Search for Porphyrins in Lunar Dust

Abstract. *Evidence for porphyrins was obtained in the Apollo 11 bulk sample of lunar dust by fluorescence spectrometry and analytical demetallation. The indicated porphyrins showed major fluorescence excitation at 390 nanometers. Abundance was about* 10^{-10} *gram of porphyrin per gram of dust. Similar pigments were found in exhaust products from tests of a lunar descent engine. The similarity of results suggests that most if not all of the indicated porphyrin aggregate of the lunar sample probably was synthesized from rocket fuel during the landing of the lunar module. These compounds may be the product of a novel high-temperature synthesis of cosmochemical interest.*

The present study was undertaken to determine whether porphyrins, tetrapyrrole organic pigments, were present in the Apollo 11 bulk sample and whether their presence in the sample was indicative of a biological origin (*1*). The results of the analyses show that pigments with spectral and chemical characteristics resembling porphyrins are present in the lunar bulk fines, but it appears that the compounds may have been introduced by the lunar descent rocket engine fueled with unsymmetrical dimethylhydrazine. Thus, while the analyses reported here evidently failed to reveal porphyrins which were unequivocally indigenous, the findings suggest that the porphyrins can perhaps be synthesized through a novel reaction involving simple compounds containing carbon, nitrogen, hydrogen, and oxygen. The pigments have also been detected in experiments simulating primitive (prebiotic) planetary conditions (*2*), and similar compounds evidently exist extraterrestrially, because porphyrins have been detected in a number of samples of carbonaceous chondrites (see *3*).

The analyses reported here are part of a comprehensive analytical program established by the Chemical Evolution Branch of the NASA Ames Research Center for the examination of all organic matter in the returned lunar samples (*4*). The group arranged to pool samples of lunar dust received by the principal investigators involved in the program. The particular sample discussed here was designated "10,086 bulk A fines" by the Lunar Receiving Laboratory.

Prior to receipt of the lunar samples, a simulation experiment was carried out on a sample of the Pueblito de Allende meteorite. Although analyses on a number of specimens of this meteorite showed porphyrins to be present in very small amounts (*5*), no porphyrins were detected in the particular sample used in the control run, and the results were substantially the same for a blank of Ottawa sand which had been fired at 1000°C for 48 hours.

The overall analytical procedure for the lunar sample is described by Ponnamperuma *et al.* (*4*). The lunar dust (54.6 g) was extracted directly with benzene-methanol (9:1), and 23 percent of the extract was made available for porphyrin analysis.

On concentration of the solution to a volume of 1 ml, weak emission bands at 600 and 630 nm for excitation at 400 and 425 nm (Turner 210 spectrofluorometer) appeared. Clear-cut excitation peaks for emission at 600 nm were observed at 390 and 455 nm, the latter being much more intense. Corresponding readings for the blank of Ottawa sand showed only a low background and no discernible peaks. Absorption spectrometry (Cary 14, 0 to 0.1 optical density) showed no distinct spectral features, only a gently rising background with decreasing wavelength. The solution was examined in an improved magnetic circular dichrometer with a superconducting magnet of 50,000 gauss; the data were evaluated by computer. Detection limits for the system were estimated to be 7 ng/ml for recognition of porphyrin compounds in metal-complexed form. Magnetic circular dichroism spectra of the concentrated extract solution, however, did not show the presence of metalloporphyrins above detection limits.

