Serovar and Drug Sensitivity of Salmonella Isolated in Kenya

Akiyoshi UTSUNOMIYA

Department of Bacteriology, Institute for Tropical Medicine, Nagasaki University

Abstract: Thirty-three strains of Salmonella isolated in Kwale District, Coast Province, Kenya, were tested for their serovars and drug sensitivities to tetracycline, streptomycin, chloramphenicol, nalidixic acid and aminobenzyl-penicillin. Number of strains belonging to O-groups B, C 1, C 2, D 1, E 1, E 4, G 1, and O were 16, 1, 1, 3, 2, 1, 7 and 2 respectively. The serovars detected were 16 as shown in Table 1, and 6 strains of Sal. kiambu, 5 of Sal. goodwood and 4 of Sal. heidelberg were predominant. As shown in Table 2, the strains resistant to one and two drugs were only 3 and 1.

Key Words: Salmonella, Drug sensitivity, Diarrhea.

Under the "Kenya/Japan Communicable Diseases Research and Control Project" sponsored by Japan International Cooperation Agency (JICA), bacteriological investigations were carried out to clarify the etiology and ecology of diarrheal diseases in Kwale District, Coast Province, Kenya. From October 1981 to July 1982, a total of 999 fecal specimens were collected from outpatients who visited the medical facilities (one Subdistrict Hospital, two Health Centers and three Dispensaries) complaining of diarrhea, abdominal pains, tenesmus or vomiting, and were examined to isolate *Salmonella*, *Shigella*, enteropathogenic *Escherichia coli* (EPEC) and vibrios.

Serological examination of the suspectable organism was done at Kenya by using commercial diagnostic antisera (Denka Seiken, Japan) for O-antigens of Shigella, OKantigens of EPEC, O-antigens of Salmonella and O- or K-antigens of vibrios, but Hantigens of Salmonella could not be detected. Serological properties for 228 strains of Shigella, 80 strains of EPEC, and 35 strains of Salmonella and their drug sensitivity pattern were reported in previous paper (Utsunomiya et al., 1982). In this paper, the serovars of Salmonella detected in Japan and drug sensitivity for each of them will be recorded and discussed.

Received for publication, May 17, 1983.

Contribution No. 1,364 from the Institute for Tropical Medicine, Nagasaki University.

MATERIALS AND METHODS

Strains: Among the isolates stocked in the semi-solid butt of Nutrient Agar (Eiken) and brought back to Japan, 35 strains of *Salmonella* were attemted to use in this study. But two strains were excluded from this study because of their death.

Serological examination: The determination of serovars for 33 strains of Salmonella was cordially examined by Dr. Riichi SAKAZAKI, Japan Salmonella Center, National Institute of Health, Tokyo.

Drug sensitivity test: The sensitivity test for antibiotics was performed by the plate dilution method, and minimal inhibitory concentration (MIC) was defined as the lowest concentration of drugs detecting no growth after incubation for 24 hrs at 37°C. Antibiotics tested were tetracycline (TC), streptomycin (SM), chloramphenicol (CP), nalidixic acid (NA) and ampicillin (aminobenzyl-penicillin, AB-PC).

RESULTS

The results of serological examination are shown in Table 1 with character of the stools and other informations obtained. Regarding the group in Genus Salmonella based on O-antigen, the numbers of strains belonging to Group B, G 1, D 1, E 1 and O were 16 (48.4%), 7 (21.2%), 3 (9.0%), 2 (6.0%) and 2 respectively. Remaining were one strain each of Group C 1, C 2 and E 4. On the other hand, 16 serovars were detected among 33 strains tested. Arranging in order, 6 (18.1%), 5 (15.1%), 4 (12.1%) and 3 (9.0%) strains were Sal. kiambu, Sal. goodwood, Sal. heidelberg and Sal. are-chavaleta respectively. Each of 2 strains were Sal. daressalaam, Sal. poona and Sal. adelaide and remaining Sal. bredeney, Sal. senftenberg, Sal. sofia, Sal. typhimurium, Sal. braenderup, Sal. zanzibar, Sal. weltevreden, Sal. ndolo and Sal. mapo were one strain each.

