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# 漢江下流 感潮水域에 있어서의 微生物分布

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# Microbial populations in Han River estuary

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## ABSTRACT

Examining the microbial populations in the Han River estuary, we conducted this experiment at six sites of the estuarine area, Chollyu-ri, Cholsan-ri, Wolgon-ri, Chogi-ri, Inhwa-ri, and Oepo-ri for 5 months since May 1967. From the results obtained it could be summarized as follows.

1) The salinity of the estuarine water increased in order of the distances from the base point of the Old Han River Bridge to every site of the estuary, and *p*H of the water, which were between 7.3 and 8.1, showing little difference each other in both the sites and dates of experiment.

2) The populations of the general bacteria and coliform group bacteria were highest at the site of Chollyu-ri, and it decreased with the downstreaming of the river water toward Oepo-ri site. As for fungi which have comparatively high tolerance to the salinity, its population was shown highest at Chogi-ri.

3) The relationship between the salinity and the number of the general bacteria have indicated that the incresed salinity reduced the growth rates of the bacteria. By this it can be assumed that the fresh-water bacteria decreased due to the sea water as well as its dilution effects.

4) The high pollution of the esturarine water was caused by the increases of inhabitants and water thrown by industries in Seoul and Kyung-In Industrial District as well as the excrements fertilized to the farms.

## INTRODUCTION

The Han River, which is one of the largests in Korea and had beautiful sceneries, originates at the Taibaek Range near DMZ area, Kangwon-do. She flows toward the Yellow Sea through several provinces, Chungchongbuk-do, Kyunggi-do and, Seoul. The river water is being utilized in many cases. It supplies drinking water to Seoulites of over four millions, industial water to many factories in Kyung-In Industrial District, and water for agricultural uses.

And then, the river water has been polluted day by day with the increasing inhabitants of Seoul and the sewage water thrown by many plants. These facts have come to close up as an issue of public damage to the residents in Kyung-In District.

Since 1965 we have been examining the pollution of the river water near Seoul and distribution of microorganisms in accordance with it. From May to September 1967 we studied the distribution of coliform bacterial group mainly, which roles as indicator organisms of water pollution, along with fungi and general bacteria at several sites of the Han River estuary where fresh water is being mixed with sea water near Kangwha Island. The results were delivered from statistical treatments, and thus, we present them here.

## MATERIAL and METHODS

For the examining the changes of microbial populations on the estuuarine water of the polluted Han River in relation to the changes of salinity, we have determined the numbers of general bacteria, coli form group and fungi, and measured and analyzed several environmental factors such air temperature, water temperature, hydrogen ion concentration, dissolved oxygen and salinity.

At each of collecting points in each of six major collecting sites (Fig. 1), we collected 300 ml. of surface, medium and bottom water. These collections were carried out monthly from May to September 1967.

All the 300 ml. samples from a major collecting site were mixed to minimize errors in sampling. From this 3,000 ml. sample, six test tubes of approximately 50 ml. of water each were saved, sealed and placed on ice, and transported to the laboratory.

On arriving at the laboratory, the six 50 ml. samples were thoroughly remixed together, and five 1 ml. samples were taken, each placed in 9 ml. of sterile water. Dilutions were continued thus to $1\times10^{2}\sim1\times10^{5}$ and 0.1 ml. samples of each were inoculated on the surface of a total of 15 agar plates, three from each of the five samples, under aseptic condition within 2 minutes after the final dilutuons were made.

Three kinds of culture media were used for the inoculations. For general bacteria, Difco standard nutrient agar was used; for the coliform group of bacteria, Eosin Methylene blue agar was used with MPN tests; and Czapek's agar for fungi. Inculated plates were inverted and placed in an incubator at 37±1°C. Coliform bacteria plates were removed in 18 hours, general bacteria in 48 hours, and fungal plates remained for from 7 to 9 days. Colonies were counted using the Quebec colony counter.

