

FINAL REPORT, 1988 CRATERS OF THE MOON LICHEN PROJECT

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Lichens were collected July 5 and 6, 1988, at Craters of the Moon National Monument and identified in July and August 1988. Air quality was evaluated by submitting a large sample of each of two species of lichens to a simple electrolyte leakage test; chemical analysis of a small sample of pine leaves was also employed as part of the air quality monitoring activity. Energy dispersal spectrometry was used to detect the presence of any heavy metals or other toxic elements in the lichen tissue; however, instrumentation problems prevented completion of that part of the project until December 1989. Reports were sent to Shelley Sparhawk and to Gerry Wright in August and October 1988. The following is a brief summary of those reports plus a summary of developments since that date.

(1) Lichens were collected at 20 locations; 23 species were identified and 22 herbarium specimens, including verification specimens of 12 species, were sent to Ms Sparhawk. (2) Electrolyte leakage measured on 71 specimens of *Letharia columbiana* and *Lecanora melanophthalma* was very low, compared to measurements made at other locations in Idaho, except from lichens collected at the Tourist Camping Area and near a parking area on the main park road approximately a mile south of the Camping Area. (3) TEM (transmission electron microscope) images did not show any serious abnormalities in the lichens collected at Craters of the Moon; minor abnormalities were noted in lichens collected at the Campground Area. (4) At some locations mistletoe had heavily infested *Pinus flexilis*, the tree on which *Letharia columbiana* was collected, and needles on some of the mistletoe infected trees showed classical fluoride toxicity symptoms; however, fluoride levels in the pine needles, although slightly elevated in leaves from the Campground Area, were not high enough to account for visible fluoride toxicity symptoms, suggesting something else, possibly the mistletoe itself, was causing the observed abnormality. (5) The abundance of *Letharia columbiana* at Craters of the Moon, together with its high level of sensitivity and favorable morphological features, indicate that it is an excellent species for biomonitoring air quality at Craters of the Moon National Monument.

Energy dispersal spectrometry (EDS) was employed to evaluate the levels of toxic elements in the atmosphere. Lichens were collected at five locations for this purpose and taken to Provo, Utah, along with lichens collected at the INEL. Thus, the INEL specimens, collected at both pollution free and pollution impacted sites, served as controls for the Craters study, and the Wilderness Area lichens from Craters served as a control for the INEL sample. The EDAX (trademark name for Energy Dispersal Analysis of X-rays) equipment, which had previously given us very

good results in our INEL lichen studies, was frequently breaking down by late summer of 1988, and was replaced at that time by a new EDAX which, it was hoped, would give better results. However, delay in setting up the equipment, followed by hospitalization of Jim Allen, the EDS operator, moved the analysis date for our material back several months, well into calendar year 1989. Finally about March 1989, I received the analysis. To my disappointment, it was purely qualitative, and it was therefore impossible to do a statistical analysis on it.

The printout implicated plutonium in three of the specimens but no indication, of course, as to how much. All three were at locations where we would least expect to find plutonium: a cross-wind site at the INEL, the Wilderness Area at Craters, and Horse Thief Canyon in the Lost River Mountains. On the other hand, there was no hint of plutonium at the one location on the INEL site where we might expect it. In July 1989, I took the 1988 data to Del Miley, EDS analyst for the INEL in Idaho Falls. By careful examination of the spectra, Dr. Miley concluded that these three specimens were showing a "ghost" spectrum (harmonics of the actual spectrum) from the gold shadowing. Each spectrum is made up of several peaks, some of which are often so close to the peaks of the spectra of other elements that they are difficult to distinguish. All of the harmonic peaks expected for gold were present, but only one peak of a spectrum that might be identified as plutonium was present. We concluded that there is no evidence of plutonium in the lichens at Craters of the Moon.