So, 7 cases of mixed infection with *Salmonella* and other enteropathogens (Utsunomiya *et al.*, 1982) were finally confirmed as Table 2. Five cases were mixed infection with *Shigella*, and the Cases 25 and 33 had bloody and mucous diarrhea caused by *Shigella*. From Case 21, different O-group of *Salmonella* were found, but the isolate of Group O could not be examined in this investigation.

Table 3 shows the MICs of each Salmonella isolate detected. Sal. goodwood isolated from Case 31 was resistant to 100 mcg/ml of AB-PC and sensitive at 25 mcg/ml of TC. The MIC to AB-PC for Sal. goodwood isolated from Case 4 was 50 mcg/ml, the MIC to TC for Sal. bredeney from Case 5 was 100 mcg/ml and the MIC to NA for Sal. kiambu from Case 33 was 25 mcg/ml. While remaining 29 isolates were highly sensitive to all of the antibiotics tested. The commonest MICs were; 1.56 mcg/ml (30 strains,

Case No.	Location	Patients' Age & Sex		Character of the stool	Serovar	O-group
1	Tiwi	10	М	formed	Sal. daressalaam#	D 1
2	Kwale	A*	Μ	muddy	Sal. arechavaleta	В
3	Kwale	А	F	mucous	Sal. goodwood #	G 1
4	Matuga	1	Μ	mucous	Sal. goodwood 	G 1
5	Tiwi	7m**	F	formed	Sal. bredeney	В
6	Tiwi	8m	F	mucous	Sal. heidelberg	В
7	Tiwi	30	Μ	muddy	Sal. senftenberg	E 4
8	Matuga	28	F	muddy	Sal. goodwood#	G 1
10	Ngombeni	30	F	watery, muddy	Sal. sofia#	В
11	Ngombeni	2m	F	watery, muddy	Sal. typhimurium	В
12	Likoni	А	F	mucous, muddy	Sal. kiambu	В
13	Matuga	2	F	bloody, mucous	Sal. braenderup	C 1
14	Waa	7	F	watery	Sal. heidelberg	В
15	Tiwi	29	Μ	muddy	Sa!. arechavaleta	В
16	Likoni	26	F	bloody, mucous	Sal. kiambu	В
17	Kwale	17	F	muddy	Sal. arechavaleta	В
18	Ngombeni	25	F	watery	Sal. kiambu	В
19	Likoni	27	Μ	muddy	Sal. heidelberg	В
20	Waa	60	F	muddy	Sal. poona	G 1
21	Matuga	18	F	muddy	Sal. kiambu	В
23	Tiwi	9m	М	mucous	Sal. daressalaam#	D 1
24	Waa	11	Μ	muddy	Sal. heidelberg	В
25	Likoni	А	F	bloody, mucous	Sal. kiambu	В
26	Waa	12	F	muddy	Sal. adelaide	0
27	Waa	1	F	mucous	Sal. poona	G 1
28	Matuga	28	F	mucous	Sal. adelaide	0
29	Likoni	23	М	mucous	Sal. zanzibar	E 1
30	Kwale	25	F	muddy	Sal. weltevreden	E 1
31	Ngombeni	14	F	watery	Sal. goodwood#	G 1
32	Ngombeni	2	F	muddy	Sal. ndolo	D 1
33	Tiwi	47	F	bloody, mucous	Sal. kiambu	В
34	Likoni	30	Μ	mucous, muddy	Sal. mapo	C 2
35	Waa	1	F	muddy	Sal. goodwood#	G 1

Table 1. Serovars of *Salmonella* isolated from Kwale District, Coast Province, Kenya (October 1981-July 1982)

A*: adult, m**: month, *: subgenus II.

,

٩,

Case 9 (Group B) and Case 22 (Group O) could not be applied to this study.

