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FATE OF THE MICROORGANISMS DISCHARGED THROUGH SUBMARINE OUTFALL II. DISAPPEARANCE OF PATHOGENIC MICROORGANISMS

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SUMMARY.

The objective of the present study was to determine the disappearance in the marine environment of the pathogenic microorganisms most frecuently detected from sewage discharged through submarine outfall. The results obtained indicate that the Pearson technique (1) is not the most suitable one for inactivation process studies of pathogenic microorganisms.

TEXT.

Microbial survival studies of pathogens in the marine environment are a useful way to know the confidence degree that the indicator microorganisms of fecal pollution give.

The sampling zone is located in the marine area affected by sewage discharges from a submarine outfall, situated between Punta de Calaburras and Fuengirola harbour (Málaga-Spain). The pathogenic microorganisms techniques and culture media used are: Staphylococcus aureus, membrane filtration (2) and BFR-O (3) as enumeration medium. Pseudomonas aeruginosa, membrane filtration (2), and mPA (4). Candida albicans, membrane filtration (2) and mCA (5). Salmonella sp, multiple tubes and MPN method; the NR10(10) (6) is used as an enrichment broth.

The T90 values were obtained from the linear correlation equations of the concentrations of the pathogenic microorganisms studied and the tracking-times (7). The T90 in all values obtained appear in Table 1, together with the T90 significative values for each experiment.

The "in situ" inactivation study of the pathogenic microorganisms using Pearson technique has the following problems: (A) pathogenic microorganisms concentrations detected in seawater are much lower than those for indicators and therefore one must not use the same tracking-times. (B) If all values equal to 0/100 ml are included to make the linear regression, one obtains overrated T90 values.

It can be concluded that the time intervals used in the present study are not the most suitable in order to aknowledge survival of pathogenic microorganisms in seawater; we propose the following ones: 0, 5, 10, 15 and 30 minutes.

A more serious control over Staphylococcus aureus is recomended, as it presents a high resistance to autodepurating factors in the marine environment.

The use of Pearson technique (1) is not considered as the most suitable for the study of microorganisms inactivation, due to their rapid disappearance because of dilution, wich makes their tracking very difficult.

More trustworthy results about the inactivation of microorganisms in the marine environment, can be obtained by using diffusion chambers or laboratory experiments.

TABLE 1. T90 values of pathogenic microorganisms for each experiment.

	T90 values (minutes)				
Microorganisms	**Exp.1**	**Exp.2**	**Exp.3**	**Exp.4**	**T90 in all**
Staphylococcus aureus	14.9	33.8	17.06	66.65	30.48
Pseudomonas aeruginosa	6.75	14.9	3.61	15.38	11.36
Candida albicans	-	34.9	7.53	7.14	22.72
Salmonella sp	6.15	12.5	6.49	9.79	8.02

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MICROBIAL STANDARDS OF WATER POLLUTION AND HEALTH HAZARDS ASSOCIATED WITH THE CONCENTRATION OF PSEUDOMONAS AERUGINOSA IN BEACHES

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SUMMARY.

The frequency of *Pseudomonas aeruginosa* hazardous concentrations for swimmers in beaches are closely related with the levels of the indicator microorganisms of fecal pollution (Total Coliforms, Fecal Coliforms and Fecal Streptococci). The potential health hazard of infective concentrations of *P. aeruginosa* is lower than 15% in beaches that fulfil the following sanitary standards: TC50 $<$1000 cfu/100 ml and FC50 and FS50 $<$ 100-200 cfu/100 ml.

TEXT.

The major source of *Pseudomonas aeruginosa* in natural surface waters is domestic sewage (1,2,3). Its isolation in these waters is associated with the degree of fecal pollution (2,3). The hazard of *P. aeruginosa* ear infections associated directly with swimming is what gives special importance to the study of this bacteria in recreational waters (2,4). Thus, Hoadley proposed that when the level of *P. aeruginosa* in swimming waters is above 100 cfu/100 ml, swimmers are exposed to infective doses for ear and eye infections (2).

The seawater samples were obtained from 15 stations located in beaches near Málaga (Spain). The microbial counts were made by the membrane filtration method (5) employing m-Endo broth, m-FC broth, m-Enterococcus agar (5) and mPA-D agar (3) for TC, FC, FS and *P. aeruginosa*, respectively.

The frequency of *P. aeruginosa* hazardous concentrations for swimmers in beaches are closely related with its levels of fecal pollution. A significative correlation is observed ($p < 0.001$) between the logarithm of median values of microbiological indicators and the probability of *P. aeruginosa* infective concentrations in studied beaches. The highest infection hazards by *P. aeruginosa* are associated with the beaches with the most deficient health quality (table 1a). In beaches fulfiling the most usual standard values of health quality in swimming waters (TC50 $<$ 1000 cfu/100 ml and FC50 and FS50 $<$ 100 cfu/100 ml), the corresponding probabilities of *P. aeruginosa* infective levels are lower than 15%. On the other hand, the use of values higher in one order of magnitude means hazards about 30% (table 1b).

The probability of *P. aeruginosa* hazardous concentrations in a determined seawater sample is very low (2%) if the concentrations of the bacterial indicators in this sample are lower than 10000 TC/100 ml or 1000 FC or FS/100 ml. Unlike the samples with values higher than these levels, which present infective concentrations of *P. aeruginosa* about 70% (table 2).

These results are in agreement with those of other authors (4,6), which find a high number of otitis cases in swimmers, if the fecal pollution levels of the beaches are higher than the mentioned standards of health quality. So, it is possible to assume that the analysis of the cited indicator microorganisms of pollution and the use of health criteria based on these microorganisms would reduce the potential health hazard associated with the *P. aeruginosa* infections in swimming waters.

TABLE 1. Relationship between XX50 values of indicator microorganisms and the probability of *P. aeruginosa* infective concentrations.

Table 1a.

PROBABILITY OF $>$*P. aeruginosa* 100 cfu/100 ml (%)	XX50 INDICATOR VALUES (cfu/100 ml)		
	TC50	FC50	FS50
10	3.5E2	1.1E2	7.0E1
50	4.5E4	7.5E3	3.5E3
90	6.6E6	5.5E5	1.7E5

Table 1b.

STANDARD		PROBABILITY OF $>$*P. aeruginosa* 100 cfu/100ml (%)
PARAMETER	CONCENTRATION (cfu/100 ml)	
TC50	10000	34
	1000	15
FC50	1000	27
	100	9.5
FS50	1000	33
	100	11.5

TABLE 2. Percentages of samples with *P. aeruginosa* infective concentrations respect to the fecal pollution level.

INDICATOR MICROORGANISM	INDICATOR CONCENTRATIONS	NUMBER OF SAMPLES	*P. aeruginosa* HAZARD CONCENTRATIONS (%)
TOTAL COLIFORMS	$<$1000	20	0.00
	$<$10000	56	1.79
	$>$10000	62	70.97
FECAL COLIFORMS	$<$100	23	0.00
	$<$1000	44	2.27
	$>$1000	66	66.67
FECAL STREPTOCOCCI	$<$100	17	0.00
	$<$1000	50	2.00
	$>$1000	72	63.89

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