Bacteriological Investigation of the Pollution at Syowa Station in Antarctica

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大腸菌を指標とした昭和基地周辺の汚染に関する調査研究

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要旨: 昭和基地周辺地域における人為汚染の実態を明らかにするために，腸内細菌 (大腸菌) を指標とした細菌学的調査を行った.

調査対象は，飲用水を中心に，水道，土壌，大気，雪および氷である．その結果，以下のことについて明らかとなった.

第 15 次隊医学担当隊員により設定された，定観測地点の土壌中から，腸内細菌 (大腸菌) は検出されなかった．しかし，飲用水，水道水からはしばしば検出された．水道水への細菌汚染の経路が追求され，人為汚染経路が明らかにされた.

現在のところ，病原細菌による汚染はなかったが，隊員の健康を守るために，適切な予防対策が講じられることが望まれた.

人為汚染の他の生物への影響が同様に細菌学的方法で調査され，一羽のオオトウゾクカモメの腸管内から大腸菌が検出された.

Abstract: Bacteriological investigation of pollution caused by human being was conducted at Syowa Station. Special attention was paid to the contamination of Enterobacteriaceae (*E. coli*).

The soil, water, air, snow and ice were examined both at Syowa Station and Mizuho Camp. The following results were obtained.

E. coli was not detected at the bacteriological pollution monitoring points which were set up by the 15th Japanese Antarctic Research Expedition (JARE–15). On the other hand, in the waterworks and in the drinking water at Syowa Station, *E. coli* was detected from many parts. But no pathogenic bacteria have been found so far. However, some means of effective prevention should be considered to ensure healthy living at Syowa Station. Some birds were also examined in terms of bacteriological pollution.

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1. Introduction

The Antarctic has been considered as the area intact from any pollution caused by human being. As a matter of fact, however, there are some innegligible problems at Syowa Station from hygienic point of view. Some of these problems are concerned with the disposal of the remnants of food, drinking water-supply and so on. As Syowa Station is an isolated and limited society, its pollution with pathogenic bacteria would become a grate danger to health. What we should do first is to realize the actual conditions of the station in which the wintering members have to keep their health. In addition, there is the problem of conservation of natural environment.

2. Sampling Sites

2. 1. Drinking water

Water for use at Syowa Station is supplied mostly in the following way: Water from Daiichi-dam and Aragane-dam is stored in a 130 k*l* tank, and is led to the residential area via a 10 k*l* tank from December to March. Ice or snow are melted in the 10 k*l* tank from April to November and is led to the residential quarter. Water is obtained from snow or ice by melting it in the tank of 10 to 20 *l* capacity at room temperature. During a short period of construction before wintering, water is supplied from Daiichi-dam directly to the workshop.

Water samples for this study were taken from Daiichi-dam, Aragane-dam, snow and ice of the places in question. The water in 130 k*l* tank, 10 k*l* tank, and other small tanks was examined. The water from every faucet along the pipelines was also sampled.

2. 2. Filthy water

The draining pipe from the bathroom is led to the tide crack of the sea together with the pipe from the lavatory. The outlet of the sewage pipe is placed windward of the 10 k*l* tank; this is a problem. Furthermore, the sewage tanks in the residential area, especially in the mess hall and the bar were important subjects of our investigation.

2. 3. Soil

Bacteriological pollution monitoring points for soil and sand were set up for continual checking at Syowa Station and its vicinity.

2. 4. Human feces

The feces of men was subjected to the investigation with special reference to the survival of Enterobacteriaceae in a cold environment.

2. 5. Effects of pollution on other creatures (penguin, McCormick's skua)

The possible effects of pollution caused by living of human beings on other creatures were investigated with a bacteriological method.

3. Methods

3. 1. Place and time of sampling

Monitoring points in East Ongul Island (March–April 1974, June 1975), Mizuho Camp (November–December 1974), and Syowa Station, water supply and drinking water (May 1974, June 1975).

