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A Study of the Microorganisms in the Air of Leningrad

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The air of large cities and industrial centers, as investigations have shown, contains a significant amount of solid and gaseous admixtures and microbes; according to the authors the latter enter the air chiefly from the soil.

In our work, which is here described, we set as our goal the determination of the number and character of the microorganisms in the air of Leningrad, in its different districts: in the dustier districts near factories and train stations and in the least dusty districts in a zone of vegetation, and for comparison, within the installations. The work was done in 1938.

Regarding the methods of studying the microorganisms of the air, it should be stated that their selection is just as difficult as is the determination of the atmospheric dust, because there is absolutely no exact method for determining their actual amount in a unit volume of air. We used the following three methods of investigation in our work: a) Koch's precipitation method b) D'yakonov's method, in which one studies the quantity of microbes in a definite volume of air that is drawn through a cylinder containing a liquid absorbent and beads; c) investigation of the microflora in freshly fallen snow.

The investigation by the first method is conducted as follows: Sterile Petri dishes of identical size (with a surface area of approx. 100 square centimeters) and containing agar on broth are set out in the survey area for 5 minutes; then they are closed and kept in an incubator at 37°C for a period of 48 hours, after which there is a calculation of the colonies that gives a representation of the amount of microorganisms that have settled on a surface of 100 square centimeters (cm<sup>2</sup>) with an exposure of 5 minutes.

The D'yakonov method was used in the following form: we poured 100 cm<sup>3</sup> of a physiological solution into, and placed several layers of glass beads on the bottom of a Drexel cylinder, through the top of which runs a short and a long tube. The glassware, thus prepared, together with the absorbent and the beads, was sterilized by steam under pressure and transported to the place of the investigation where it was then utilized.

The air was drawn in with a hand-operated piston pump, which was connected to the short tube of the Drexel cylinder so that a suction of air occurred through the cylinder's long tube, which reaches almost to its bottom. With this, after every stroke of the piston and the passage of the air through the physiological solution the cylinder was shaken so as to disintegrate the dust particles, by the action of the beads, and to liberate their accumulations of microorganisms.

Many authors note that in dust particles there exist microbic accumulations each of which will produce one colony in a culture and will be considered as one microbic cell; the crushing of the dust particles leads to their separation and to the release of the microbes and, consequently to an increase in the number of colonies growing after culture. This is the basis for the use of the beads in the D'yakonov method. After drawing 20 liters (l.) of air through the Drexel cylinder the absorbent was cultured on a series of dishes containing meat-peptone agar, whereupon the cylinder was shaken again prior to the culture. The dishes, as in the first case, were kept in an incubator 48 hours at 37°C, after which we calculated the quantity of microbes for 1 cubic meter (m<sup>3</sup>) of air. We used both methods at the same point simultaneously, during which the exposure of the open Petri dishes and the aspiration of the air were made at the level of the respiration zone.

For the investigation of the snow we collected it during a snow fall (at all points simultaneously) into sterile glassware from which we cultured the melted water onto agar and kept it in an incubator as described above; after counting the colonies we computed their number for 1 cm<sup>3</sup> of melted water.

Concerning the selection of a place for an investigation, we marked the points both with the greatest amount of solid atmospheric contaminants (at the points of the most smoke and street traffic) and with the least contamination - in a vegetational zone.

These points are as follows: 1) near an industrial plant; 2) near a second industrial plant; 3) near the Moscow station; 4) near a factory; 5) the city center - Lassal Square; 6) a garden-the yard at the hospital im. (in name of) Erisman; 7) Sosnovka; 8) Central Park of Culture and Rest on the Kirovskiy Islands; 9) and for comparison - within an installation - in a laboratory where several people work. The snow was collected at only 5 points.

