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Otitis externa by *Pseudomonas aeruginosa* associated with whirlpools

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SUMMARY

Over a period of about 1 year, otitis externa occurred in at least 300 visitors to a recreational park. The infections were associated with the presence of *Pseudomonas aeruginosa* in insufficiently chlorinated whirlpools.

INTRODUCTION

Pseudomonas aeruginosa infections, notably skin rash and otitis externa, associated with the use of inadequately chlorinated swimming pools have been reported with increasing frequency. Whirlpools or other small-sized swimming pools have been commonly incriminated as sources of infection (Hopkins, Abbot & Wallace, 1981; Ford-Jones *et al.* 1981), but larger swimming pools have been involved also (Reid & Porter, 1981; Seyfried & Fraser, 1978). The number of affected bathers was generally small, ranging from 2 to 75.

This paper describes an outbreak that affected at least 300 visitors to a recreational park over a period of about 1 year. The recreational park was opened to the public in July, 1980. Shortly afterwards patients with ear complaints which were diagnosed as otitis externa, were often examined during regular consulting hours at the park. The number of patients examined quickly rose to 100 in July and 83 in August. It is possible that many more infections occurred among people who did not seek medical attention or waited to do so until after their return home (most visits to the park were short). In this period, ear-swabs were taken from 14 patients and cultured by the Regional Public Health Laboratory at Hilversum. All cultures yielded fluorescent pseudomonas. Otosporin® (polymyxin B sulphate 10000 E ml^{-1}; neomycin sulphate 5 mg ml^{-1}; hydrocortisone 10 mg ml^{-1}; 2 drops

four times daily for 1 week) was prescribed and all the patients recovered uneventfully.

The high number of otitis externa cases was attributed to initial operating problems with the rather complicated equipment used for treating the water in the various swimming pools. Following technical measures in September the number of cases dropped steadily reaching zero in January, 1981. However, in the following months new patients were seen which prompted us to perform a more detailed investigation.

MATERIALS AND METHODS

The swimming pool complex

The swimming pool complex is part of a large recreational park where approximately 3000 visitors can be accommodated in 600 bungalows. During their stay, lasting usually between 3 days and 2 weeks, visitors can use the pools free. The complex consists of a large hall where several pools are situated: a large swimming pool in which artificial waves can be generated (including a childrens' bath) and a 'bubble-bath' with strong aeration. Next to these pools are a sauna, a solarium and a gymnasium. Pot plants and other decorations are situated throughout the hall. An outdoor swimming pool is connected to the hall by a sluice. Two whirlpools are also situated outdoors. Water temperatures in the pools are within the range of 25–30 °C, except for the whirlpools. Whirlpool I is maintained at 30–32 °C and whirlpool II at 35–37 °C.

Water treatment in the pools is largely automated. The water from the large indoor pool (900 m^3, turnover $2\frac{1}{4}$ h) is treated by coagulation, followed by dual-layer filtration (hydro-anthracite/sand). Disinfection is obtained by dosing with sodium hypochlorite solution and pH-control is by hydrochloric acid. Both additions are automatically controlled by measuring free residual chlorine (DPD) and pH, respectively. Redox potential is also monitored continuously. About 10 % of the return-flow from the pool is treated prior to coagulation/filtration by a Chloramine-separator®, intended to reduce combined residual chlorine by irradiation (infrared and ultraviolet).

Water from the outdoor pool (150 m^3, turnover $1\frac{1}{2}$ h) is treated similarly but the addition of chlorine is controlled by redox-measurement and there is no treatment by Chloramine-separator®. The bubble-bath (60 m^3, turnover 1 h) is circulated with treated water from the large pool. The water in the whirlpools (2 m^3 each) is filtered separately through sand filters (turnover $\frac{1}{4}$ h) and continuously exchanged with water from the outdoor pool (4 m^3/hour/pool). Disinfection was initially performed by means of slow dissolving chlorine tablets in the inflowing water, occasionally supplemented with hand-addition of calcium hypochlorite powder. At a later stage, pumps for the continuous addition of a sodium hypochlorite solution were installed. These pumps add a constant amount of hypochlorite solution from 10 a.m. to 10 p.m., and are connected to the blower-controls in such a way that more chlorine is added when the blowers are in use.

The bather load was reasonably constant throughout the year and was estimated to range from 2000 to 3000 persons a day for the whole complex.