In the fall of 1989, BYU purchased new EDS (Energy Dispersive Spectrometry) equipment from another company, not EDAX. On December 21, 1989, I reanalyzed the material which I collected in the summer of 1988 including some of the material collected at five locations at Craters of the Moon. Even though the lichens were very old (I estimate the age of individual specimens of *Letharia columbiana* collected at Craters of the moon at 40 years for the youngest, at least 200 years for the oldest, and the age of *Lecanora melanophthalma* as considerably older than that) and had been accumulating foreign substances during their entire life span, there was no evidence of appreciable accumulation of heavy metals or other toxic elements in the ones collected at Craters of the Moon. A small amount of copper was found in all of the specimens but its concentration was less in the lichens collected at Craters than in those collected at any of the sites at the INEL (Table 1). Antimony, lead, cadmium, and manganese were also present in some of the lichens from the INEL: manganese was detected in two cells out of 30 evaluated, one of them at Craters of the Moon, and cadmium in one *Lecanora* cell. We specifically checked for plutonium in the same specimens where it had previously been reported, and there was no evidence of it. I believe we can safely conclude that atmospheric pollution is at a very low level at Craters of the Moon National Monument, and that plutonium is not present.

Although the cadmium and manganese values in Table 1 are so few, so scattered, and so low as to lack any statistical signifi-

TABLE 1. Average level of polluting elements in five specimens of *Letharia columbiana* collected at Craters of the Moon National Monument and at the INEL and one specimen of *Lecanora melanophthalma* collected at the INEL. The subjective quality rating is based on direction and distance from the main source of pollution (the ICPP) on a scale of 0-10 with 0 = clean air and 10 = badly polluted urban air

SUBJECTIVE AIR QUALITY RATING	COLLECTION SITE	Cd	Mn	Sb	Cu	Pb
0	Wilderness #20	0	.13	0	.57	0
1	INEL Crosswind-5 km	0	.08	.14	.74	3.5
3	INEL Sec Downwind-5 km	0	0	.33	.68	4.2
4	INEL Sec Downwind-1 km	0	0	0	.69	5.0
5	INEL Prim Downwind-5 km	0	0	.04	.88	5.6
x	<i>L. melanophthalma</i>	.29	0	0	.57	7.8

cance, the averages for antimony, copper, and lead represent measurable levels in a relatively large number of cells. Both lead and copper increased in lichen thalli as the estimated level of pollution at the collection site increased ($r = 0.91$ for lead and $r = 0.73$ for copper, the former statistically significant at $P = 0.05$). This is of special interest because the *Letharia columbiana* specimens collected at the INEL were transplants from a pollution free site in the Lost River Mountains which had been exposed to the pollution from the ICPP for only 7.5 weeks. (The *Lecanora melanophthalma* specimen was also a transplant and had been exposed to pollution at the secondary downwind 5 km site for 13 months; although included in the table it was not included in the calculation of the correlation coefficients, partly because we know that different species of lichens respond differently to exposure to pollution and partly because it had been exposed for a longer period of time.)

Even though our data are limited, Table 1 indicates that the Wilderness Area of Craters is generally free from air pollution. On the other hand, lead, mercury, and possibly some other forms of pollution are high along the west boundary of the Monument near U. S. Highways 20/26, even higher than at the worst sites at the INEL (Rope and Pearson 1990). To understand why this is so, one must know something of the history of the two areas in addition to the biology of lichens. Since lichens are not only extremely long-lived but do not shed leaves, as many other plants do, and do not have excretory systems as animals do, they accumulate much larger quantities of heavy metals and other elements than do most organisms. Lead from the exhaust fumes of automobiles, buses, and trucks have become concentrated in the thalli of lichens near the highway at Craters. Prior to about 1950, on the other hand, the entire INEL area was far removed from automobile traffic and other sources of atmospheric pollution and the lichens there are still relatively free of lead. Mercury, which is soluble in hydrosulfuric and sulfurous acids (H_2S and H_2SO_3 ,

respectively), tends to be concentrated in basalt outcroppings along faults where sulfurous hot springs existed in the recent past, probably about 2000 years ago along the west border of the Monument. The lichens we have collected along some of the fault scarps on the west boundary of the Monument have been much higher in mercury than in any of the lichens collected at the INEL.