Table 2. The	cases o	of mixed	infection	with	Salmonella
--------------	---------	----------	-----------	------	------------

Case 8: Sal. goodwood and Shigella flexneri 4
Case 15: Sal. arechavaleta and Shigella dysenteriae 2
Case 21: Sal. kiambu and Sal. Group O (can not be studied)
Case 25: Sal. kiambu and Shigella flexneri 1b
Case 27: Sal. poona and Shigella flexneri 6
Case 30: Sal. weltevreden, EPEC O86a:K61 and EPEC O144:Kx2
Case 33: Sal. kiambu and Shigella flexneri variant X

	Table 3. M	IC of Salmo	<i>nella</i> isolates	(mcg/ml*)		
No.	Serovar	TC	SM	СР	NA	AB-PC
1	Sal. daressalaam	1.56*	6.25	3.13	3.13	0.39
2	Sal. arechavaleta	1.56	6.25	3.13	3.13	0.39
3	Sal. goodwood	1.56	6.25	3.13	3.13	0.39
4	Sal. goodwood	1.56	6.25	3.13	3.13	50
5	Sal. bredeney	100	6.25	3.13	3.13	0.39
6	Sal. heidelberg	1.56	6.25	6.25	3.13	0.39
7	Sal. senftenberg	1.56	3.13	6.25	3.13	0.39
8	Sal. goodwood	1.56	12.5	1.56	1.56	0.39
10	Sal. sofia	1.56	6.25	3.13	3.13	0.39
11	Sal. typhimurium	1.56	6.25	3.13	3.13	0.39
12	Sal. kiambu	1.56	12.5	3.13	3.13	0.39
13	Sal. braenderup	1.56	12.5	3.13	3.13	0.39
14	Sal. heidelberg	1.56	12.5	3.13	3.13	0.39
15	Sal. arechavaleta	1.56	12.5	3.13	3.13	0.39
16	Sal. kiambu	1.56	12.5	3.13	3.13	0.39
17	Sal. arechavaleta	1.56	12.5	3.13	3.13	0.39
18	Sal. kiambu	1.56	12.5	3.13	3.13	0.39
19	Sal. heidelberg	1.56	12.5	3.13	3.13	0.39
20	Sal. poona	1.56	12.5	3.13	3.13	0.39
21	Sal. kiambu	1.56	6.25	3.13	3.13	0.39
23	Sal. daressalaam	1.56	6.25	3.13	3.13	0.39
24	Sal. heidelberg	1.56	6.25	3.13	3.13	0.39
25	Sal. kiambu	1.56	6.25	3.13	3.13	0.39
26	Sal. adelaide	1.56	6.25	3.13	3.13	0.39
27	Sal. poona	1.56	6.25	3.13	3.13	0.39
28	Sal. adelaide	1.56	6.25	3.13	3.13	0.39
29	Sal. zanzibar	1.56	12.5	3.13	3.13	0.39
30	Sal. weltevreden	0.78	3.13	3.13	3.13	0.39
31	Sal. goodwood	25	12.5	1.56	3.13	$100 \angle$
32	Sal. ndolo	1.56	12.5	3.13	3.13	0.39
33	Sal. kiambu	1.56	12.5	3.13	25	0.39
34	Sal. mapo	1.56	12.5	3.13	3.13	0.39
35	Sal. goodwood	1.56	6.25	3.13	3.13	0.39

ŧ

Table 3. MIC of Salmonella isolates (mcg/ml*)

TC: Tetracycline, SM: Streptomycin, CP: Chloramphenicol, NA: Nalidixic acid, AB-PC: Aminobenzyl-penicillin (Ampicillin)

90.9%) to TC, 6.25 mcg/ml (16 strains, 48.4%) and 12.5 mcg/ml (15 strains, 45.4%) to SM, 3.13 mcg/ml (29 strains, 87.8%) to CP, 3.13 mcg/ml (31 strains, 93.9%) to NA and 0.39 mcg/ml (31 strains, 93.9%) to AB-PC.