3. 2. Water

The water was sampled aseptically and kept in the sterilized test tube. Therefter, it was cultivated on the cultural plate in a chamber of the Environmental Science Laboratory.

3. 3. Soil and sand

The sample was taken aseptically into the plate with a sterilized spoon from the earth surface. Water was added at the ratio of 1 m*l*/1 g soil, and mixed in a mixer. Then the supernatant fluid was cultured with the ordinary media.

3. 4. Snow and ice

Samples were picked up aseptically, melted at room temperature, and cultivated by usual procedure.

3. 5. Culture of bacteria

Nutrient agar medium, Brain-heart infusion, BTB (Drigalski modified agar medium), EMB, and MacConky agar medium were used in most cases. The sample was cultivated at 37.0°C for 24 hours. The bacterial colony was transplanted to the identification field and cultivated again at 37.0°C for 24 hours. The culture media were prepared at the Environmental Science Laboratory and also at Mizuho Camp, if necessary.

E. coli is used as the indicator of the pollution caused by human being, because it is the most popular bacteria among Enterobacteriaceae and generally used as an indication of pollution. *E. coli* itself is not pathogenic and it is the most useful microorganism for determining the grade of man-caused pollution.

4. Results

4. 1. Sampling site

As shown in Table 1, 335 samples were taken at Syowa Station and 52 samples at Mizuho Camp. Fig. 1 indicates the monitoring points set by JARE–15. These points were set up in radial arrangement of 16 directions on the concentric circles with radius of 100 m, 200 m, 400 m, 600 m, and so on around the center point, the bar (that was used as the living and mess room by JARE–1 to JARE–7). Some of the total 75 points were shifted a little from the should-be points on the map for convenience. Each point

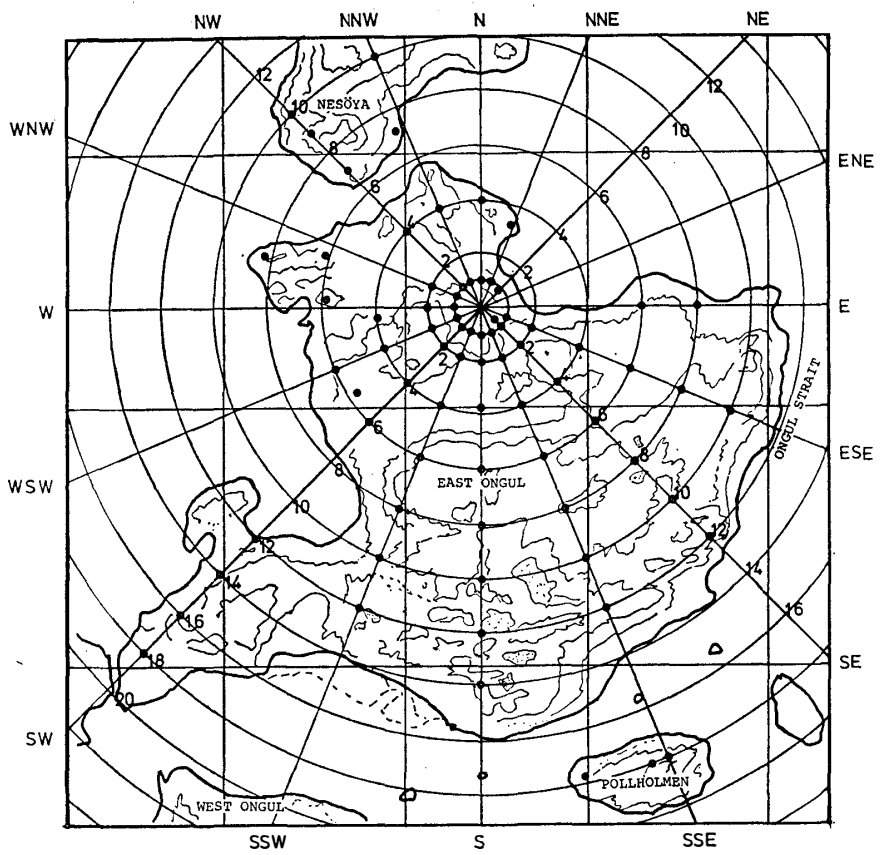

Fig. 1. Sampling points for monitoring bacterial pollution of soil by man in East Ongul Island. The points were set by JARE-15 (medicine).

is named after the distance and direction from the center point; for example, "S–6" means South 600 m from the bar, and "NNW–10" North-North-West 1,000 m from the bar.

