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MICROBIOLOGICAL BURDEN ON THE SURFACES OF THE AIMP SPACECRAFT

PART 1

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NASA GODDARD SPACE FLIGHT CENTER
GREENBELT, MARYLAND

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SUMMARY

The Anchored Interplanetary Monitoring Platform (AIMP) spacecraft was decontaminated and monitored for microbiological contamination during the first stage of assembly.

The decontamination procedure, using isopropyl alcohol, resulted in a two-log reduction in the microbial population.

Extrapolation and projection of the counts obtained to the completely assembled spacecraft indicated that the total spacecraft would have a population of 1×10^7 viable organisms before decontamination, and 2.8×10^5 viable organisms after decontamination.

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PART 1

INTRODUCTION

The decontaminating the Anchored Interplanetary Monitoring Platform (AIMP) was performed in compliance with the "NASA Spacecraft Decontamination Policy" for the decontamination of lunar landing hardward as stated in Management Manual 4-4-1.*

The AIMP was decontaminated and assembled in a class 100 laminar crossflow room. All surfaces of the spacecraft were monitored for microbiological contamination immediately before being occluded. This program, which is probably the first attempt to measure the microbiological contamination of a spacecraft during assembly, should provide valuable data, as well as valuable experience and methodology, for decontaminating and sterilizing vehicles for planetary landing.

The primary objective of the AIMP is to measure interplanetary magnetic fields, solar plasma, energetic particles, and micrometeorite fluxes in the vicinity of the moon. As the primary concern is the successful mission of the spacecraft, no decontamination or microbiological procedure which would jeopardize the mission is being used.

This report describes the decontamination and monitoring procedure during the first stage of assembly, and the results. The work was performed November 30, 1965.

MATERIALS AND METHODS

Spacecraft

Figure 1 shows the configuration of the AIMP spacecraft. The surfaces sampled were:

*The Spacecraft Integration and Sounding Rocket Division was assigned the responsibility for decontamination of AIMP. The Space Biology Branch provided technical direction and assistance and determined the microbial burden on the spacecraft surfaces.