DISCUSSION

In this study, 33 Salmonella strains isolated from Kenyan residents living in rural and lowland area of Kenya were classified to 16 serovars. Excepting Sal. daressalaam, Sal. goodwood and Sal. sofia who belong to subgenus II, 13 serovars are subgenus I, and Sal. kiambu, Sal. goodwood and Sal. heidelberg were predominant serovars in order. It can not be found the relation between the serovar and the location or the other information. Sal. typhi, Sal. paratyphi A, Sal. schottmuelleri (Sal. paratyphi B) and Sal. hirschfeldii (Sal. paratyphi C) were not found in this investigation. In another African population, Bockemühl (1977) reported the Salmonella infection rates from the urban population of Lomé, capital of Togo, West Africa from 1971 to 1973. From patients suffering from intestinal diseases and healthy populations, 12,166 stool specimens were examined. The isolates of Salmonella were 412 (3.4%) and classified to 115 serovars. Sal. agona (35 strains, 8.4%), Sal. agama (26 strains, 6.3%), Sal. typhimurium, Sal. virchow, Sal. elisabethville (20 strains each, 4.8%) were the predominant serovars, and the predominant serovars in Kenya stated above were not found in Togo. Regarding the same serovars as isolated in Kenya, Sal. braenderup (8 strains), Sal. bredeney (7 strains), Sal. adelaide (3 strains), Sal. senftenberg and Sal. poona (one each strain) were isolated. On the other hand, Zen-yohji (1978) reported that Sal. typhimurium (13.0%), Sal. infantis (7.3%), Sal. thompson (5.6%), Sal. schwalzengrund (4.3%), Sal. panama and Sal. senftenberg (4.2% each) were the predominant serovars among 3,801 Salmonella-strains isolated from Japanese domestic residents during the period between 1966 and 1977. There is big difference in predominant serovars between Kenya and Japan. In Japan, Sal. kiambu, the most predominant serovar in Kenya, was found in a report (Abe et al., 1981). One strain was isolated from traveller's diarrhea at Osaka Airport Quarantine Station in 1979, but the suspected regions for infection was not stated.

Regarding the drug sensitivities of isolates, 3 strains (Sal. goodwood-Case 4, Sal. bredeney-Case 5 and Sal. kiambu-Case 33) were resistant to one drug, and 1 strain (Sal. goodwood-Case 31) was resistant to 2 drugs. But remaining 29 strains were highly sensitive to 5 antibiotics tested. Although the most of diarrheal patients have been treated with TC in Kwale District, the incidence of highly resistant Salmonella to TC was very low. In contrast, 763 (20.1%) out of 3,801 Salmonella-strains isolated from Japanese residents were resistant to antibiotics with various patterns. In the case of Sal. typhimurium, 40% were resistant to SM and TC, and 10.9% were resistant to SM, TC and kanamycin (KM). Also 58.7% of Sal. panama were resistant to TC and KM

(Zen-yohji, 1978). According to the traveller's cases in Japan (Abe *et al.*, 1981), 15 strains out of 145 were resistant (more than 25 mcg/ml) to antibiotics. Two strains of *Sal. krefeld* were showed multiple resistance to SM, CP, TC, KM and AB-PC. One strain (*Sal. anatum*) was resistant to SM, CP, TC and AB-PC. Five strains (two strains of *Sal. indiana*, and one strain of *Sal. panama*, *Sal. agona* and *Sal. kiambu*) were resistant to SM and TC. Also 3 strains (*Sal. stanley*, *Sal. anatum* and *Sal. haifa*) were resistant to TC, and 4 strains (*Sal. london*, *Sal. derby*, *Sal. worthington* and *Sal. blockley*) were resistant to SM. Drug sensitivity test of isolates from Togo (Bockemühl, 1977) was not done.