As a result of the study of the microflora of freshly fallen snow we received the following data: (see table)

From the presented figures it is evident that the greatest quantity of microbes was detected near the plants and the train station, where there is a great amount of both vehicular and pedestrian traffic and where, according to our investigations, the largest amount of dust was found. The least amount of microbes was shown on the Kirovskiy Islands, in the Central Park of Culture and Rest. Atmospheric precipitations, snow in particular, absorb the dust particles suspended in the air and the microorganisms implanted on them. It is possible to make a comparative evaluation of the contamination of the air at various points of the city

by the number of these microorganisms.

We have a very scanty material for comparison of our findings with the results of snow investigations by other authors, because such works are too few. Shelk detected 4-6 bacteria per  $\text{cm}^3$  in glacial snow collected at an altitude of 2,000 meters; Yanovskiy, in investigating the snow in Kiev in 1888, found from 2 to 463 colonies per  $\text{cm}^3$ ; Gorovits - Vlasova, in 1926-1927 in Dnepropetrovsk, detected from 120 to 260 colonies per  $\text{cm}^3$  of freshly fallen snow, and from 155 to 2,690 colonies in snow that had lain for 2-3 days. By comparing our results with the cited data we come to the conclusion that the number of microorganisms in the air of Leningrad is greater than the number that was in the air of Kiev in 1888 and in Dnepropetrovsk in 1927, which can be completely explained both by the more developed street traffic and by the greater number of population as well as by the significantly increased number of factories.

In regards to the microbes in the snow, we attempted to determine the relationship between the quantity of microbes and the temperature of the air; with this, it proved that with collections of snow made at the same place at temperatures of  $-10^\circ\text{C}$ ,  $-4^\circ\text{C}$ , and  $-2^\circ\text{C}$  there was no decrease in their number at the lower temperatures. Concerning the species of microbic flora in the snow, we found the following: 1) pigmentary and non-pigmentary cocci and sarcinae; 2) sporogenic saprophytic bacilli (B. subtilis, B. mesentericus, B. megaterium, B. mycoides); 3) molds (Aspergillus niger and Mucor mucedo).

With the use of the precipitation and air-aspiration methods in the different points of the city, we established that the amount of microbes increases from the winter to the spring months, with the maximal number in May, which completely confirms the connection of the bacterial flora of the air to the dust (soil).

In addition, the number of atmospheric precipitations in the period preceding the days of the investigation and the velocity of the wind undoubtedly exerted an influence; thus, in the first half of May, when the majority of May's investigations were made, there was a comparatively small number of precipitations [6 mm (?see text) for the first ten days] and the wind on the days of the investigation reached 7 -6 -4 m. At the end of May (no investigations) and in June the number of precipitations increased significantly; the investigations in June were conducted with a comparatively moist soil and a higher relative humidity, and the number of microorganisms decreased in June at the majority of the points: a decrease of colonies, as compared with May, was also observed in September. Unfortunately, we conducted no investigations in July and August. It is necessary to indicate that in the process of the investigation we observed an increase in the number of microbes at the same point with an increase of traffic; in September with open Petri dishes were conducted near a factory on holidays, when a large crowd of people were on the street, and this produced a sharp increase in the colonies. It is interesting to note that microbes in the air also precipitate with fog droplets: in our investigations in the spring months, on the Kirovskiy islands, we sometimes observed a low fog, whereupon its droplets precipitated onto the agar; in those areas of the dish we observed a solid growth of microbes and an

increased number of them as compared with the clear days.

For a comparison of the quantity of microbes in the atmospheric air and in an inner setting, we conducted parallel investigations in a work room - a laboratory where several persons were employed; the figures resulting from the investigation by the precipitation method were smaller in the majority of cases than in the atmospheric air, with the exception of some experiments in parks. Such results seem to contradict the established fact that there are more microbes in the air of installations than in the air outside; and, actually, in the determination of the bacterial flora by means of air aspiration, we always found more microbes/m<sup>3</sup> in the laboratory than in the air outside; their decreased number in the precipitation method is related to the fact that in a room there is evidently a finer dust and its precipitation is delayed; as a result of this, more dust, and the microbes contained with it, will settle in 5 minutes on the same surface area outside than in an installation.