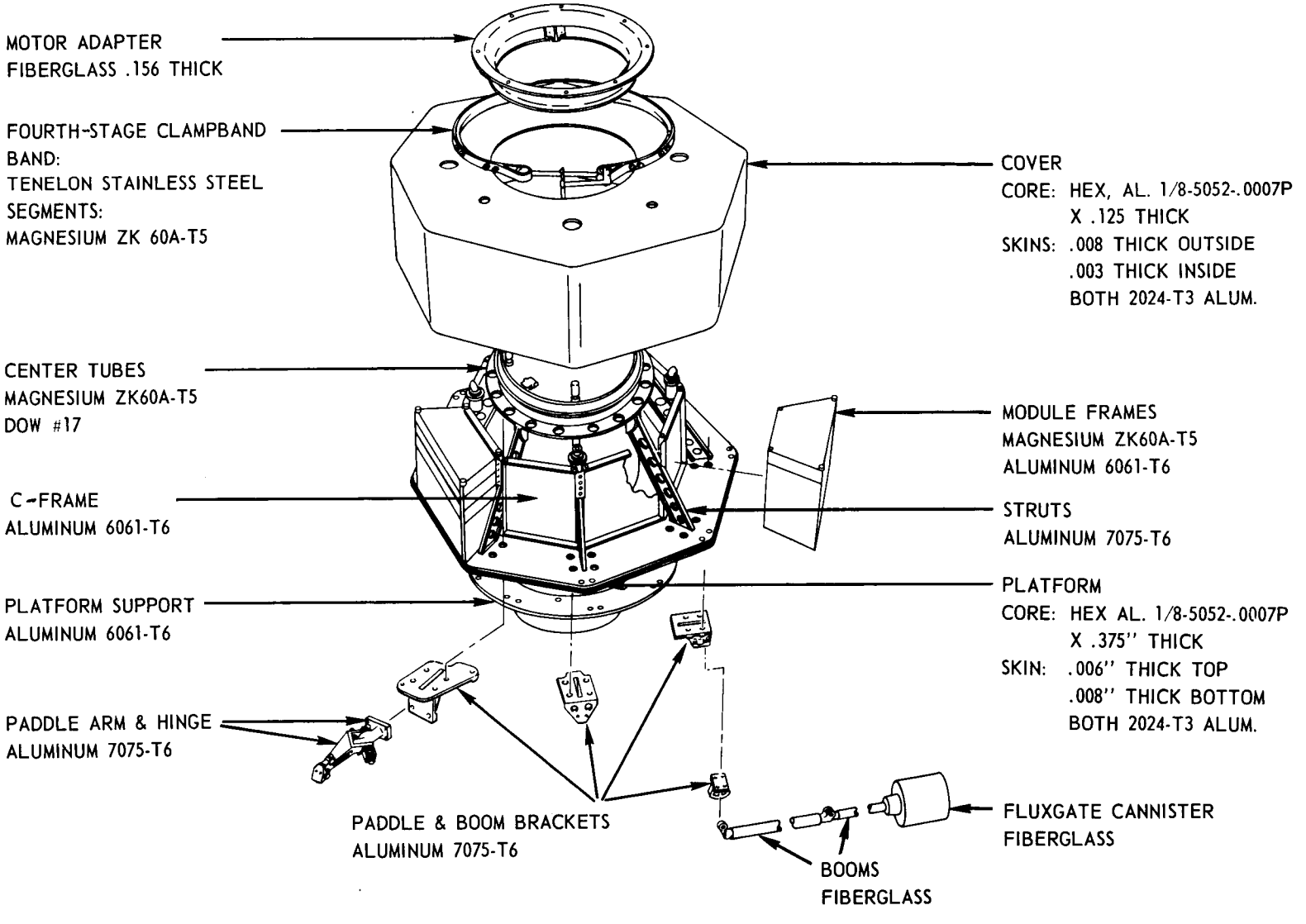

Figure 1. AIMP Spacecraft Showing Spacecraft Configuration and Structural Materials

Mating Platform Surface for C-Frame — This platform is octagon-shaped, 27 inches from facet to facet. Each facet is 6 inches long and there is a hole in the center of the platform 10 inches in diameter. The surface area of the platform is approximately 400 square inches. The surface of each of the eight sections of the platform which mate with the C-frame was sampled. The total area of each mating surface sampled was 3.5 square inches. Facets are lettered A through H.

C-Frame Base — The C-frame is also octagonal, with sections lettered A through H to match the underlying platform. The base of each of the eight sections of the C-frame which mate with the platform has a surface area of 3.5 square inches.

Mating Platform Surface for Lower Support Ring — The opposite side of the mating platform for the C-frame has a highly polished aluminized surface. The surface of this platform which mates with the lower cone-support ring was sampled.

Lower Cone-Support Ring — This ring supports the C-frame which is bolted to it. The ring has a surface area of 136 square inches. The surface of the ring which mates with the underlying platform was sampled.

Figure 2 is an artist's concept of the spacecraft.

Decontamination

All surfaces of the spacecraft were sampled before and after decontamination. The decontaminating agent used was 2-propanol (90 percent). After the "dirty" samples had been taken, the spacecraft was cleaned with the alcohol in the receiving area of the clean-room complex and brought through an air shower into the class 100 laminar crossflow room.

The spacecraft was placed approximately 1 foot in front of the bank of air inlet filters for decontamination and assembly. All personnel remained downstream of the spacecraft at all times during the assembly.

The surfaces of the spacecraft were then decontaminated again with 90 percent 2-propanol, and "clean" samples were taken. The alcohol was applied with cleanline wipers (ester type polyurethane foam) which were changed frequently. Swab samples were taken after the alcohol had completely evaporated, usually within 10 minutes after decontamination.