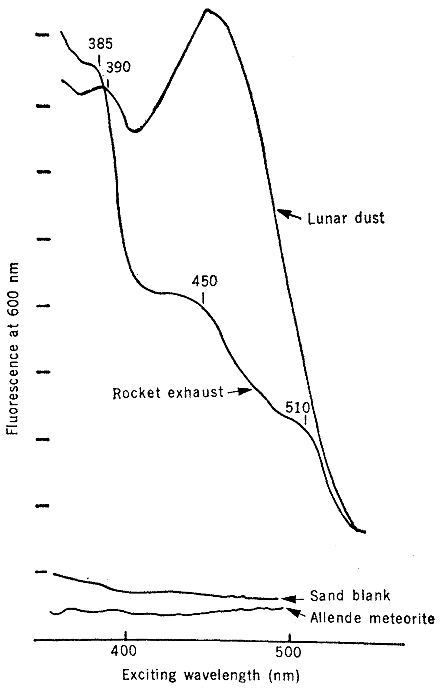

Fig. 1. Fluorescence excitation spectra for extracts of (i) lunar dust from the bulk sample, (ii) dunite exposed to the exhaust of a rocket engine fired at NASA White Sands Test Facility, (iii) crushed Pueblito de Allende carbonaceous chondrite, and (iv) Ottawa sand ignited at 1000°C for 48 hours. Instrument operating parameters were set for highest sensitivity in all cases except that of the rocket exhaust, in which fluorescence was suppressed by a factor of 20.

Free-base porphyrins fluoresce freely, but most metal complexes fluoresce only faintly. Thus, controlled demetallation is useful for the detection of traces of porphyrin metal complexes (*6*). The extract solution was divided into two portions, one four times the volume of the other. Each was reduced to dryness in a spectrofluorometer cell and taken up in 1 ml of diethyl ether. The smaller sample showed a faint excitation band at 410 nm for emission measured at 660 nm upon addition of 2 percent of the demetallating reagent, methanesulfonic acid (MSA). The intensity of the band increased at 7 percent MSA, and at 19 percent MSA the band was replaced by an excitation peak at 388 nm, which became more intense on further addition of MSA.

Recovery of the presumed free-base porphyrins was accomplished by neutralizing the solution with sodium acetate and extracting with ether. The ether layer showed faint excitation bands at 385 and 450 nm. Transfer to 6*N* HCl also showed a weak peak at 390 nm for emission measured at 600 nm, and this peak was interpreted tentatively as a Soret band of porphyrin pigments. To extend the identification of the apparent porphyrin, it was complexed with copper to quench its fluorescence. This was done by transferring the porphyrin to glacial acetic acid and adding cupric sulfate. The excitation band disappeared after a few minutes at room temperature, in accordance with the behavior of authentic porphyrins.

The same analysis was repeated on the other portion of the lunar extract, which corresponded to 10 g of lunar dust. Essentially the same results were obtained, with the emergence of a band at 390 nm for emission at 600 nm on the addition of 20 percent MSA. After demetallation, the fluorescence of the pigments in ether was still weak, but definite emission could be detected at 630 nm for excitation features at 390 and 415 nm. Extraction with 6*N* HCl produced an aqueous layer with emission peaks at 600, 628, and 685 nm. A strong excitation peak was observed at 390 nm, and it appeared to be accompanied by a much weaker peak at 550 nm. The "porphyrins" were transferred back to ether and reextracted with HCl of increasing concentration. The acid strength necessary to transfer the pigments from the organic layer was 4*N* HCl; from this we tentatively concluded that few polar substituents were present to promote the solubility of the pigment in the aqueous phase. Formation of the copper complex resulted in the complete suppression of fluorescence. On the assumption that the pigment was porphyrin, the amount present was estimated to be 1 ng, for a concentration of 10^{-10} g of porphyrin per gram of lunar dust.

The sand blank was examined in the same manner, and it showed no indication of porphyrins above background. In addition, reagents, glassware, and handling procedures were checked for possible contaminants.

A similar demetallation analysis was carried out on the benzene eluate from silica-gel chromatography of the major portion of the benzene-methanol extract of the lunar dust. The hexane and methanol eluates were not available for porphyrin analysis. The benzene eluate left after the completion of other tests appeared to show the presence of a small amount of porphyrin pigments.