Concerning the quantity of colonies received with the D'yakonov method, it should be noted that here, too, there is an increase noted in their number in the spring months; there is no strict parallelism between the amount of the precipitating microbes and the microbes that are in a unit volume of air, and this can hardly be expected, considering that in an investigation by the D'yakonov method the dust particles are crushed and the microbic accumulations are broken up, whereas only the larger dust particles with their contained microbes will be able to precipitate onto the open dishes during the comparatively short period of time. For this very reason we can never adhere to the opinion held by Omelyanskiy, who considers that approx. as many microbes as there are contained in 10 l. of air will precipitate on a dish's surface area of 100 cm<sup>2</sup> in five minutes; our findings conflict sharply with this opinion.

Transferring to an evaluation of the methods that we used, let us point out the following: the investigation of the bacterial flora in freshly fallen snow can serve as a relative method for a comparative evaluation of the degree of air contamination at different points of a populated area, with a simultaneous collection of samples. Concerning the precipitation method, it entices with its simplicity, but gives doubly approximate figures, which are dependent, evidently, on the type and sizes of the dust particles and air currents that are formed in the air; using the precipitation method with two identical dishes at the same time, at the same point, at the same level and situated 35-40 cm from each other, we never observed the same number of colonies: the difference in the number of colonies was sufficiently significant.

The method of determining the microbes in a unit volume of air by its aspiration should be considered the most precise. It gives no large fluctuations in the quantity of microbes in a simultaneous investigation, with the use of similar equipment and at the same rate of aspiration.

The D'yakonov accessory for breaking up the dust particles and accumulations of microbes is fully expedient.

### Conclusions

1. The number of microorganisms in the outside air, as well as the amount of solid and gaseous admixtures in it, can serve as an indicator of the degree of contamination at different points of a populated area.

2. In view of the fact that the chief source for the contamination of the air with microbes is the soil, the number of microbes in the air in the same populated area depends on the number of atmospheric precipitations, the degree of moisture in the soil, the growth of street traffic, and the velocity and direction of the wind.

3. The greatest number of microorganisms/ $m^3$  of Leningrad's air (55-64 thousand) was detected during the most significant wind and at the points of the most street traffic near the factories and the train station; in a vegetational zone the air contains a significantly smaller number of microbes (10-35 thousand).

4. A determination of the number of microorganisms in the air of an installation, which was conducted in parallel with the investigation of the atmospheric air, showed that there are many more microorganisms/ $m^3$  than there are outside; there are fewer precipitated onto Petri dishes in 5 minutes in a room than occurs outside, which can be explained by the fact that the room's dust is finer, settles slower and carries with it fewer microbes during a short interval of time.

5. The air is contaminated by the soil microbes that are most resistant to drying and ultraviolet radiation; we detected micrococci and sarcinae, saprophytic sporogenic bacilli of mold and yeast.

6. The determination of the air's microflora by the precipitation method is extremely simple, but it gives only approximate data and can be used for an investigation in different points only with the condition that there is a simultaneous analysis and a homogeneity of the dust.

7. The method of determining the microbes in aspired air should be recognized as the more precise, particularly the supplement proposed by D'yakonov with which there is a crushing of dust particles and a breakdown of the microbial accumulations.

8. Concerning the method for the study of the microorganisms by the investigation of snow, it can give comparative indicators of the degree of contamination at different points, with the condition that there is a simultaneous collection of freshly fallen snow.

### Literature

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The number of microorganisms/cm<sup>3</sup> of melted snow water.

Place where snow was collected	In February			In March		
	Max.	Min.	Average of all tests.	Max.	Min.	Average of all tests
1. Near a factory	860	80	460	1200	120	506.6
2. Near 2nd factory	-	-	-	1000	30	443.3
3. Near the Moscow train station	800	100	450	600	80	379.0
4. Yard at Erisman Hosp.	500	20	260	600	18	109.0
5. Central Park	160	10	50	200	9	43.9