Finally the author likes to add here that *Sal. kiambu*, the most predominant serovar in this study, was firstly isolated in 1951 from children dead by lung tuberculosis who lived in Kiambu (Kelterborn, 1967) which is a neighboring town of Nairobi, Kenya.

ACKNOWLEDGEMENTS

The author wishes to thank Prof. Tatsuro NAITO, Department of Bacteriology, Institute for Tropical Medicine, Nagasaki University, for his advice and reviewing the manuscript. The author is also grateful to Dr. Riichi SAKAZAKI, National Institute of Health, Tokyo, for his help in identify the serovars of isolates.

REFERENCES

- Abe, H., Kanda, T., Yanai, Y., Hashimoto, S., Ogawa, R., Miyata, Y., Shinbo, M., Harada, K., Tsukamoto, T., Kinoshita, Y., Arita, M., Takeda, Y. & Miwatani, T. (1981): Bacteriological study of traveller's diarrhoea (1) Isolation of enteric bacteria from patients with traveller's diarrhoea at Osaka Airport Quarantine Station during 1979. J. J. A. Inf. D., 55 (10), 679-690. (in Japanese with English abstract)
- Bockemühl, J. (1977): Salmonellosis and shigellosis in Togo (West Africa), 1971-1973 II. Infections in the urban population of Lomé. Tropenmed. Parasit., 28, 377-383.
- Kelterborn, E. (1967): Salmonella-species, Erstfunde, Namen und Vorkommen. p. 211.
 S. Hirzel Verlag Leipzig., cited from Lapage, S.P., Taylor, J., Nicewonger, C.R. & Phillips, A.G. (1966): New serotypes of *Salmonella* identified before 1964 at the Salmonella Reference Laboratory, Colindale. Int. J. system. Bact., 16, 253-297.
- 4) Utsunomiya, A., Mori, K., Hayashi, T., Iwanaga, M., Naito, T., Gatheru, Z.W., Sang, F., Koske, N. & Kaviti, J.N. (1982): Bacteriological study on the diarrhoeal diseases in Kwale District, Coast Province, Kenya. Trop. Med., 24 (4), 235-252. (in Japanese with English abstract)
- 5) Zen-yohji, H. (1978): Salmonellosis. J. J. A. Inf. D., 52 (4), 95-97. (in Japanese)

ケニアで分離されたサルモネラ属菌の血清型と薬剤感受性

宇都宮明剛(長崎大学熱帯医学研究所病原細菌学部門)

1981年10月から1982年7月までの間、ケニア国南部海岸地区で下痢症の原因細菌の調査を行 い、999の下痢便検体から赤痢菌228株,病原大腸菌80株と共に、サルモネラ属菌35株を分離した (宇都宮ら,1982). 本報ではサルモネラ属菌の33株について、その血清型と分離株個々の薬剤 感受性値を記述した.血清型別の結果、8種類のO群と16種類の血清型がみられ、Sal. kiambu が最も多く6株(18.1%),次いでSal. goodwood 5株(15.1%),Sal. heidelberg 4株(12.1 %),Sal. arechavaleta 3株(9.0%)で、Sal. typhimurium は1株であった.またチフス 菌、パラチフス菌は検出されなかった.薬剤感受性試験の結果では、100 mcg/ml 以上の耐性を 示したものはアンピシリンに対する1例のみで、多剤耐性株も見出されなかった、1966年-1976 年の日本国内分離株、および1979年内に大阪空港検疫所で調査され、旅行者下痢患者から分離さ れた薬剤耐性サルモネラ属菌の耐性頻度と比較して、ケニア住民から分離されたサルモネラ属菌 は、極めて高い感受性を示したといえる。

熱帯医学 第25巻 第2号, 65-71頁, 1983年6月

Erratum

In "Bacteriological Study on the Diarrhoeal Diseases in Kwale District, Coast Province, Kenya" by Utsunomiya, A. *et al.* (Tropical Medicine, Vol., 24, No. 4, pp. 235-252, 1982), on the part of Salmonella in Table 14, NA should be AB-PC and AB-PC should be NA.