4. 2. Soil, sand, snow and ice

Soil and sand samples from these monitoring points were subjected to the examination at the Environmental Science Laboratory. No *E. coli* was detected in these samples.

Half the bacterial species found in these soil or sand samples are gram positive rod, and gram negative species including Enterobacteriaceae (*E. coli*) were 32 or 24.8% of all identified species.

Bacterial species isolated from water, snow, ice, soil, sand, and air were classified in terms of gram-stain and microscopic findings. As shown in Table 1, 22 gram negative rods and 29 gram positive rods were found in the water samples from Syowa Station. On the other hand, a relatively large number of gram positive cocci were detected in the air. In the soil and sand, rods were found more than cocci. At Mizuho Camp, most

Table 1. Samples collected in Syowa Station and Mizuho Camp, and type of bacteria.

Syowa Station

Sample	Sample number	G⁻/Bac.	G⁺/Bac.	G⁻/Mic.	G⁺/Mic.
Water	111	22	29	3	0
Soil	133	4	9	0	3
Snow and ice	51	4	12	1	3
Air (room)	27	0	9	1	17
Others	13	2	9	0	1
Total	335	32	68	5	24
Total detect. number=129 (100%)		(24.8%)	(52.7%)	(3.8%)	(18.6%)

Mizuho Camp

Sample	Sample number	G⁻/Bac.	G⁺/Bac.	G⁻/Mic.	G⁺/Mic.
Water	4	0	1	0	0
Soil	0	0	0	0	0
Snow and ice	30	10	10	0	0
Air (room)	15	2	4	0	3
Others	3	0	1	1	1
Total	52	12	16	1	4
Total detect. number=33 (100%)		(36.4%)	(48.5%)	(3.0%)	(12.0%)

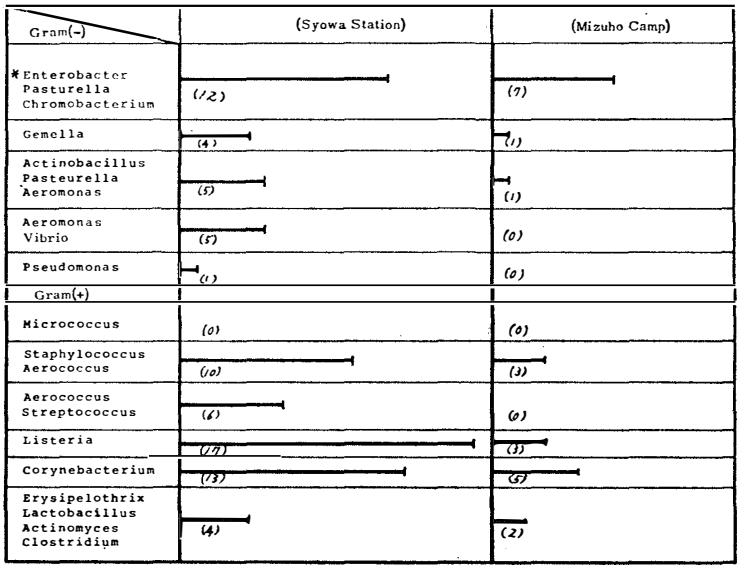

Fig. 2. Bacterial flora classified according to the Cowan's method.

species found in the snow and ice samples were rods and there were only a few cocci.