Figures 1-5 are electron microscope photographs of cross sections of *Letharia columbiana* thalli collected at Craters of the Moon and at the INEL. Three tissues are present in *L. columbiana* and other fruticose lichens: a parenchyma tissue, the cortex, which completely surrounds the thallus and is made up entirely of fungal cells; a chlorenchymatous inner cortex, or algal layer, containing both algal and fungal cells; and a prosenchymatous medulla containing only fungal cells. These tissues can be seen in Fig. 1 which is a scanning electron microscope (SEM) picture of a specimen of *Letharia columbiana* collected in the Wilderness Area of Craters of the Moon National Monument. Note how compact the cells in the cortex are and how loose the medulla is. Arrows point to three algal cells, the largest of which appears to have haustoria from a hypha in the medulla penetrating it.

Figures 2-5 are transmission electron microscope (TEM) photographs of lichen tissues. Fig. 2 shows cells from the medulla of three lichens, the upper and bottom ones collected at two of the most heavily polluted sites at the INEL, the center one of a healthy lichen collected in the Wilderness Area at Craters showing well formed concentric rings typical of healthy lichenized fungi; the concentric rings are damaged to the extent that they are hardly visible in the top and bottom cells. Fig. 3 and 4 show the effects of pollution on the internal membrane systems in lichens: the upper cells in each case from heavily polluted sites, the lower cell in Fig. 3 from the Campground Area at Craters of the Moon and the lower cell in Fig. 4 from a pollution free site at the INEL. The upper photo in Fig. 5 shows the plasma membrane pulled away from the cell wall and badly torn (arrow, lower left) in a lichen from the most heavily polluted site at the INEL; the lower photo shows the membrane also pulled away from the cell wall but still intact in a lichen from the slightly polluted Campground Area.

In the summer of 1990, I would propose developing an interesting display for the enjoyment of park visitors telling about the lichens at Craters of the Moon, their diversity and distribution, how to recognize some of the more common species, something of their evolution and their role in ecological succession, and, of course, what they can tell us about air pollution and its effects on plants and animals, including humans. I have additional transmission electron microscope pictures that show damage to cell membranes and organelles in lichens exposed to high levels of atmospheric pollution, in addition to the originals of the ones copied in Fig. 2-5, that could be used in such a display.

I would be happy to help develop a display for use at the

Information Center or elsewhere at Craters of the Moon. I would also be happy to collect lichens along the east boundary of the Monument to monitor any possible pollution coming from either the INEL or the phosphate plants around Pocatello using either the simple (and inexpensive) Pearson Electrolyte Leakage Test or Energy Dispersal Analysis, or both. I realize, of course, that budget problems always exist; however, by running the material from Craters with the INEL material, two important goals can be achieved: incorporating the Craters material into a larger batch of material cuts the cost of energy dispersal analyses significantly, and we at the INEL gain the advantage of additional controls for comparison with our material collected on our established transects radiating from our major source of pollution, the ICPP (Idaho Chemical Processing Plant). The results we get from these control sites would also be available to you. There is such a wealth of lichens here that Craters of the Moon ought to be the outstanding National Park unit for the display of this resource. And really good, clean air is becoming such a rare commodity, that you ought to do all you can to advertise its presence at Craters.

