THE EFFECTS OF SUNLIGHT ON BACTERIA.

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INTRODUCTION.

1. Bacteria

Definition

Classification

II. Bacteria

(a) Fungi without power of photosynthesis.

(b) Bacteria dependent upon, excepting in nitrifying groups, organic matter for food.

(c) Light of no value to bacteria.

(d) Thru degeneration have lost power of photosynthesis and power to withstand light.

EXPERIMENTS WITH LIGHT AND BACTERIA.

Exp. A.

Exp. B.

Exp. C.

Exp. D.

Exp. E.

Exp. F.

CONCLUSIONS.

I. Direct Sunlight

II. Different rays of spectrum.

III. Presence of oxygen.

IV. Sunlight: Tendency to keep growth of bacteria in water in check.

V. Bacteria at different depths of water.
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Bacteria are one celled microscopic plants which reproduce by fission and are propagated by sporulation. In the evolutionary classification of plants they hold the following positions.

Plant Kingdom

Sub Kingdom. Thallophyta

Division ll. Lethallophyta or true thallophytes.

Sub Division 1. Schyomycetes, fission fungi or bacteria.

As the above classification indicates the bacteria are supposed to be degenerate forms of fission plants, probably springing from the algae. Most of them being parasitic or saprophytic they have lost their power of photosynthesis, it in this degenerate form being no longer of use to them. Others of the nitrofying group derive their energy in an unknown way from certain mineral salts.

As these plants gradually formed the saprophytic habit they lost the need of light to assist in forming starch. Those that burrowed into the interior of the medium in which they were growing were more protected from the external world than those living upon the surface. The result is their descendants which of course also inherited the tendency to leave the surface were more successful in the struggle for existence than those living upon the surface or exterior,
This growth in the darkness however has a decided degenerating effect upon the bacteria. Their progenitors which lived in the light, providing that we except the theory that they sprung from the algae, had functions which prevented the light having any deteriorating effect upon them. As they formed the habit of growing in the darkness however this function of resisting the adverse effects of light was no longer needed, consequently it slowly wasted away. We therefore expect to find the bacteria unable to stand the presence of light, stronger than that to which they are accustomed.

Experiment A.

Test tubes containing one cc of a two day old bouillon culture of the bacillus subtilus were prepared; one was exposed to the direct rays of the sun on April 15th for two hours while the other was kept in darkness. At the end of the time of exposure tubes were inoculated, three from each of the above cultures and these set away to incubate. At the end of twenty four hours they were examined. Those tubes inoculated with the culture exposed to the light showed no growth; those not so exposed were slightly turbid.

Thirty six hours after inoculation the tubes from exposed culture which we will call No. 1 appeared as Figure 1 in Plate I. While those inoculated with the other culture which we will call No. 2 appeared as Figure II, Plate I. A white growth is noticed upon the surface of the medium in Figure 2, while Figure 1 shows no such growth.

In 48 hours tubes No. 1 appeared as Figure 3, Plate I there
still being no growth in it while tube No. 2 appeared as Figure 4 the growth being more abundant.

In 60 hours tubes No. 1 appeared as Figure 5, Plate I and tubes No. 2 appeared as Figure 6, Figure 5 still showing no growth. Observations were made of the tubes during a period of fourteen days in which no growth took place in any eg of the tubes of No. 1. Apparently all the bacillus in the culture exposed were destroyed by the light.

Experiment B.

Another unique method tried for testing the effects of sunlight as a germicide is illustrated in Figure 1, Plate II. In this experiment on the lower surface of a sterile pasteur was pasted the letter K made out of black paper. A plate culture was made in this out of fourteen cc of bouillon agar inoculated with one cc of a three day old bacillus bouillon culture.

After the agar in the plate had become firm or set, the plate was exposed to the sunlight for three hours the bottom of the plate being turned towards the sun so that the K would protect a portion of the culture from the sun's rays. This plate was then set away to incubate. At the end of twelve hours the part of the culture protected by the letter was well developed so that the K shown in bold relief, appearing as Figure 1, Plate III. In this case a few colonies developed in the agar unshielded from the sun. This was probably due to certain germs being protected by opaque foreign matter in the agar.

Experiment C.

This experiment was performed to test the sunlight's germicidal effect upon the water in the reservoir of the Manhattan
City Water Works.

On the 8th day of June, a day on which the sun shown brightly through out; the reservoir water was examined. In this examination plate cultures were made in the following manner: one cc of the reservoir water was mixed with 99 cc of distilled stiral water. An agar bouillon plate culture was made from this being innoculated with one cc of the mixture. This culture was made at the reservoir immediately after taking the water from it so that the number of germs in this water had no time to increase. The morning after the plate was made, at sun up, another plate was made at the same place and both plates set away to develop. At the end of two days the colonies were counted. The plate which was made in the evening had upon it twenty one colonies which of course was 1/100 of the number of the bacteria in one cc of the evening reservoir water. This would give 2100 bacteria present in the evening to one cc of water.