As the first step of identification of bacterial species, Cowan's method (COWAN, 1974) was used for each sample. The result is shown in Fig. 2. Many species are to be included in Enterobacteriaceae.

4. 3. Water supply

The water supply and sewage system at Syowa Station is illustrated in Fig. 3. Special attention was paid to the 130 k*l* and 10 k*l* tanks for storing water. Enterobacteriaceae were detected in the water of 130 k*l* tank and in the water supplied from 10 k*l* tank to the bathroom, dining room, bar, and Geoscience Laboratory.

These species were found also in the water of Daiichi-dam and Aragane-dam. The mess hall, meteorological station, telecommunication room, and Environmental Science Laboratory have no water pipe from the water tank, and water is obtained by melting ice in small tanks at room temperature. Enterobacteriaceae were detected in every

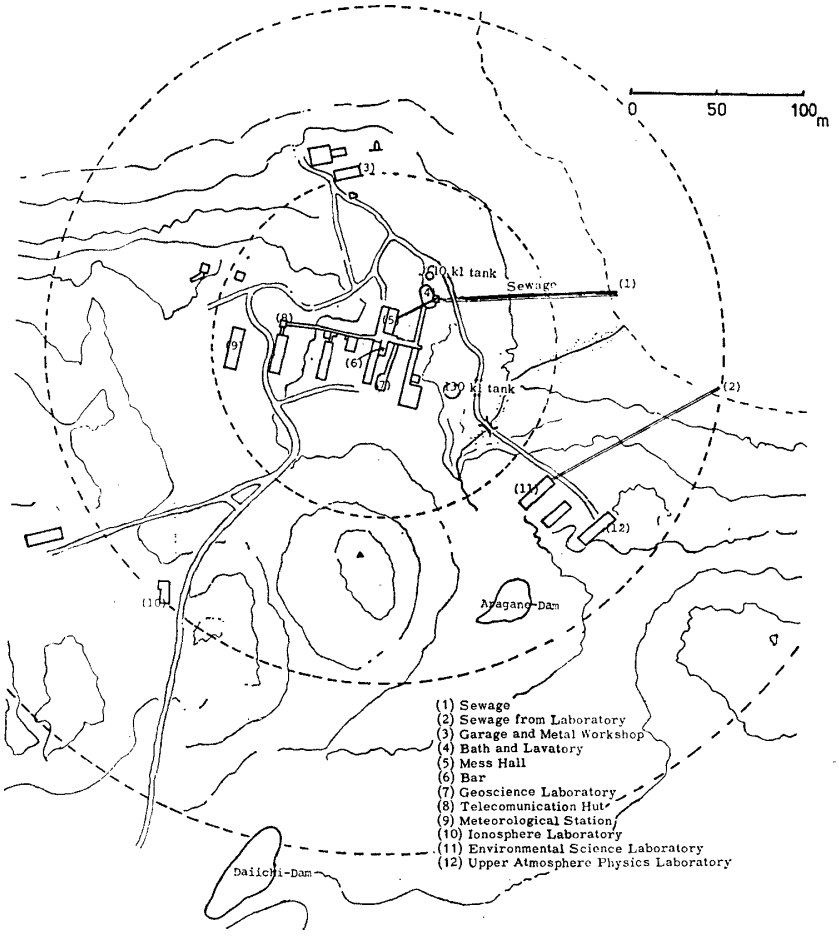

Fig. 3. The waterworks at Syowa Station. Water for bathroom (4), kitchen (5), and bar (6) is supplied from 10 kl tank through pipeline.

Table 2. The number of Enterobacteriaceae and other bacteria in the waterworks and drinking water of Syowa Station.