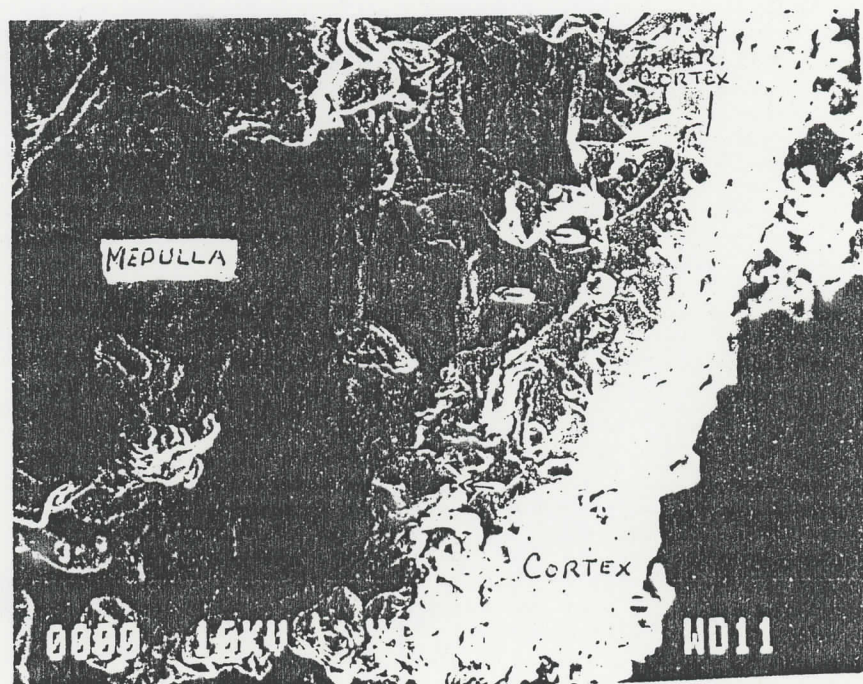
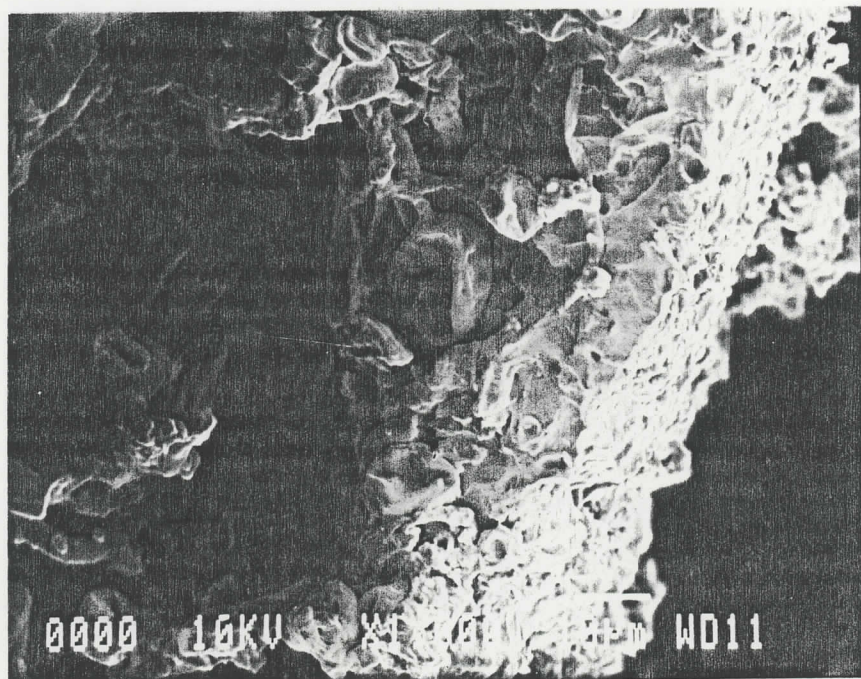


FIG. 1. Scanning electron micrograph of cortex, inner cortex (algal layer), and medulla of *Letharia columbiana*. Arrows point to three algal cells in the inner cortex.