The plate made in the morning contained 3888 colonies which times one hundred shows that there were 388800 present in the morning. This experiment indicates that during the time of the absence of sunlight the bacteria increased from 2100 to 388800 per cc.

Figure 1, Plate II, shows a plate as it appeared from the evening test and Figure 2 as it appeared in the morning test, there being 21/38880 as many colonies present in Figure 1 as in Figure 2.
Experiment D.

The following experiment shows the number of bacteria present in the different depths of the reservoir water. The activity of sunlight in water at various depths is here illustrated. The following apparatus was used to obtain samples of water at the various depths in this experiment. A tube (a) Figure 1, Plate III is provided with a cork (b) which is held in place by the rubber band (c). To the wire loop (d) in the cork a string (n) is attached, by which the cork can be drawn out of the vial as far as the band will allow it to go. Upon releasing the string the rubber band forces the cork back into the vial.

This tube is fastened to a block of wood (e) which is grooved on one side to fit the tube and on the other to fit the bamboo pole (f) by means of the rubber bands g and g. Bamboo pole (f) is long enough so that so that samples of water may be taken as deep as fifteen feet if so desired. It is marked with a scale showing the feet and half feet, reading from the tubes upward. Wire loops, 1, 2, 3 etc keep the string (u) which is used to pull open the cork, in place. By means of this arrangement water at any desired depth may be obtained.

The water selected for testing was that of the Manhattan City Reservoir. This reservoir is open and broad enough so that the sun plays upon its surface from morning until evening on a clear day. Samples were taken at depths differing by two feet, from the surface down to a depth of six feet and plates made in the same way as in Experiment C. After four days
incubation the colonies were counted. The number of bacteria found present were as follows:

<table>
<thead>
<tr>
<th>Depth</th>
<th>Bacteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Surface</td>
<td>2100</td>
</tr>
<tr>
<td>2 feet deep</td>
<td>98100</td>
</tr>
<tr>
<td>4 feet deep</td>
<td>136800</td>
</tr>
<tr>
<td>6 feet deep</td>
<td>360000</td>
</tr>
</tbody>
</table>

By the above experiment it appears that the sun's rays influence the growth of bacteria in water to the depth of at least four feet in clear water.

**Experiment E.**

The following will show something of the nature of the action of the sunlight. One cc of a three day old culture of *bicusulis subtilis* were placed in each of the sterilized tubes. From one of these the air was expelled by means of hydrogen gas being introduced in it and then the tube was hermetically sealed with sealing wax. These tubes were then exposed to the direct rays of the sun for four hours.

At the end of this time two tubes of bouillon were inoculated from each of these cultures, two from the sealed and two from the unsealed. In twenty four hours the tubes were examined and it was found that those tubes inoculated with the culture exposed in the absence of air were turbid while those inoculated with the culture in presence of air showed no growth. This experiment indicates that the presence of oxygen or air is an important factor in the germicidal action of light. We show no drawings of Experiment E as the farther development is very much the same
as those shown in Plate I.

Experiment F.

Four test tubes each containing one cc of a three day old B Subtilus culture of bouillon were prepared. No. 1 culture was exposed to the direct sunlight for ten hours. No. 2 exposed to the blue, green and violet rays for the same period, the other rays being shut off by means of blue glass. However, a spectroscope examination of this glass showed that it transmitted a small part of the red rays. No. 3 was exposed to the red rays for the same time the other rays as the spectroscope showed being completely cut off by the red glass as the spectroscope showed. No. 4 was kept in the darknes.

At the end of the exposure tubes were inoculated from each of the above and then set away to develop. In twenty four hours tubes from No. 1 showed no growth; Tubes No. II no growth; Tubes No. III were turbid while in Tube No. IV was a thin membrane ( )

In 38 hours tubes No. I showed no growth, tubes No. II were slightly turbid, tubes No. III had upon the surface a membrane and tubes No. IV a heavy growth. The experiment from which this data was made was not entirely satisfactory.

From this the white light seems to be most active; the blue and the violet the next while the red is the least. (The data from the above experiment contain some error however owing to a lack of time to give to the experiment.

In conclusions of the foregoing experiments we would say the following may be deducted.

I. Sunlight has deteriorating effect upon the growth of
bacteria even acting as a germicide.

II. The direct rays of the sun are the most active, the blue and violet the next, and the red the least.

III. The presence of oxygen aids in the germicidal effects of sunlight.

IV. The sunlight tends to keep the bacteria in check in clear water thru which it shines.

V. The deeper the water is from the surface the less active will be the sun upon the bacteria.