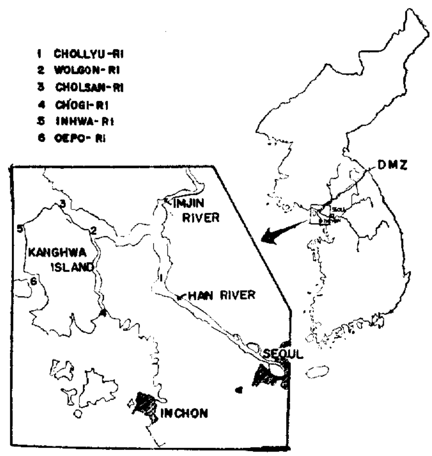


**Fig. 1.** Experimental sites where water samples were taken.

Salinity was determined by Mohr's method. First we obtained the chlorinity by using the silver nitrate titration method with potassium chromate as an indicator. And thus, the salinity was calculated by the following equation (Martin Knudtsen, 1901, and Strickland, 1965);

$$\text{Salinity} = 0.03 + 1.805(\text{Chlorinity})$$

Dissolved oxygen was determined by using Winkler's thiosulphate titration method with starch solution as an indicator (Strickland, 1965). And the hydrogen ion concentrations were measured with a Beckmann *p*H meter, Model G.

## RESULTS and DISCUSSION

Maximum, minimum and average values of the physical and chemical factors, such as air temper-

**Table I.** Physical and chemical factors of the estuary taken at the time of visit from May to September 1967.

| Factors | Maximum | Minimum | Average |
|---|---|---|---|
| Air temp.(°C.) | 31.0 | 16.0 | 25.35 |
| Water temp.(°C.) | 29.3 | 12.0 | 22.71 |
| *p*H | 8.1 | 7.32 | 7.63 |
| D.O. (mg./*l*) | 8.9 | 7.6 | 8.2 |
| Salinity (‰) | 25.69 | 0.06 | 8.073 |

-ature, water temperature, hydrogen ion concentration, dissolved oxygen and salinity, measured and analyzed at the sites and in the laboratory during this experimental period are shown in the Table I.

And the regional characteristics of each experimental site in the Han River estuary are shown respectively in the Table II.

**Table II.** The distances and average values of several environmental factors of each experimental site obtained from May to September 1967. The distances were measured from the base point of the old Han River Bridge following the stream.

| Name of Site | Distance (Km) | *p*H | D.O. (mg/*l*) | Salinity (‰) |
|---|---|---|---|---|
| Chollyu-ri | 34.3 | 7.62 | 8.2 | 0.116 |
| Wolgon-ri | 57.5 | 7.52 | 8.9 | 3.224 |
| Cholsan-ri | 64.8 | 7.56 | 8.3 | 5.224 |
| Chogi-ri | 74.0 | 7.68 | 8.0 | 10.183 |
| Inhwa-ri | 77.3 | 7.66 | 7.6 | 12.126 |
| Oepo-ri | 87.8 | 7.69 | 8.1 | 17.566 |

As shown in the Table I and Fig. 2, according as the distances from Seoul increased, the salinity increased. And the hydrogen ion concentration of each site were similar to those of Wood (1959) in the estuary of the Lake Macquarie.

The Table III shows that the average numbers of the general bacteria, coliform group and fungi collected at each site. In the case of coliform bacteria, the numbers of them showed decreasing intention from 165,000/ml. of Chollyu-ri to 27,000/ml. of Oepo-ri in the order of distance. However, it insists on the high pollution of Han River estuary, comparing with the number of Tokyo Bay, about 20,000/ml. (Nieta, 1901), and with the small number of the Lake Macquarie's estuary (Wood, 1959).

In the case of bacteria, from report of bacterial population which carried out with the surface water and close to the bottom water in Gulf of Manaar, India, its number was 100–850/ml. (Velankar, 1955).

In the Lake Macquarie, Wood(1955) described the number as 5 to 13,000, and it resulted from the inflow of nutrients into the lake by the inflows of fresh water from the land after heavy rains. He asserted that this was one of the main factors of microbial population in water.