Samples of rocket exhaust were made available from the test firing reported by Simoneit *et al.* (*7*). These comprised samples from trap A, a trap for the exhaust gases cooled to liquid-nitrogen temperatures, and trap E, one containing dunite. Material from trap A showed an intense fluorescence background on which a broad excitation band (in methanol and ether) appeared at 450 nm for emission in the range 600 to 700 nm. In HCl, the exhaust products turned a bright yellow, and an excitation band at 390 nm for emission at 610 nm was clearly present. Absorption spectrometry showed only the yellow color. While the foregoing results suggested porphyrins in the free-base state, MSA demetallation of the total exhaust material—that is, without any chemical fractionation—showed evidence for metal complexes with a 412-nm excitation band appearing at 4 percent MSA. This spectral feature persisted to a concentration of 23 percent MSA; at higher concentrations of MSA a much stronger band emerged at 385 nm and shifted gradually to 392 nm. Attempts to recover free-base porphyrins from the foregoing aqueous acid layers were unsuccessful, however.

Material from trap E gave similar results. Extraction of the sample was carried out in the same manner as that for the lunar sample. A broad excitation band was evident at 450 nm, as in the case of trap A material, but on an intense background. Addition of MSA gave typical demetallation results for metalloporphyrins. At a concentration of 25 percent MSA a strong peak emerged; it increased in intensity with increasing acid content up to 50 percent MSA. Final location of the band was at 387 nm, and the corresponding emission peak was at 595 nm. Magnetic circular dichroism analysis of material extracted from trap E showed a large number of compounds, but none could be definitely identified as porphyrins.

To confirm and extend the previous results and to demonstrate adequate

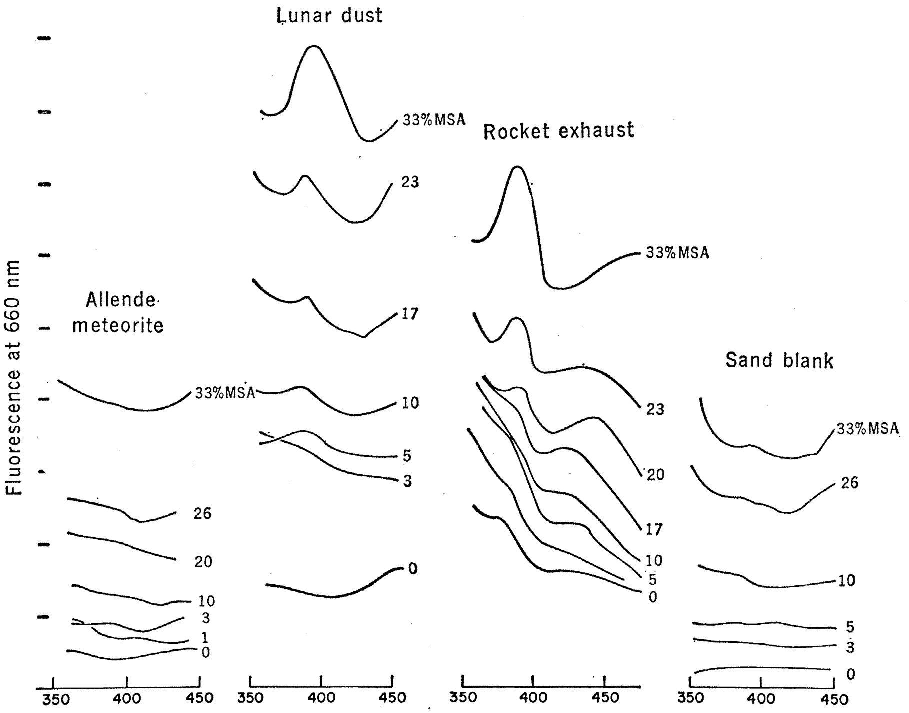

Fig. 2. Demetallation analysis of extracts showing emergence of excitation peaks in the region of the Soret band of porphyrins with increasing content of methanesulfonic acid (MSA). Operating parameters of the instrument were the same in all cases; portions taken for the analyses of lunar dust and rocket exhaust were, respectively, 75 and 11 percent of the total extracts.