Analysis of pool water

Samples were taken at the outlet side of the larger pools, and in the centre of the whirlpools. During sampling, temperature, free and combined residual chlorine (DPD-method, back-titration with ferrous-ammonium sulphate), pH and redox-potential were measured directly. Further chemical analysis (permanganate-value, ammonia, total nitrogen, turbidity and bicarbonate) was performed in the laboratory using standard analytical methods. Samples for bacteriological analysis were neutralized immediately with sodium thiosulphate and the analysis commenced within 15 min. Two 100 ml volumes were filtered through two Gelman GN6 membrane filters (47 mm, pore size 0·45 μm) which were placed on two selective media: 0·4 % enriched Teepol-agar (TA) (Report, 1969) for enumeration of total coliforms and mPA-B agar (Dutka & Kwan, 1977) for enumeration of *Pseudomonas aeruginosa*. The agar plates were kept at ambient temperature (20–30 °C) for 3–5 h for resuscitation and then placed in an incubator (TA at 37 °C for 1 day and mPA-B at 41·5 °C for 2 days). After incubation, typical colonies (yellow on TA; brown to black on mPA-B) were counted. Typical colonies on mPA-B were subcultured onto milk agar (MA, Brown & Scott Foster, 1970) for confirmation. Growth on MA-plates was recorded after incubation at 41·5 °C for 1 day. Plates showing growth were examined for pigment production and casein hydrolysis after an additional overnight incubation at room temperature. Total colony counts at 37 °C were also made using the pour plate technique with Plate Count Agar (Oxoid CM325) incubated at 37 °C for 2 days.

Environmental swab samples

Cotton swabs taken at various places in the complex, were refrigerated and transported to the laboratory, and they were cultured within one day. Swabs were streaked on mPA-B agar (41·5 °C for 2 days) and subsequently transferred to asparagin-broth (AB) (Drake, 1966) which was incubated at 37 °C for 2 days. All cultures showing growth were then streaked onto mPA-B and treated as described for the analysis of pool water.

Ear swab samples

Starting May 1981, whenever possible, each patient with a diagnosis of otitis externa completed a questionnaire and an ear-swab was taken. The earswabs were placed in Stuart medium and transported to the laboratory by mail. Swabs were streaked on 7 % sheep blood-agar and McConkey-agar (37 °C for 1 day) and mPA-B agar (41·5 °C for 2 days). *Ps. aeruginosa*-like colonies on either media were subcultured on MA, as described.

Typing of Pseudomonas aeruginosa

Pure cultures which grew on MA and hydrolysed casein and exhibited pigment production in most cases, were considered to be *Ps. aeruginosa* provided that the following biochemical characteristics were found: oxidase and catalase positive, Gram-negative rod, oxidative metabolism of glucose, and positive reactions for arginine-dihydrolase, utilization of malonate as sole C-source, splitting of gluconate and reduction of nitrate to nitrogen.

Each strain was typed routinely by four techniques. Our typing methods differ from standard practice as follows:

(1) *Serological typing* (Habs, 1957). The type-strains were those received from Prof. dr. H. Habs (Bonn), from the Central Public Health Laboratory, Colindale (London) and from dr. T H. Siem (Amsterdam). Typing is performed in Microtiter plates under four different conditions at the same time: the antigens are grown at 30 and 37 °C; serum is used at 4× and 8× the homologous titer. Sixteen serotypes were recognized: L(ányi 11), (2) A, (2) B, (5) C, (5) D and 1, 3, 4, 6–13.

(2) *Active pyocin typing* (pyocin production procedure; Gillies & Govan, 1966; Govan, 1978).

(3) *Passive pyocin typing* (pyocin sensitivity pattern; Osman, 1965). The indicator pyocines were prepared with the aid of Mitomycin C from the 8 Gillies strains obtained from dr. J. S. Kuipers (Groningen). After application of the indicator pyocines incubation was at 24 °C (not 37 °C).

(4) *Bacteriophage typing*. Nineteen bacteriophages were obtained from the Central Public Health Laboratory, London, in 1970, one bacteriophage was sent by Prof. J. Beumer, Brussels. They were diluted by steps of $\sqrt{10}$ (= 3·16).

RESULTS

The total number of patients examined per month during the outbreak, which lasted more than a year, is given in Table 1. A more detailed description of the distribution of cases with time since March 1981 is shown in Fig. 1. This shows that there are clusters of cases with time.