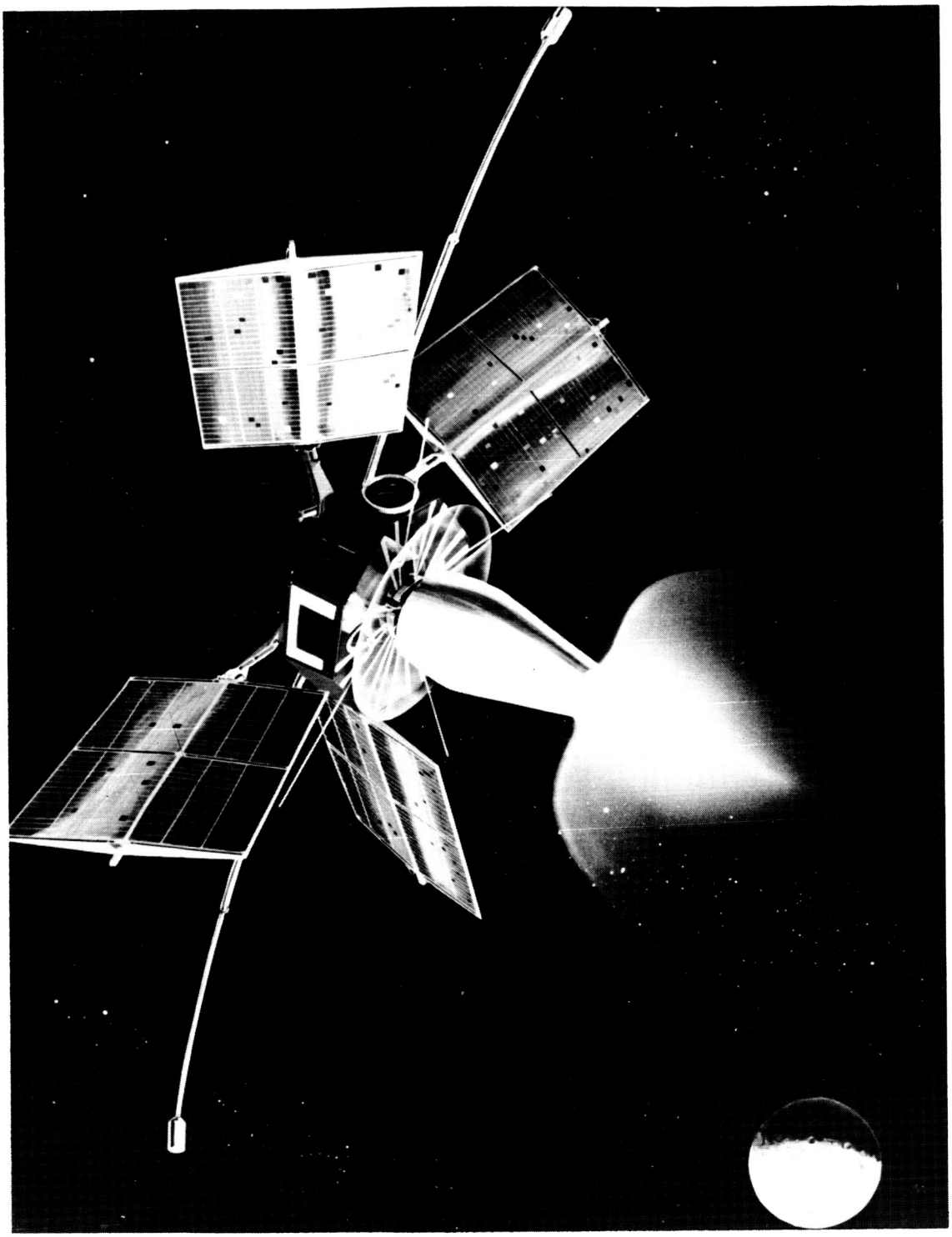

Figure 2. Artist's Concept of AIMP Spacecraft

Sampling Procedure

Sterile cotton swabs wetted with sterile distilled water were used to sample all surfaces of the spacecraft. Screw-cap tubes containing 10 ml of distilled water were sterilized by autoclaving for 15 minutes at 121°C. Cotton swabs were sterilized, five to a test tube (16 × 150 mm), by autoclaving 30 minutes at 121°C.

