THE ROLE OF MICROORGANISMS IN THE WEATHERING OF ROCKS

N. A. Krasil'nikov, Institute of Microbiology of the Academy of Sciences USSR, Moscow

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I. Microflora of the Surface Layer of Rocks

Although microbiology has amassed sufficient observations of the geological /318* activity of microorganisms, many scientists — not only among geologists, but also among pedologists — nevertheless take the viewpoint that the process of "rock weathering," the breaking down of rock, and its transformation into marl proceed under the effect of physicochemical and mechanical factors. It is the opinion of many pedologists that biogenic and, in particular, microbiological factors are excluded in this process (Kravkov [Ref. 4], Zakharov [Ref. 3], etc.).

Vernadskiy (Refs. 1, 2) wrote that to study the processes of rock "weathering" merely from the viewpoint of physiochemical phenomena reveals the backwardness of this field of chemical geology. The process of rock weathering, he wrote, is a biologically sluggish process. The approach to the study of this process must be biogeochemical -- in other words physical, chemical, and biological agents participate in the breakdown of rocks.

Polynov (Ref. 6) was the first to set about studying the initial stages of soil formation in detail from these viewpoints. He and his collaborators are concentrating their attention on lichens and studying their effect on the process of rock destruction (Polynov (Refs. 6, 7], Yarilova [Ref. 10]). These papers give graphic proof of the destructive effect of lichens.

Our own observations show that microorganisms — bacteria, fungi, actinomycetes, and algae — actively participate in the breakdown of rocks. In the preceding work (Ref. 5), we quoted data on the massive development of various microorganisms in the body of lichens and remarked that lichens are also accumulators of these creatures in the surface layer of rock.

It is well known to specialists that rocks are frequently overgrown with lichens (Fig. 1). These puzzling symbionts frequently cover the surface of mountain massifs in a continuous thick layer. As a rule, the rock beneath them is altered to a greater or less depth, and a surface layer is formed which is clearly colored (Fig. 2). This layer differs from the basic unchanged rock in a number of other chemical and physiological properties. It is less strong, may be easily worked, and more often than not is colored brownish or light yellow.

The thickness of this layer varies in different cases depending on the rock of the stone massifs, the conditions of the site, and the continuance of lichen growth on them. It fluctuates from 1 to 5 mm, but in isolated cases, goes as high as 10 mm.

The altered layer on the surface of the rocks is also often observed on massifs which are not overgrown with lichens. It also is customarily of differing thickness -- from 1 to 5 mm -- and has brownish coloration and reduced strength.

^{*}Numbers in the margin indicate pagination in the original foreign text.

This altered surface layer of rocks was the subject of our investigations. /319 We were interested in the presence of life in this impaired surface layer. Do or do not microorganisms exist there? For this purpose, we hammered off small pieces of rock having an altered surface layer.

The specimens collected were wrapped in sterile paper and taken to the laboratory, where they were subjected to microbiological analysis. In many cases the rock specimens were processed on the day they were collected in the Microbiology Department of the Academy of Sciences of the Armenian SSR. Most of the specimens, however, were sent to Moscow to the Institute of Microbiology of the Academy of Sciences USSR, where they were studied after a more or less protracted sojourn in a dry state (from 1.5 to 2 months from the time collected).

In the laboratory the surface of the collected specimens of rock monoliths was mechanically freed of lichens, then flushed and carefully washed in sterile water to cleanse it of surface microorganisms. After being dried in steel filings, the rock of the altered surface layer was scraped off coat by coat. The fine powder obtained was inoculated in a certain weight onto nutrient media, agarized and liquid.

In each specimen we investigated scrapings taken at 3 or 4 levels. The first, the topmost, was of 0-2 mm; the second was at 3-5 mm from the surface; the third was in a deeper part 6-8 mm (not always), and the last in a deeper part which did not change.

The largest number of specimens was gathered from the basaltic rocks of Armenia, then tufas -- red and black -- were investigated, and a few specimens of granite and limestone.

For the innoculations of the scrapings taken from the rocks we used nutrient media of varying compositions, i.e., meat-infusion agar, meat-infusion broth, grape must agar, Capek's synthetic medium, and Ashby's anitrogenous medium.

The test material was inoculated onto the agarized media in Petri dishes directly as powder. Less often the powder (scraping from the rock) was first diluted in water, and the aqueous suspension obtained was incoulated onto the media in the usual way. The cultures were incubated at 20-25° for 5-20 days.

About 50 specimens of basalts and tufas in all were examined in this way. The results of these investigations show that the surface layer of rocks covered with lichens is, when it seems changed to the eye, populated with microorganisms to a greater or less degree. We determined from 5000 to 100,000 germs per gram of rock, but their distribution in the altered layer is not uniform.