Clinical data on 31 patients is summarized in Table 2. Three patients were thought to have otitis media in addition to otitis externa. Most patients had been swimming in all the pools within the complex and had their first swim shortly after arrival at the park. The interval between arrival at the park and the onset of symptoms was usually 5–7 days (17×), but shorter periods of 1–2 days were also noted (7×). One patient was already ill upon arrival at the park, ear complaints having started during a stay at a similar recreational park in another part of the country. Most patients were between 10 and 20 years of age, and 13 had a previous history of ear-complaints. Of these, only 5 were considered to have a predisposition for otitis externa.

From these 31 patients, 28 ear-swabs were obtained and 23 yielded *Ps. aeruginosa*, usually in pure culture on blood-agar plates. This bacterium was also frequently isolated from swimming water and environmental swab samples (Table 3). In samples from the whirlpools, *Ps. aeruginosa* was present in 7 out of 20 samples, the maximum number of colonies found was 350 per 100 ml. Two of the four negative samples taken on July 16 were taken after the pools had been closed to the public for 3 days – as a consequence of the high incidence of cases in the first half of July – and the other two were taken 1 h after the pools had been reopened. Other pools were also contaminated with *Ps. aeruginosa*, but with a lower frequency and in lower numbers than the whirlpools. Swabs samples from the environment were frequently positive. On May 6, all 19 wet surfaces sampled yielded *Ps. aeruginosa*. These included plant pots, wooden and synthetic parts of the constructions, and floor-tiles adjacent to the pools. Although cleaning and

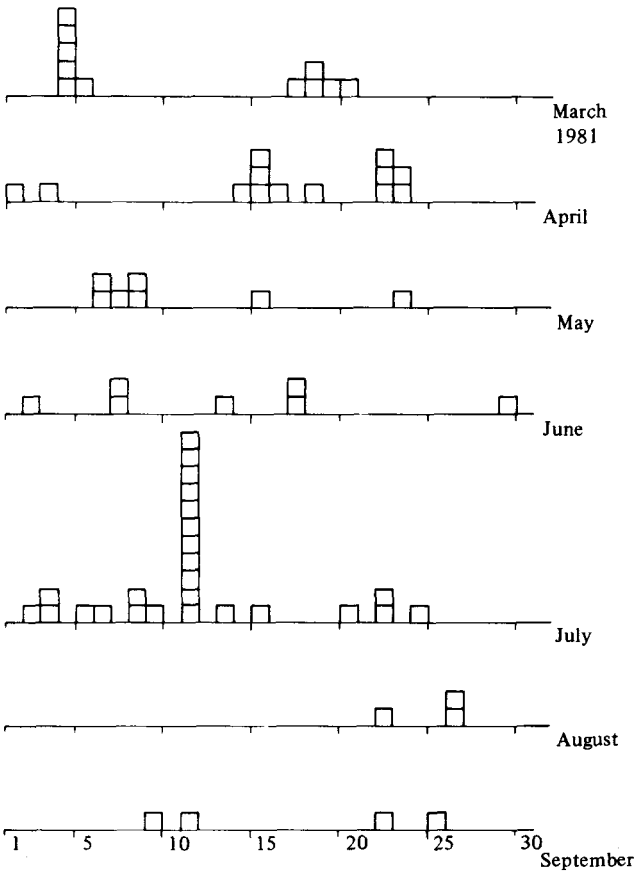

Fig. 1. Number of patients with otitis externa by day of visit to physician

Table 1. *Otitis externa among visitors of a recreational park*

Year and month	Number of cases
1980 – July	100
August	83
September	13
October	6
November	5
December	2
1981 – January	0
February	3
March	11
April	14
May	8
June	7
July	25
August	3
September	4
Total	284

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Table 2. *Clinical data from* 31 *patients with otitis externa*

Complaints and symptoms (by questionnaire)		Signs (by physician)	
Ear-ache	31	Auditory canal:	
(two sided)	6	Reddish	30
Pain upon chewing	20	Moist	21
Clogged nose	9	Secretion	8
Cold	6	Scaly	4
Fever	3	Aberrant tympanum	7

Table 3. *Isolation of* Pseudomonas aeruginosa *from a swimming pool complex*

Date (1981)	Whirlpools	Other pools	Environment
6 May	2/4*	2/9†	19/23
15 June	3/4	1/4	2/10
16 July	0/4‡	0/5	1/10
12 August	2/4	0/6	7/7
29 September	0/4	0/5	11/19
Total	7/20	3/29	40/69

* No. of positive samples/no. of samples analysed (100 ml).
† Including two non-chlorinated pools yielding *Ps. aeruginosa*.
‡ Whirlpools not in regular use.

disinfection of the complex were intensified *Ps. aeruginosa* was still isolated from swab samples on all the sampling days although with a varying frequency.