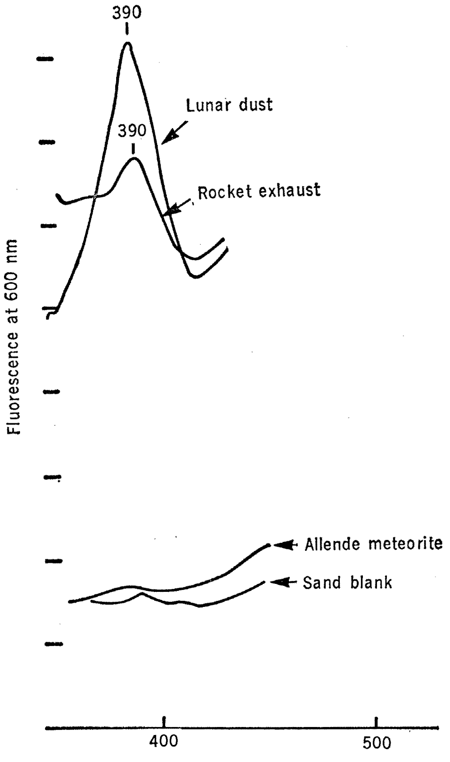

Fig. 3. Fluorescence excitation spectra in Soret region for pigments recovered from MSA demetallation analysis. Operating parameters for spectrofluorometer were the same in all cases. Solvent, 6*N* HCl.

control over laboratory procedures, an additional set of samples was analyzed. Approximately 10 g each of the same lunar dust, trap E dunite, Pueblito de Allende meteorite, and Ottawa sand were extracted with a solution of benzene-methanol (9:1). The extracts were identified by a code unknown to the analysts (G.W.H. and E.P.), and complete analyses were carried out. Figure 1 shows fluorescence excitation curves for the extracts. Demetallation caused by the addition of MSA is illustrated in Fig. 2, and Fig. 3 shows the results of procedures for the recovery of free-base porphyrins. The results from the lunar and exhaust samples confirmed the earlier analyses, and what appear to be free-base porphyrins were recovered from the products of MSA demetallation of both the lunar dust and the rocket exhaust. Results for extracts from both the sand and meteorite were negative.

Organic matter is undoubtedly synthesized extraterrestrially, as indicated for example by the carbonaceous matter of chondrites and the generally accepted graphitic interstellar dust (*8*). Simple compounds containing nitrogen, carbon, hydrogen, and oxygen are reported in interstellar dust clouds (*9*) and in stellar atmospheres (*10*). Recent theories (*11*) postulate that much of interstellar space is occupied by thermally stable gas phases at 10^4 °K, which suggests that high-temperature synthesis of organic matter including porphyrins throughout space is within reason.

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References and Notes

1. J. E. Falk, *Porphyrins and Metalloporphyrins* (Elsevier, New York, 1964); C. Ponnamperuma and N. W. Gabel, *Space Life Sci.* **1**, 64 (1968); K. A. Kvenvolden and G. W. Hodgson, *Geochim. Cosmochim. Acta* **33**, 1195 (1969).
→ G. W. Hodgson and C. Ponnamperuma, *Proc. Nat. Acad. Sci. U.S.* **59**, 22 (1968).
3. G. W. Hodgson and B. L. Baker, *Geochim. Cosmochim. Acta* **33**, 943 (1969).
4. C. Ponnamperuma, K. Kvenvolden, S. Chang, R. Johnson, G. Pollock, D. Philpott, I. Kaplan, J. Smith, W. Schopf, C. Gehrke, G. Hodgson, I. Breger, B. Halpern, A. Duffield, K. Krauskopf, E. Barghoorn, H. Holland, K. Keil, *Science*, this issue.
5. G. W. Hodgson, E. Peterson, K. A. Kvenvolden, C. Ponnamperuma, in preparation.
6. G. W. Hodgson, E. Peterson, B. L. Baker, *Mikrochim. Acta* 805 (1969).
7. B. R. Simoneit, A. L. Burlingame, D. A. Flory, I. D. Smith, *Science* **166**, 733 (1969).
8. B. Donn, N. C. Wickramasinghe, J. P. Hudson, T. P. Stecher, *Astrophys. J.* **153**, 451 (1968).
9. L. E. Snyder, D. Buhl, B. Zuckerman, P. Palmer, *Phys. Rev. Lett.* **22**, 679 (1969); A. C. Cheung, D. M. Rank, C. H. Townes, D. D. Thomson, W. J. Welch, *ibid.* **21**, 1701 (1968).
10. M. S. Vardya, *Mon. Notic. Roy. Astron. Soc.* **134**, 347 (1966).
11. G. B. Field, D. W. Goldsmith, H. J. Habing, *Astrophys. J.* **155**, L149 (1969).
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Organogenic Elements and Compounds in Surface Samples from the Sea of Tranquillity

