacillus</td>
</tr>
<tr>
<td>Zinc Works</td>
<td><em>P. 'Marilandica'</em></td>
<td>resistant</td>
<td>1</td>
</tr>
<tr>
<td>&quot;Miasteczko Śląskie&quot;</td>
<td><em>P. 'Hybrida 194'</em></td>
<td>resistant</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>(P. 'NE 49')</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Zinc Works</td>
<td><em>P. × candidans</em></td>
<td>resistant</td>
<td>1</td>
</tr>
<tr>
<td>&quot;Wełnowiec&quot;</td>
<td><em>P. 'Marilandica'</em></td>
<td>moderately resistant</td>
<td>3</td>
</tr>
<tr>
<td>Zinc Works</td>
<td><em>P. alba</em></td>
<td>resistant</td>
<td>2</td>
</tr>
<tr>
<td>&quot;Orzel Bialy&quot;</td>
<td><em>P. × candidans</em></td>
<td>resistent</td>
<td>1</td>
</tr>
<tr>
<td>Zinc Works</td>
<td><em>P. × candidans</em></td>
<td>resistent</td>
<td>1</td>
</tr>
<tr>
<td>&quot;Szopienice&quot;</td>
<td><em>P. × berolinensis</em></td>
<td>resistent</td>
<td>1</td>
</tr>
<tr>
<td>Cooper Works in Legnica</td>
<td><em>P. × berolinensis</em></td>
<td>moderately resistant</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td><em>P. × candidans</em></td>
<td>resistent</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>P. × canescens</em></td>
<td>moderately resistant</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td><em>P. 'Hybrida 275'</em></td>
<td>(P. 'NE 42')</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>P. 'Marilandica'</em></td>
<td>resistant</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>P. nigra 'Fastigiata'</em></td>
<td>susceptible</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td><em>P. 'Robusta'</em></td>
<td>moderately resistant</td>
<td>3</td>
</tr>
<tr>
<td>Iron and Steel Works &quot;Lenin&quot;</td>
<td><em>P. × candidans</em></td>
<td>not assayed</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>P. 'Marilandica'</em></td>
<td>resistant</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>P. nigra 'Fastigiata'</em></td>
<td>resistant</td>
<td>2</td>
</tr>
<tr>
<td>&quot;Czarna Huta&quot; near Tarnowskie Góry</td>
<td><em>P. 'Gerlica'</em></td>
<td>not assayed</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>P. sp.</em></td>
<td>not assayed</td>
<td>1</td>
</tr>
</tbody>
</table>
Genus *Erwinia* — bacteria of this genus were isolated from the bark surface of the resistant varieties *P. 'Marilandica'* and *P. nigra 'Fastigiata'* exposed to the pollution emitted by the Lenin Ironworks and from the moderately susceptible varieties *P. × berolinensis* and *P. nigra 'Fastigiata'* growing close to the "Legnica" Works.

Genus *Cellulomonas* — these microorganisms were obtained from *P. alba* growing on the cemetery in Brzeziny Śląskie in the neighbourhood of the Zinc Works "Orzel Bialy" and from the bark surface of *P. 'Gerlica'* exposed to the emission of the chemical plant "Czarna Huta" near Tarnowskie Góry.

**DISCUSSION**

The results obtained confirmed the supposition that the surface bacterial microflora developing on poplar bark may serve as index of the greater or smaller resistance to gases and dusts emitted by industrial plants. It results from Table 2 that the bacterial flora was more differentiated on the bark of varieties considered as moderately resistant, notwithstanding the kind of pollution source. From the bark of moderately susceptible and sensitive varieties such as: *P. × berolinensis, P. × canescens, P. 'Hybrida 275', (P. 'NE 42'), P. nigra 'Fastigiata' and P. 'Robusta' which grow in the protective belt of the Copper Works in Legnica, a greater number of bacterial species was collected than from the bark of *P. 'Marilandica'* which is a decidedly resistant variety within the same range. There is a marked correlation between resistance of the tested poplar varieties to industrial pollution, and the occurrence of bacterial microflora on their bark. This is particularly true for varieties of known origin which grow in experiments started by the Institute of Dendrology in Kórnik in the protective belt of the Copper Works in Legnica.

It would seem, on the basis of the results obtained, that the variety *P. nigra 'Fastigiata'* will be susceptible when exposed to the conditions prevailing around the Lenin Iron Works, and not, as it was believed up till now, resistant. There also is a relation between the higher or lower resistance of poplar varieties to industrial pollution, and injury of the shoots and branches caused by fungal pathogens. The varieties *P. nigra 'Fastigiata' and P. 'Robusta'*, under the conditions prevailing around the Copper Works in Legnica, belong to trees moderately susceptible to the emitted dusts and gases. The differences in the bacterial flora occurring above all in susceptible plants, are very wide in the above mentioned varieties. Symptoms characteristic for the fungus *Cryptodiaporthe populea* causing one of the most dangerous and common poplar bark diseases were found on these trees. On the other
hand, in the moderately susceptible variety P. × berolinensis necrosis
disease caused by Valsa sordida was observed. These diseases probably
infect the species and varieties of poplars weakened by other factors,
in the present case by the poisonous compounds in the pollution.

On the basis of these preliminary results, bacteria were not selected
which would serve as index of pollution of the zone close to the emis-
sion sources. Balicka, Węgrzyn and Varanka (1975) assumed
as such test strains bacteria of the genus Pseudomonas and yeast which
occurred on the leaves of alfalfa and sugar beets growing within the
pollution range of the Copper Works in Głogów.

It could be concluded from the description of these authors that
these bacterial colonies belonged to the fluorescent group of the genus
Pseudomonas to which species pathogenic to plants also belong.

In order to complete the present investigations, it is indispensable
to isolate once more the bacterial and fungal microflora from the bark
surface of the same varieties of known origin and to test the resistance
reaction of the fungal microflora to industrial pollution under labor-
atory conditions.

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Mikroflora bakteryjna izolowana z powierzchni kory topoli rosnących na terenach nadmiernie skażonych zanieczyszczeniem przemysłowym powietrza

Streszczenie

Jesienią 1976 roku z powierzchni kory topoli rosnących w strefach ochronnych kilku zakładów przemysłowych wyizolowano bakterie z rodzaju Bacillus, Pseudomonas, Flavobacterium, Erwinia i Cellulomonas. Stwierdzono, że skład jakościowy i ilościowy powierzchniowej mikroflory bakteryjnej zmienia się w zależności od stopnia odporności topoli na działanie emitowanych przez zakłady przemysłowe pyłów zawierających duże ilości metali ciężkich.