In order to study the possible relationships between the strains of *Ps. aeruginosa* isolated from the various sources, typing of pure cultures was carried out. (Table 4; this table includes data on 4 strains isolated in March–April 1981 by the Regional Public Health Laboratory at Hilversum). Among the strains from patients, serotype 11 and seriologically non-typable strains predominated. By pyocin typing and phage typing it could be demonstrated that in the course of time many different types were found, but certain types (designated as NT_1, NT_2 and 11_1, see also Table 5) were found among patients more than once. In addition type NT_1 was isolated on one occasion from a whirlpool and a swimming-belt; type 11_1 was found on a wooden bridge situated over the major swimming pool. Samples from this bridge always yielded *Ps. aeruginosa* and different types were isolated from the same sample as shown on August 12. *Ps. aeruginosa* types isolated from the bridge were also found in the sauna-bath (8_1), on floor-tiles (1_1), in whirlpools (6_1) and from one patient (10_1).

Chemical, physical and bacteriological analysis of the water revealed various defects in the treatment. In general, the amount of organic matter was very high in all pools (permanganate value was between 10 and 25 mg l^{-1}; Kjeldahl-nitrogen between 1 and 7 mg l^{-1}). Free chlorine levels were frequently below the desired level of 0·5 mg l^{-1}, especially in the whirlpools. In relation to the high level of organic matter, combined chlorine levels were also high, usually between 1 and 2 mg l^{-1}. In the course of 1981, more attention was paid to the treatment of the

Table 4. *Typing of* Pseudomonas aeruginosa *strains from patients, swimming pools and surfaces* (1981)

Patients		Pool water		Surfaces	
Date of first symptoms	Type*	Date and pool	Type	Date and location	Type
20 March	NT	06 May		06 May	
16 April	NT_1	Whirlpool II	NT_1	Window-frame	NT
23 April	NT_1	Whirlpool II	NT	Swimming-belt	NT_1
23 April	1	Saunabath	8_1	Wooden bridge	8_1
02 June	NT_2	Kneipp-bath	8_1	Plantpot	1
02 June	NT_2				
14 June	11_1	15 June		15 June	
15 June	11_1	Whirlpool I	6_2	Wooden bridge	NT
26 June	NT	Whirlpool II	6_2		
30 June	NT_1	Outdoor pool	6_2		
01 July	11				
01 July	NT_2	12 August		12 August	
07 July	6_1	Whirlpool I	6_3	Floor-tiles	C
08 July	NT_2	Whirlpool I	6_4	Floor-tiles	1_1
09 July	11	Whirlpool II	6_3	Floor-tiles	6
10 July	6_1	Whirlpool II	6_4	Floor-tiles	NT
12 July	11_1			Wooden bridge	6
22 July	10_1			Wooden bridge	10_1
04 August	3			Wooden bridge	11_1
12 August	NT_1			Wooden bridge	6_3
13 August	NT_1			Wooden bridge	1_1
20 August	6				
21 August	1				
20 September	11_1				
23 September	NT				

* Number: serological type (0-antigen), NT = serologically non-typable.
$_1$, $_2$ etc.: code for identical pyocin- and phagetype (if no subscript is added, the type was unique).

Table 5. *Typing results of strains common to patients and pool water or environment*

Serological type and code	Number of strains	Pyocin type		Bacteriophage type†
		active	passive	
NT_1	8	12D	48A	21, 119X
		12D	41A	21, 119X, 352
10_1	2	15D	NT	16, 21, 68, 119X
		15D	54C	16
11_1	5	12D	57C	16, 21, 119X
		12D	NT	21, 119X

* Code modified after Farmer (1970):
1: + + +, 2: + + −, 3: + − −, 4: − − −, 5: − + −, 6: − + +, 7: − − +, 8: + − +
A: + +, B: + −, C: − −, D: − +, NT = 44C.
† Strong reactions only.