Just before sampling, the swab was inserted into the tube of distilled water and moistened. Excess water was removed by wringing out the swab on the inside wall of the tube. Quantitative data were obtained by sampling 4-square-inch (2 by 2 inches) area outlined by sterile Kraft paper templates. Because templates were sometimes prepared to fit the configuration of the spacecraft, some templates had sampling areas measuring 1 by 4 inches and some, 0.5 by 4 inches (2 sq. in.).

The surface within the squared area was scrubbed approximately 25 times in one direction and 25 times at right angles. If a piece part was small, the entire surface area was sampled and measured. Several template samples were taken from large areas.

The swab was then aseptically broken off into a tube of sterile distilled water. Each tube was shaken mechanically for 5 minutes in a gyro-rotary shaker followed by vigorous manual shaking for 2 minutes. This procedure broke up the cotton swab adequately.

Colony counts were made by plating out 4-ml aliquots in duplicate into sterile disposable petri plates (Fisher). Culturing was usually accomplished within 1 hour after breaking off the swab in the tube of distilled water.

Media

Tryptic soy agar (Difco) was used as the growth medium. Twenty ml were added to each petri plate containing the 4-ml aliquots from each tube.

Screw-cap tubes containing 10 ml of sterile distilled water were used for "washing" the cotton swabs.

Incubation

All plates were incubated at 35°C for 72 hours. Negative plates were incubated an additional 24 hours.

Clothing

All personnel who entered the clean room wore lint-free coveralls, boots, cap, hood, face mask, and sterile rubber surgical gloves. Clean-room procedure was observed at all times (air shower was used before entering the clean room, etc.).

Tools

All tools were decontaminated with 90 percent 2-propanol and, when not in use, were kept immersed in the alcohol during assembly. When possible, tools were sterilized.

RESULTS

Table 1 shows the microbiological counts obtained from the mating surfaces of the C-frame and mating platform. Decontamination with 2-propanol reduced the count on the C-frame from 8832 organisms per square foot before decontamination to 72 organisms per square foot after decontamination. Counts from the platform mating surface were reduced from 48,462 organisms per square foot before decontamination to 200 organisms per square foot after decontamination. The number of organisms occluded between the C-frame and mating platform, based on counts obtained, was 54. This number was obtained by adding the counts obtained from each facet of both the C-frame and platform mating surface. The total area occluded was 56 sq. in.

Table 2 shows the microbiological count obtained from the lower cone support ring and mating platform surface. Paper templates with a 4 sq. in. sampling area were used and samples were taken at random. The average microbial count on the platform was 173 per 4 sq. in. (6228 per sq. ft.) before decontamination and 52 per 4 sq. in. (1883 per sq. ft.) after decontamination. The average count obtained from the lower cone support ring was 3 organisms per 4 sq. in. (97 per sq. ft.) after decontamination. The high average count from the platform after decontamination was due to one sample which yielded a count of 492 organisms per 4 sq. in. (17,712 per sq. ft.). The other 9 samples from the same surface yielded counts ranging from 2.5 per 4 sq. in. to 16 per 4 sq. in. (90 to 576 per sq. ft.) after decontamination (Table 2). The total microbial population occluded between the lower cone support ring and the mating platform surface was 1852, based on a total occluded surface area of 272 sq. in.

Table 1

Microbial Contamination of C Frame and Mating Platform Surface (11-30-65)

Facet	Before Decontamination				After Decontamination			
	Platform		C Frame		Platform		C Frame	
	Org/3.5 in^2	Org/ft^2	Org/3.5 in^2	Org/ft^2	Org/3.5 in^2	Org/ft^2	Org/3.5 in^2	Org/ft^2
A	107	4,402	70	2,880	2.5	103	4	164
B	450	18,500	35	1,440	2.5	103	0	0
C	392	16,100	107	4,402	6.2	254	4	164
D	525	21,600	1032	42,500	5.0	206	1.2	50
E	7092	290,200	165	6,800	14.0	575	1.2	50
F	272	11,200	147	6,050	5.0	206	1.2	50
G	167	6,900	125	5,150	1.2	50	1.2	50
H	432	17,800	35	1,440	2.5	103	1.2	50
Average Organisms/ft^2		48,337		8,832		200		72
Total Organisms Occluded: P + C = 56 in^2 = 54								