Place	Enterobacteriaceae (/m*l*)	Other bacteria (/m*l*)
1. Outlet of sewage pipes	1×10^{4}–10^{8}	1×10^{4}–10^{8}
2. Bathroom: Water supply	2–4	0
3. Bathroom: Warmer water supply	0–2	0
4. Mess hall: Melted ice water	4–7×10^{1}	1–2×10^{2}
5. Kitchen: Water supply	2×10	1×10^{2}
6. Kitchen: Warmer water supply	0	0
7. Bar: Water supply	3×10^{1}	1×10^{1}
8. Bar: Warmer water supply	0	0
9. Hand washing (disinfected)	0	0
10. Generator room: Water tank	3–4	3–7
11. Generator room: Warmed water room	0	0–2
12. 10 k*l* tank	1–2×10^{1}	4×10^{2}
13. 130 k*l* tank	1–2×10^{1}	3×10^{1}
14. Geoscience Labo.: Water supply	3	0
15. Meteorological Station: Melted ice water for drinking	8.2×10^{2}	2.3×10^{3}
16. Ionosphere Station: Melted ice water for drinking	1.4×10^{2}	3.5×10^{2}
17. Telecommunication room: Melted ice water for drinking	3.4×10^{2}	1×10^{4}
18. Observation Station: Melted ice water for drinking	0	1.9×10^{3}

water tank except that in Environmental Science Laboratory where distilled water is provided for drinking (Table 2).

Usually the members at Syowa Station drink tea, green tea, and coffee. But in the mess hall they often drink unboiled water from the tank, in which Enterobacteriaceae are detected at the ratio of 4–7×10^{1}/m*l*. In the water of the Observation Station, no Enterobacteriaceae where found, but there were other kinds of bacteria at the ratio of 1.9×10^{3}/m*l*. From the snow below the sewage pipe, $1\times10^{4}\sim10^{8}$/m*l* Enterobacteriaceae were detected.

Fig. 4 shows these results schematically. The asterisk shows the site where Enterobacteriaceae were detected.

The pollution of the 10 k*l* water tank should not be overlooked, because it has been used and will be used more frequently than any other tanks at Syowa Station. As shown in Fig. 3, the sewage pipe is placed windward of the tank. To examine the dispersion of bacteria from the outlet of the sewage pipe, the snow between the outlet of pipe and the 10 k*l* tank was sampled. Enterobacteriaceae were detected 1×10^{3}/m*l*

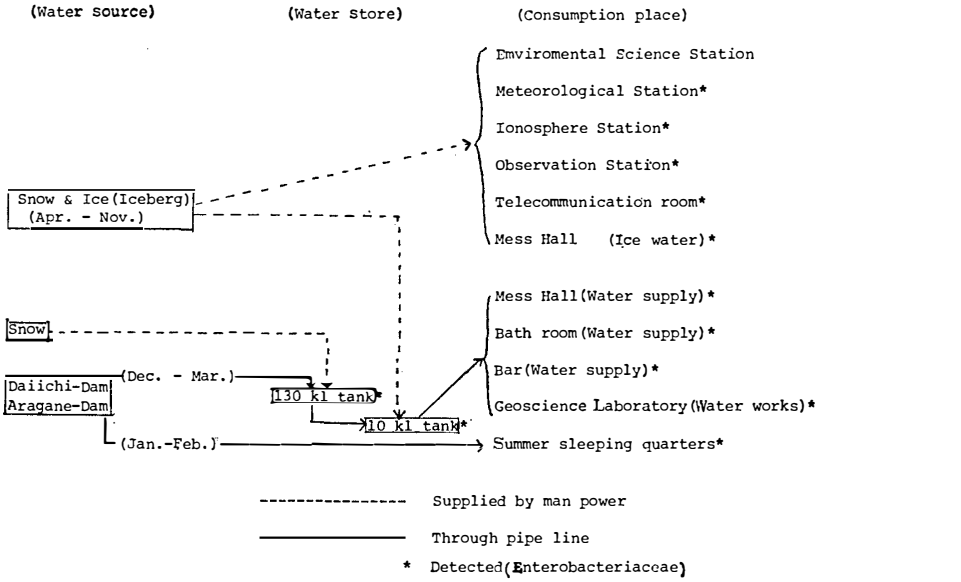

Fig. 4. The route of water supply at Syowa Station. The 10 kl tank and 130 kl tank were polluted with Enterobacteriaceae. The ice water for drinking in mess hall was remarkably contaminated.

in the samples taken near the outlet of the sewage pipe. However, the dispersal of bacteria seemed to be restricted in a small area around the pipe-outlet, and no microorganism of human origin was detected at the site 10 meters apart from the 10 k*l* tank.