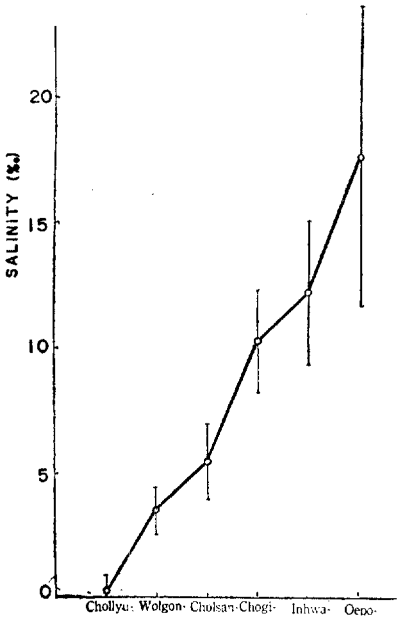


**Fig. 2.** Graph shows the salinity in each experimental site.

**Table III.** Numbers of microorganisms per ml. of water in Han River estuary.

| Microorganism / Locality | General bacteria | Coliform bacteria | Fungi |
|---|---|---|---|
| Chollyu-ri | 266,000 | 165,000 | 5,200 |
| Wolgon-ri | 146,000 | 140,000 | 2,300 |
| Cholsan-ri | 111,000 | 78,000 | 3,700 |
| Chogi-ri | 36,000 | 32,000 | 23,000 |
| Inwha-ri | 31,000 | 29,000 | 4,000 |
| Oepo-ri | 31,000 | 27,000 | 13,000 |

Otherwise, Oppenheimer(1960) set forth that the aerobic bacteria from the surface water of Texas Bay ranged from $5\times10^5$ to $5\times10^6$, and from $3\times10^5$ to $6.5\times10^6$ in the sediment of the Bay.

And in the study of Botany Bay and Port Hacking, two estuaries near Sydney, Australia, bacterial counts were obtained from $5\times10^6$ to $5\times10^7$ for the water at the surface and the tidal edge, and from $5\times10^7$ to $3\times10^9$ for sediments. There were little significant difference between the pollution of the exposed sediments at low tide and those covered by 3 feet of water (Wood, 1953).

The total numbers of the bacteria determined in this experiment were medium, comparing with those reported by the above investigators. However, the large amount of coliform bacteria of the Han River estuary resulted, we presume, from the influence of the inflow of foul water drained from Seoul and many of farm lands in the vicinity of the river, to which farmers usually manured a lot of excrements from men and/or animals. Most of the coliform bacteria contaminated from river water diminishes in the sea water from after about 4 weeks (Nieta, 1961).

But continuous flowing of the coliform bacteria from the Han River seems not to give rise to the natural decomposition of the coliform bacteria remarkably.

The relation of locality as to the distance of Han River water way to the salinity of each site was shown in the Fig. 2, and the relationship between the number of bacteria and salinity was illustrated in the Fig. 3. Salinity of the sea water usually marks 33 to 37‰, and 35‰ in open ocean. About 85% of contents of the salinity is consisted on

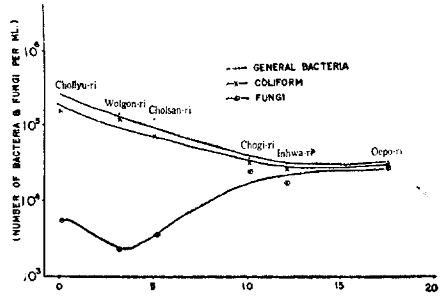


**Fig. 3.** Sites show the the changes of the microorganisms as the salinity increases.

sodium chloride. As indicated in the Table I, the maximum salinity in the Han River estuary during the period was 25.69‰ of Oepo-ri. This means that larger amount of salinity were there than that of average in the usual estuaries. It may be said the numbers of the bacteria decreases as the distance closes to the sea due to the dilution effect by the sea water.

However, the result of our investigation, in which we had inoculated the water sample from Chollyu-ri on the media having sodium chloride in different concentrations (Table IV and Fig. 4), showed that

**Table IV.** Growth index of general bacteria on nutrient broth agar with different concent rations of sodium chloride.

| Concentration of NaCl (%) | Number of Colonies per ml. | Survival Per cent |
|---|---|---|
| 0.00 | 116,500 | 100.0 |
| 0.25 | 71,000 | 60.9 |
| 0.50 | 67,000 | 57.5 |
| 0.75 | 60,000 | 51.5 |
| 1.00 | 55,000 | 47.2 |
| 2.00 | 18,500 | 15.7 |
| 3.00 | 16,000 | 13.7 |
| 4.00 | 13,000 | 11.2 |

the formation of colonies of the general bacteria diminished as the concentration of sodium chloride increased. The growth index of the bacteria from river water reported by Salle(1961) showed similar intention with that of our experiment indicated in the Table IV and in the Fig. 4. And by the specific

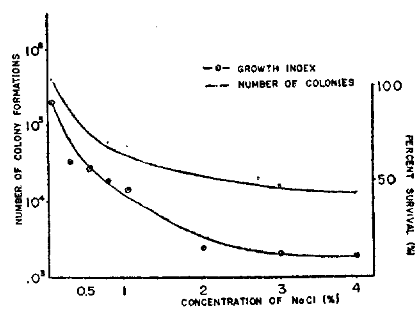


**Fig. 4.** Growth index of general bacteria on nutrient agar media which were added by different concentrations of sodium chloride.

salinity requirement the fresh-water bacteria distinguish from the marine ones (Korinek, 1927).