Abstract. *Organogenic elements, mainly carbon, nitrogen, phosphorus, and sulfur are present in the particulate material and in a breccia rock from Tranquillity Base in amounts ranging from 5 to 4200 parts per million. The major compounds of carbon released by heating are carbon monoxide and carbon dioxide; the former predominates. Small amounts of other compounds of carbon have also been observed. Sulfur can be released as hydrogen sulfide by treatment with acid. The carbon isotopic* $\delta^{13}C$ *values are definitely nonterrestrial (+ 13 to + 18.5 per mil).*

An igneous origin has been considered likely for the Sea of Tranquillity (*1*), and has since been confirmed by a preliminary examination of its surface rocks (*2*). This precludes the existence of large amounts of organic matter on this part of the lunar surface. Therefore, it was considered pertinent to analyze for those elements and compounds that are necessary for prebiological or biological synthesis of organic molecules (*3*). The compositions of the fine particulate material (type D) and a small specimen of breccia (type C) were studied with different methods of analysis. To avoid contamination and the introduction of artifacts, the samples were analyzed as directly as possible and with a minimum of handling. Thus the fines were neither sieved nor pulverized and the breccia was cleaved only once.

Spark-ionization mass spectrometry was used to determine the concentration of hydrogen, carbon, nitrogen, phosphorus, and sulfur (*4*). We used a mass spectrograph (CEC X21-110) that operated at a resolution of approximately 3000, and at a relative sensitivity that ranged from 0.0005 to 0.03 ppm. Each sample, of about 250 mg, was sparked against a pure gold electrode. Up to 20 mg of material from each sample was actually analyzed. If the sample is not homogenous, variations in the analytical values will be observed depending on the composition of the area sparked. All the samples were isolated from the laboratory environment by means of glove boxes up to and including insertion into the mass spectrometer vacuum system. Cryogenic rough pumping, ion pumping, and preliminary sparking assured a minimum of contamination present during actual measurements. The fine material was vacuum encapsulated, hydrostatically pressed to 4570 kg/cm² and then cleaved for analysis. The small breccia rock was cleaved and immediately analyzed at selected surfaces. Calibration analyses with six National Bureau of Standards specimens of Special Ingot

Table 1. Spark ionization mass spectrographic analysis for organogenic elements in type D (fines, sample 10086,6) and type C (breccia, sample 10002,54) lunar samples. Values expressed in ppm.

Element	m/e	Bulk fines			Breccia		
		Ia	Ib*	II	Fresh inner cut	Outer surface (dark area)	Outer surface (white spot)
H	1	1.2	1.2	0.41	0.13	0.011	0.083
C	12	9(5)†	24	41	27	6	5.5
N	14(7)	14	20	7	15	10	10
P	31	270	187	110	110	250	59
S	32	4200	590	1000	2800	2100	1000

* Same as Ia analyzed after 2 weeks of exposure to the terrestrial environment. † Second analysis performed the following day on the same sample without its removal from the instrument.