In our repeated experiments of air bacteria, nothing was detected in the air. As it is quite difficult to protect the caltivating plates from freezing, though the time of exposure outdoors was only 5–10 minutes, a special box was made to keep them warm outdoors using a handy heater.

4.4. Fresh snow

Fresh snow was taken from 28 sampling points, which are indicated in Fig. 5. No microorganism was found in any sample.

4.5. Survival test of enterobacteriaceae in cold natural environment

In the feces left on the sand in East Ongul Island during April 1974 and January 1975, Enterobacteriaceae were detected. Enterobacteriaceae were found also in the feces left on the snow at Mizuho Camp through three winter seasons since JARE-13.

4.6. Penguin and McCormick's skua

Table 3 shows the results of the investigation of Enterobacteriaceae in the internal organ of two Adélie penguins and a McCormick's skua. Enterobacteriaceae were not found in the internal organ of Adélie penguins, but were detected in the digestive tract from the anterior part of stomach to the anus of a McCormick's skua.

McCormick's skua sometimes comes flying to Syowa Station and customarily eats

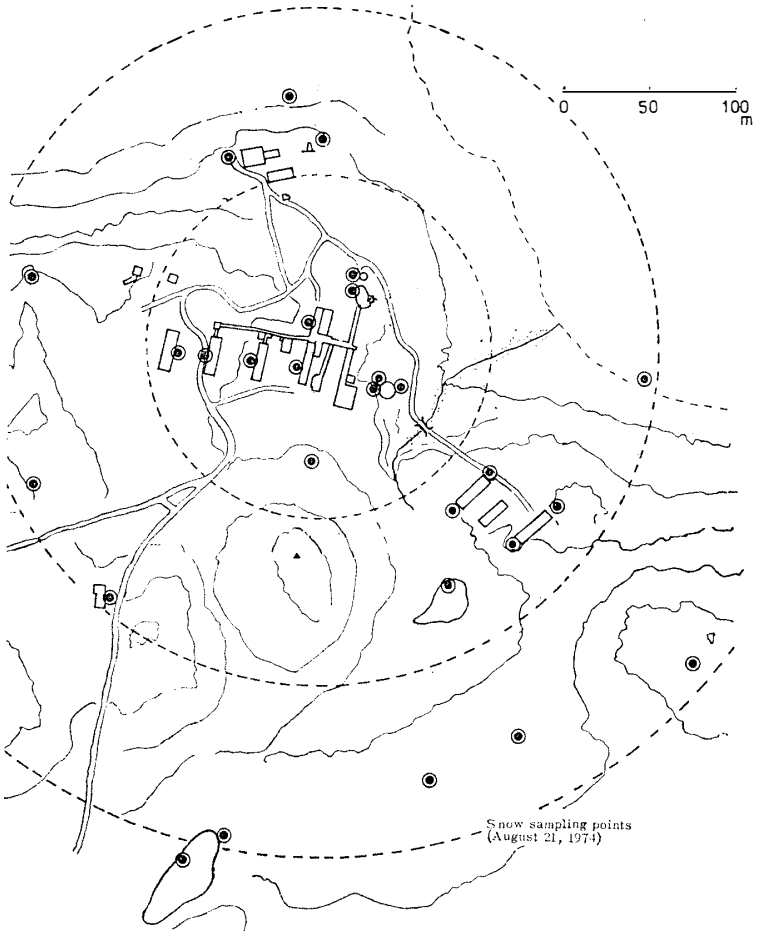

Fig. 5. The sampling points of fresh snow at Syowa Station (⊙). No E. coli was detected.