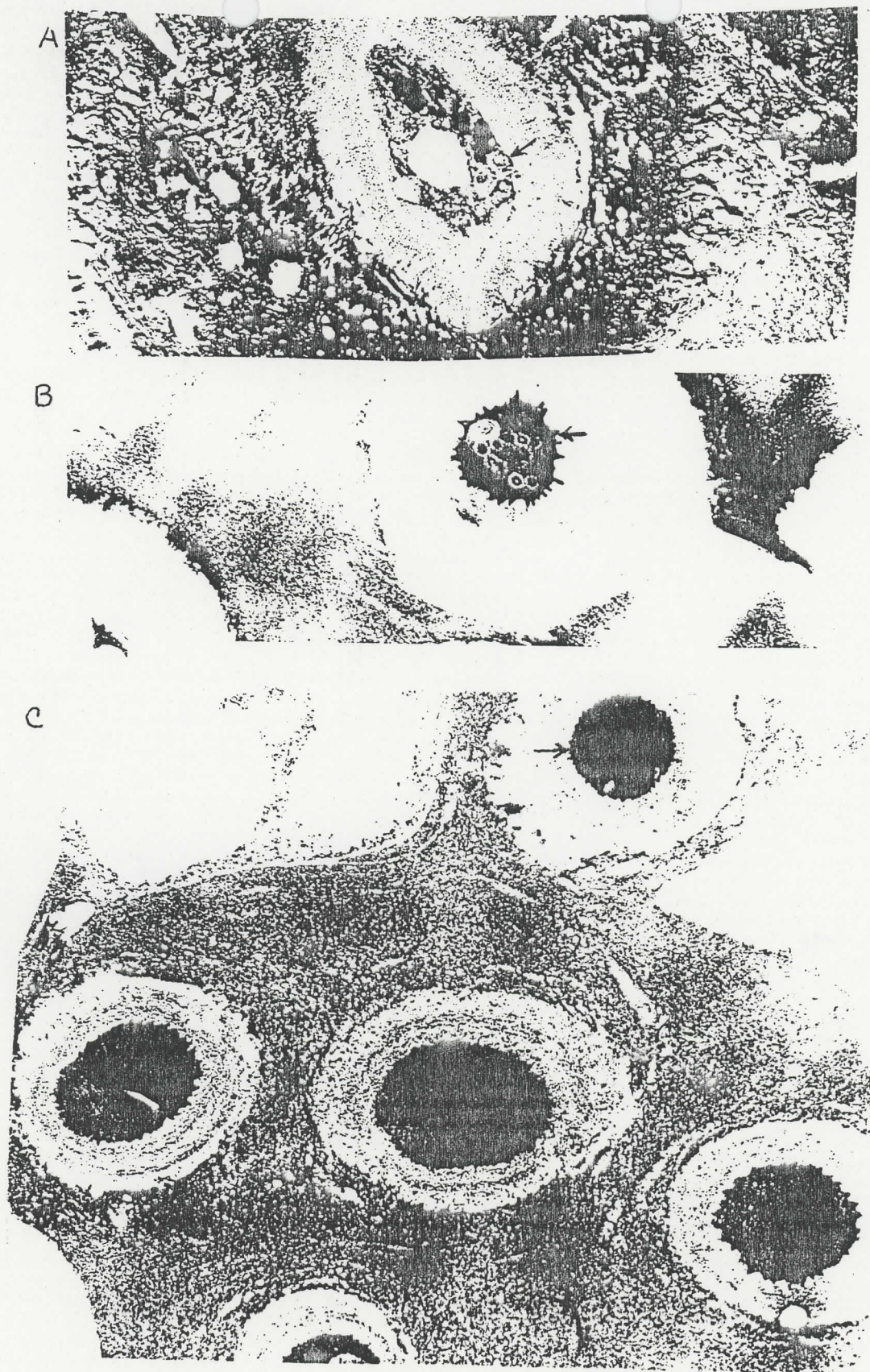


FIG. 2. Transmission electron micrographs of fungal cells in the medulla of *Letharia columbiana*. About six structures called concentric rings can be clearly seen in the middle photograph taken of a healthy lichen but the concentric rings in the top photo (INEL site 215° 3 km) and bottom photo (55° 3 km) are damaged, probably from exposure to SO_2 , and not readily visible.



FIG. 3. TEM photos of a pollution damaged cell (215° 3 km) and a slightly damaged cell (Craters #5, Campground Area).