The greatest number of microorganisms was detected in the first level 0-2 mm from the surface. As depth increases, their number decreases, and they are not found at all beneath the altered layer in the untouched mass of the rock (Table I). Table I gives the average readings of these analyses.

The microbe population develops most abundantly, as these figures indicate, in the surface layer of the basaltic and granite masses, and less so in the tufa at all were detected on this medium, while the bacteria evinced no differences from the bacteria growing on meat-infusion agar.

TABLE II. GROUP COMPOSITION OF MICROORGANISMS IN SURFACE LAYER OF BASALTS AND TUFA. NUMBER OF CELLS PER GRAM OF ROCK

	Basalt			Tufa		
Nutrient Medium	Bac-	Actino-	ľ	Bac-	Actino	_
	teria	mycetes	Fungi	teria	mycete	Fungi
Meat-infusion agar Capek agar Ashby agar	4 500 10 000 150 000	50 200 500	20 200 100	200 6 500 10 000	10 50 50	0—10 10 0—10
Grape must agar	-	_	1 000		_	20

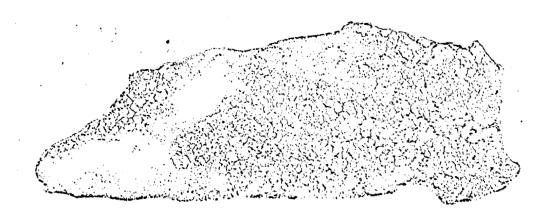


Figure 1.



Figure 2

It must be noted that, although the number of colonies on the Ashby agar is considerably larger than on the Capek agar, the colonies themselves are very

rocks.

In the monoliths below the altered layer, the rock was generally sterile (relatively, of course) providing there were no fissures in it. In other words, the presence of microorganisms in this part is not detected by present-day methods.

We examined 20 specimens of rock gathered from the non-lichen-covered areas and separate stones in various regions of Armenia, ten of which had an altered surface layer and ten did not. Analysis of these specimens indicates that the specimens with an unaltered surface layer are not populated with microorganisms. Microbes are not discovered on these masses or are found in slight number and only on the very surface of the monoliths (Table I). Apparently they are adven- /320 titious forms.

TABLE I. NUMERICAL DISTRIBUTION OF MICROORGANISMS BY LEVEL IN SURFACE LAYER OF ROCKS. NUMBER OF CELLS PER GRAM OF ROCK

Depth	L	ichen Gro	Not Lichen Grown		
in mm	Basalt	Tufa	Granite	Basalt	Tufa
0-2 3-5 7-10 20-30	100 000 45 000 5 000	10 000 5 000 100 0	150 000 65 000 10 000	5 000 100 0 0	1 700 0 0 0

In the monoliths free of lichens and having an altered surface layer, microorganisms were detected in considerable numbers. In two specimens of limestone and in four of basalt and granite in which there were perceptible surface layer changes but no lichens, we discovered developing bacteria, actinomycetes, yeasts, and fungi, as well as algae. The 0-3 mm surface layer of the limestone specimens contained from 50 to 100 thousand microbe cells, including about 5000 algae. In the granite and basalt specimens, there proved to be 60 to 80 thousand cells, among which were also algae in the amount of about 2000 per gram.

In qualitative composition, the microflora of the rocks investigated differ (Table II). The great majority is comprised of bacteria with considerably fewer yeasts, fungi, and actinomycetes.

Asporogenous forms of the genera Bacterium and Pseudomonas, and then mycobacteria, are the principal bacteria found; sporogenous bacilli and micrococci are rarely encountered. All these organisms are colorless. Pigmented forms are discovered in rare cases and only among representatives of the <u>Bacteriaceae</u>.

We had no opportunity for more detailed study of the microorganisms found in other rocks, but Table II illustrates their relationship to nutrient substrata. The findings in Table II demonstrate that the greatest number of bacteria grows on protein media. On must agar we counted only fungi and yeasts; no actinomycetes

small and punctiform, barely perceptible or entirely imperceptible to the naked eye, and most of them had to be examined and counted with a loupe or a low-power microscope. These colonies consist of asporogenous bacteria and mykobacteria. Characteristic of them is their inability to grow in successive subinoculations onto nutrient media. We tried to culture these bacteria on different media — synthetic (Capek's, Ashby's, etc), protein (meat-infusion agar, gelatin), liquid, and agarized. As a rule, they do not grow in the inoculations from Ashby-agar.

Here we apparently have more or less specialized forms of bacteria and mycobacteria which require that a specific medium be present for their growth, since they have become adapted to certain conditions of existence.

The bacteria which develop on ordinary protein media, as well as on Capek's synthetic medium, perceptibly differ in their characteristics from many soil forms which grow in the same media. They subinoculate well onto nutrient media.