Table 2

Microbial Contamination of Lower Cone Support Ring and Mating Platform Surface

Before Decontamination			After Decontamination				
Sample* No.	Platform		Sample* No.	Platform		Support Ring	
	Org/4 in^2	Org/ft^2		Org/4 in^2	Org/ft^2	Org/4 in^2	Org/ft^2
1	135	4,860	1	0	0	3.7	133
2	255	9,200	2	0	0	0	0
3	43	1,550	3	0	0	1.2	43
4	697	25,000	4	16	576	7.5	270
5	345	12,400	5	2.5	90	3.7	133
6	20	720	6	6.2	223	2.5	90
7	8	288	7	492	17,712	5.0	180
8	185	6,670	8	0	0	1.2	43
9	25	900	9	3.7	133	1.2	43
10	15	540	10	2.5	90	1.2	43
Average Org.	173	6,213	Avg. Org.	52	1,883	2.72	98
Total Organisms Occluded: (P + R) = 272 in^2 = 1,852							

*Samples were taken randomly and do not represent the same areas.

DISCUSSION

The microbial population was significantly reduced by decontamination with 2-propanol. The one exception to this was sample 7 from the platform after decontamination (Table 2). The high counts from this sample indicate that this area of the platform was not properly decontaminated.

If it is assumed that the swab technique is only 50 percent efficient, a doubling of all the counts obtained will give a close approximation of the microbial population on the surfaces sampled.

The above data represent the first samples taken for microbiological contamination of the AIMP during the initial assembly of the spacecraft. Sampling for microbial contamination will continue for each phase of the assembly until the spacecraft is completely assembled and ready for launch. The samples will be taken from all surfaces immediately before launch. When the spacecraft is completely assembled, the counts will be averaged and an attempt will be made to estimate the microbiological population of the total spacecraft. Whenever possible, the entire surface of a spacecraft part will be sampled. If the surface area to be sampled is large, a number of samples will be taken and the counts averaged to get a representative distribution of the population.

Assuming that the average count per square foot obtained in this initial survey is representative of the total spacecraft, the total count per spacecraft when completely assembled may be estimated.

The spacecraft, when assembled, will have a total surface area of 500 square feet. By averaging the counts per square foot of spacecraft shown in Tables 1 and 2, one arrives at an estimated count per 500 square feet of spacecraft: Before decontamination, the estimated count per spacecraft is 1×10^7 organisms; after decontamination, the estimated count is 2.8×10^5 organisms. This represents approximately a two-log reduction in microbial numbers. Throughout this study and through each assembly phase, an estimate will be made of the number of viable organisms per square foot of surface of the spacecraft.

The primary concern throughout this study is the successful mission of the spacecraft. Therefore, extreme care will be taken so that decontamination and microbiological procedures will not jeopardize the successful completion of the mission.

No known procedure is capable of recovering 100 percent of the organisms present, but the standard "damp swab" procedure used in this study is easy to use, lends itself to any surface configuration and is reproducible; recovery is equivalent to any other procedure.

Sterile distilled water was used as the suspending medium and wetting agent for the swab because it is compatible with the spacecraft surface and leaves no residue. Saline could not be used because it would cause degradation of the surfaces; peptone or any other organic medium would leave a residue. Because the wash fluid is cultured within 1 hour, there should not be any injury of cells due to plasmolysis.

Alcohol was chosen for the decontaminating agent because it is compatible with surfaces of the spacecraft and acceptable with project engineers. Alcohol is also a very good decontaminating agent. It is safe, relatively inexpensive and readily obtained. Alcohol acts quickly, evaporates readily, and leaves no residue. It also exerts a cleaning action. Isopropyl alcohol was chosen because it is more bactericidal than ethyl alcohol.* It is also a better fat solvent than ethyl alcohol and is tax-free and nonpotable.

* Anon: Cleaning, Disinfection, and Sterilization. State of California Department of Public Health, Bureau of Hospitals. 1962. p. 20.