Table 3. E. coli within the gastrointestinal tract of two Adélie penguins and a McCormick's skua. E. coli was detected from the skua.

Part of internal organ	Density of *E. coli*-fage (/ml)	Inspected fage	RNA-fage (%)	RNA-fage group I	II	III	IV
Adélie penguin							
Stomach (anterior)	0	0	0	0	0	0	0
Intestine (super.)	0	0	0	0	0	0	0
Intestine (medius)	0	0	0	0	0	0	0
Intestine (infer.)	0	0	0	0	0	0	0
Anus	0	0	0	0	0	0	0
McCormick's skua							
Stomach (anterior)	3	1	0(0)	0	0	0	0
Intestine (super.)	2×10^1	2	0(0)	0	0	0	0
Intestine (medius)	3×10^1	4	0(0)	0	0	0	0
Intestine (infer.)	1×10^3	26	0(0)	0	0	0	0
Anus	1×10^3	38	7(18.4)	0	7	0	0

remnants of food in the garbage, which may account for the result mentioned above.

5. Discussion

The purpose of this study is to make clear the actual situation of man caused pollution at Syowa Station and its vicinity, with special reference to bacteriological pollution.

The water supply system in the residential area of Syowa Station was found to be contaminated with Enterobacteriaceae. The bacterial contamination of drinking water in the 10 k*l* and other tanks is a serious problem to consider. Every man handles these tanks with gloved hands, which might be responsible for water pollution. Therefore, the gloves worn by eight men were subjected to the bacteriological examination. Six out of eight pairs were found to be contaminated with Enterobacteriaceae.

At Mizuho Camp, however, water is obtained by melting the snow which is treated carefully with specially prepared gloves. This may explain the absence of enterobacteriaceae in the water there.

As shown in Table 4, the water temperature in the 10 k*l* tank is 11.9°C which is warm enough for these bacteria to survive or even to proliferate. Water, especially drinking water, in these small living areas must be kept as clean as possible. As the present condition of water supply and sewage system is far from satisfactory, we should take a positive step to prevent any possible troubles in future.

As far as Enterobacteriaceae contamination is concerned, problems might be found

Table 4. The water temperature in the waterworks and water tanks in Syowa Station.

Place	Water temp. (°C)	Room temp. (°C)
10 k*l* tank	19.5	
130 k*l* tank	6.0	
Generator room (Warmer water tank)	48.5	
Generator room (Water tank)	35.0	
Kitchen: Water supply	30.0	
Kitchen: Warmed water supply	40.5	
Kitchen: Filty water in room	23.0	17.5
Melted ice water for drinking in mess hall	1.5	
Bar: Water supply	19.5	
Bar: Warmed water supply	46.0	12.0
Bar: Filty water in room	9.5	
Bathroom: Water supply	32.0	36.0
Bathroom: Warmed water supply	50.0	

20 May 1974, 15:00–15:30 Air temperature: −12.0°C Wind velocity: 0.3 m/s

primarily in the sewage system. As a matter of fact, snow, soil, and water are contaminated mainly along the sewage pipes. Another problem is the feces which is usually left outdoors on the snow for a long period of time and Enterobacteriaceae certainly can survive in this condition.

At the bacteriological pollution monitoring points set up by JARE-15 no bacterial pollution is recognized at present. We should make a good use of these points to continually check the possible expansion of man-caused pollution hereafter.

6. Conclusion

Bacteriological investigation of soil, snow, ice, and air pollution caused by human being was carried out in the area of Syowa Station and Mizuho Camp. The area 100 m apart from the station is not bacteriologically polluted as yet. In residental regions, however, most parts, paticularly the drinking water, are contaminated by enterobacteria. It is most likely that the working gloves and clothes worn by the members while handling snow and ice to make water, can carry enterobacteria into drinking water.

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Reference

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