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Table 6. *Incidence of* Pseudomonas aeruginosa *in relation to free chlorine content*

Free chlorine mg l^{-1}	*Ps aeruginosa* in 100 ml	
	present	absent
< 0·10	7 (88 %)	5
0·10–0·29	3 (30 %)	7
⩾ 0·30	0 (0 %)	25

water and as a result the water quality with regard to the above parameters improved markedly in all pools during the investigations.

As can be seen from Table 6, *Ps. aeruginosa* was only found in samples with free chlorine below 0·30 mg l^{-1} (pH was usually between 7·5 and 7·8). In all but one of the samples positive for *Ps. aeruginosa*, coliforms were detected and the colony count at 37 °C was high (100–5000 ml^{-1}).

DISCUSSION

The high incidence of otitis externa cases among visitors to the recreational park may have been related to the fact that the visitors spent a long time in a warm, humid environment for several consecutive days. This may have made the ear more susceptible to infection, particularly because most visitors frequently spent some time outdoors. According to this hypothesis, a more or less constant number of patients might be expected throughout the year. As can be seen from Fig. 1, however, there was a marked clustering of cases in time. From this, it may be concluded that other factors must have been involved in the establishment of the infections.

It is suggested that the infections are related to the frequent presence of *Ps. aeruginosa* in the bathing water, which was associated with inadequate chlorination. The available evidence pointed mainly towards the whirlpools as sources of infection, especially in 1981. Originally, the water in these pools was chlorinated by slowly dissolving chlorine tablets positioned in the inlet pipes. These tablets did not liberate sufficient chlorine to guarantee a satisfactory level under all conditions. Furthermore, the tablets were not always renewed at the proper time, especially in the first half of 1981. Consequently, the whirlpools contained low levels of free chlorine on various occasions, and better chlorination equipment was recommended. Automatic pumps dosing a sodium hypochlorite-solution were installed in the beginning of July and it was about a week before these operated properly. In the first two weeks of July many otitis externa cases were seen.

Since September 1981, no cases of otitis externa have been seen. This coincided with the moment that microbiological and chemical analysis no longer revealed treatment problems. This also strongly suggests an association between insufficient water treatment and cases of otitis externa.

It is probable that there was a constant input of bacteria from the environment of the pools into the water. From the typing data, it appeared as if the bridge over the major pool (constructed in such a way that effective cleaning is extremely

difficult) was one of the permanent reservoirs of *Ps. aeruginosa* in the complex. When insufficient free chlorine is present, it is possible that the bacteria may survive or even multiply in the swimming water.

If the bathing water was an important source of infection, the isolation of similar types of *Ps. aeruginosa* from patients and from external sources would be expected. To a certain extent this indeed was found. However, the data on typing should be interpreted with care. The strains were collected over a long period of time, and those from pools only on certain days. On other days, or other times of the same day, different types may have been present in the pool water. Therefore a great number of different types may be found. From this point of view, the isolation of strains common to patients and pool water or pool environment may be considered as an additional argument for the incrimination of the swimming pools as sources of infection.

The maintenance of a sufficient level of free chlorine was a particular problem in whirlpools, where the bather loads were extremely high (usually 8–12 bathers were present in only 2 m^3 of water). Chlorine was further reduced by strong aeration. Additionally, a high water temperature increased the rate of chlorine consumption and also increased the load of organic matter per bather (Althaus & Pacik, 1981).

These investigations do not indicate that the quality criteria for swimming water presently applied in the Netherlands should be tightened. *Ps. aeruginosa* was never found in samples where the free chlorine level was above 0·5 mg l^{-1} (the present guideline) and in all but one of the positive samples, the bacteriological criteria (absence of coliforms in 50 ml, colony count at 37 °C less than 100 ml^{-1}) were not met. The problem clearly is related to the need to maintain good quality at all times. This was only obtained by the proper attention of adequately trained personnel.

Sampling and chemical analyses were performed by the Laboratory for Environmental Chemistry, National Institute for Public Health (Dr A. W. Fonds, Dr H. R. F. Reijnders). Bacteriological analyses were carried out by Ms W. M. Hogeboom and Mr M. During; typing of *Ps. aeruginosa* strains were performed by Mrs J. H. L. De Jong. The authors gratefully acknowledge Dr J. Rechsteiner who provided information and strains isolated by the Regional Public Health Laboratory, Hilversum. They thank Dr H. F. Jansma and Dr H. W. B. Engel for helpful discussions and Dr J. S. Colbourne for correcting the English manuscript.

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