Summing the above results there were definite relationships between the decreasing in numbers of bacteria and increasing of salinity as shown in this experiment of the Han River estuary. It can be said the decrease in number of bacteria could occur with the increase in salinity of water in which they lived. Further study on the adaptation of the fresh-water bacteria to marine bacteria are interesting and needed.

Among the microorganisms identified from the estuarine water in this period of study the proteolytic bacteria such as *Escherichia coli*, *Bacillus subtilis* and *B. megaterium*, and the bacteria belonging to *Pseudomonadales* were predominant. And also we could find such bacteria as *Mycoplana* spp., *Aerobacter* spp., and *Shigella* spp., and such fungi as *Saccharomyces* spp., *Actinomyces* spp., *Penicillium* spp., *Aspergillus* spp. and *Rhizopus* spp.

The microbial flora of the water in Lake Macquarie examinated by Wood (1959) was found that proteolytic bacteria formed a greater percentage of the whole flora in the sediment. *Actinomycetes* and other fungi were very numerous in late summer. Coliform bacteria had not been found in uncontaminated estuarine water. Their presence indicated recent contamination (Wood, 1959).

Comparing with the above, the results of our examinations suggest the need of some sewage plants in the area of the Han River.

## CONCLUSION

In the Han River estuary the general bacterial counts ranged from 31,000 to 266,000, coliform group were 27,000 to 165,000, and fungi were distributed from 2,300 to 23,000 per milliliter of the water in the period of May-September 1967. These numbers marked very higher than that of international security limit in public health. Furthermore, coliform bacteria, which are indicators for the rate of water pollution, formed 81.6% of general bacteria. By these facts it can be said the water of the estuary was highly polluted. And it was also confirmed by that the proteolytic bacteria such as *Bacillus subtillis* and *B. megaterium* took the place of predominance in the microflora of the estuary. The high pollution of the estuary, it was assumed, resulted from the drained water from Seoul and its vicinity, thickly populated districts, and from the sewage water thrown by many industrial plants in the area of the down stream of the river. And since the dirty water was discharged into the sea without any purification processes before its inflow into the river, many of the water-closets in the residences of inhabitants were not constructed ideally, and also the excrements of men and animals were usually fertilized to the farm lands, it seemed possible for the water of the Han River estuary to show such high contamination.

As the water of the Han River progresses toward the sea, its salinity increased, and the fresh water became diluted, and according as the increasing salinity and dilution, the number of polluting microorganisms decreased. It seemed this decreasings of the bacterial counts due to mainly the week tolerance of the fresh-water bacteria in marine water and influence of dilution effect by the sea water partly.

The study of water pollution needs to examine the whole flora of heterotrophic, fresh-water and marine bacteria, to disclose the correlations between the flora and their environmental factors, especially the pollution rate, and to investigate the self-clarification of estuaries, through a long period. In this country continuous pursuit of this problems are firmly demanded in the future.

## 摘 要

한강하구의 오염미생물의 분포를 검토하기 위하여 전류리, 월곳리, 철산리, 초지리, 인화리, 외포리에서, 1967年 5月부터 9月까지 5개월간 매월 조사하였는바 그 결과는 다음과 같았다.

1) 서울의 제1한강교를 기점으로 한강수로를 따라 각 조사지점까지의 거리순으로 salinity의 증가를 보여주었으며 *p*H는 7.3~8.1로 전 조사기간을 통하여 각각 큰 차이를 보이지 않았다.

2) 일반세균과 대장균군은 전류리에서 가장 많았고, 외포리에 이르기까지 점감하였으며, salinity에 비교적 저항성이 큰 fungi는 초지리에서 가장 많았다.

3) Salinity와 general bacteina와의 관계는 salinity의 증가에 따라 growth rate가 감소함을 보았는데, 이는 fresh-water bacteria가 한강 하구에서 해수의 영향으로 점차 감소할 수 있음을 의미한다.

4) 한강수의 오염도는 서울과 경인공업단지의 인구증가, 공장폐수의 증가, 그리고 농경작지에 사용된 인분등의 영향을 크게 받음을 알 수 있다.

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