We conducted similar studies of limestone specimens collected from different sites in the central zone of the Soviet Union. We subjected the surface layer of monoliths in quarries and of individual slabs and stones found on the surface of the ground and in various structures to microbiological analysis. In most cases the surface of the samples tested was free of lichens, but in a cross-section from the surface there was a thin altered layer distinguished by its darker color. Analysis indicated that these specimens contain a large number of bacteria in the surface layer, and fewer mycobacteria, fungi, and actinomycetes. Many algae are discovered here — green and blue-green. On inoculation onto Ashby agar, the different specimens display a count of from 10 to 80 thousand cells of bacteria and up to 2000 of algae per gram of rock surface layer. This research is not yet concluded while the microorganisms remain uncounted, but the general picture of the findings is the same as in the research on the Armenian limestone specimens.

We observed a unique process of limestone rock disintegration in the fields near Kinel' in the Kuybyshev Oblast. On the slope of a hilly elevation in the fields, there is a large number of small stones whose undersides are greatly corroded and look like sponges. After careful cleansing and washing in sterile water, the sponge-like eroded surface was microbiologically analyzed. The stones were hammered apart; on the fracture the altered surface layer was scraped off and inoculated onto nutrient media.

The analyses showed only the presence of bacteria and a very few actinomycetes. We discovered no algae. The bacteria do not penetrate deeper than 5 mm into the stones. The number of bacteria counted was from 50 to 200 thousand per gram of surface layer of the stone specimens under investigation.

In summing up the above, it may be inferred that bacteria take a great part in the disintegration of rocks. Under the lichens, and in many cases regardless of the presence of lichens, the surface of hard rocks — basalt, tufa, limestone, granite, and, we must assume, others — is settled principally with bacteria, mycobacteria, actinomycetes, fungi, and algae — autotrophs and heterotrophs. With their excretions, these organisms destroy rocks from the surface and invade their depths, forming a surface layer or "weathered crust" which is visible to

the eye. Polynov (Ref. 8) is therefore right in regarding bacteria as the first settlers on uninhabited rocks. These microorganisms destroy the rock with their $\frac{322}{}$ acid excreta, while the products of rock decay are assimilated as sources of mineral nutriment in the form of separate elements.

In the cases where bacteria and other microorganisms develop in the surface layer of rock under lichens, they utilize the organic excretions of the lichens as sources of energy. From our preceding investigations (Ref.5) it is evident that lichens may feed a vast number of microorganisms with their excreta.

In rocks free of lichens, the microorganisms take advantage of the carbon nutriment sources excreted by free-living algae which, as the above makes clear. are almost always discovered together with bacteria, actinomycetes, and fungi.

Their development is not limited to littoral and submarine limestone rocks. Algae, which as a rule are less specialized, inhabit various rocks, not only those bathed in water, but on the dry land, on mountain tops, and in valleys.

Tauson (Ref. 10) has observed massive concurrent development of algae and bacteria on rocks on mountain tops of the Pamirs and the Caucasus, where they form large "coatings." Together with bacteria, mycobacteria, and fungi, they penetrate into the depth of the rock, dissolve it, and thus prepare soil for other subsequent settlers of the plant and animal world. It is these koinoses consisting of free-living bacteria, actinomycetes, fungi and algae which play the primary and most important role in the breakdown of rocks under present-day conditions of soil formation.

It must be remarked that many bacteria, and perhaps fungi and actinomycetes, are themselves capable of assimilating carbon dioxide from the atmosphere and of synthesizing organic matter, using in this the energy which they produce by oxidizing the inorganic reduced compounds of the rocks.

Such bacteria -- autotrophic or protrophic -- can exist without algae or lichens. Polynov (Ref. 9) and other scientists regard these bacteriaas the very first microbe settlers which prepare soil for other microorganisms.

Conclusions

- 1. Microbiological investigations have been carried out on the impaired surface layer of rocks -- basalt, granite, tufa, and limestone -- overgrown with lichens and free of them.
- 2. It has been ascertained that this layer is inhabited by a great number of organisms. The great majority consists of bacteria and mycobacteria with considerably fewer fungi and actinomycetes.
- 3. There is a larger number of microorganisms in the surface layer of basalt, granite, and limestone rocks than in tufa rocks.
- 4. Free-living algae are detected in relatively great quantities on rocks not overgrown with lichens and they are seemingly necessary concomitants to bacterial and fungous flora.
- 5. In their biological characteristics, the bacteria of rocks are unique, differ from the ordinary forms, and belong chiefly to the autotrophs developing

on synthetic media with mineral sources of nitrogen.

6. These bacteria together with free-living algae are the first settlers on the original virgin rocks. They prepare the soil for lichens and other organisms.

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