FIG. 4. TEM photos of a pollution damaged cell (55° 1 km) and a healthy cell (295° 10 km) showing the difference in the internal cell membranes making up the endoplasmic reticulum and dictyosome structures

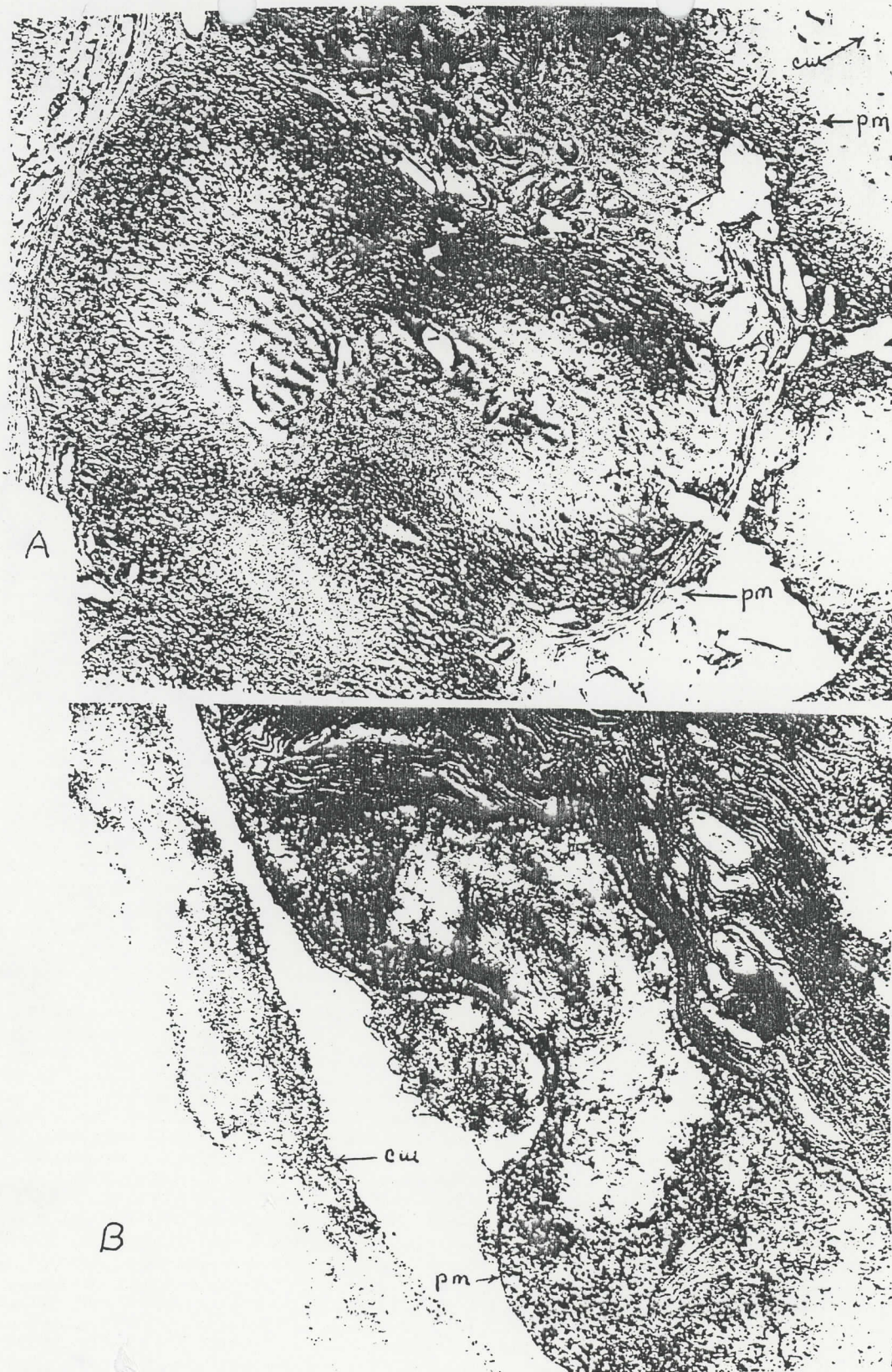


FIG. 5. TEM photos of a severely pollution damaged cell from the INEL (55° 1 km) and a slightly damaged cell from Craters of the Moon (#5, Campground Area). Note that the plasma membranes are pulled away from the cell walls in both cells, but that the plasma membrane is intact in the lower cell and disrupted and torn in the upper cell.