

## The seasonality of bacterial quality of water in a tropical developing country (Sierra Leone)

By R. C. WRIGHT

*Rural Water Supply Programme SIL/79/009, UNDP, P.O. Box 1011, Freetown, Sierra Leone*

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### SUMMARY

Natural water sources used as drinking-water supplies by rural settlements in Sierra Leone were examined monthly over a one-year period to detect any seasonal variations in bacterial quality. The 37 °C colony count, levels of selected faecal indicator bacteria and the incidence of *Salmonella* spp. were monitored. A seasonality was demonstrated for all the variables, counts generally increasing with the progression of the dry season, culminating in peaks at the transition from dry to wet season. Some complications with respect to the interpretation of counts of faecal indicator bacteria from raw tropical waters are noted.

### INTRODUCTION

A large proportion of the rural population in the developing world takes water from natural sources directly for drinking (WHO, 1973; Feachem, McGarry & Mara, 1977). These sources are exposed to contamination with faecally derived organisms. The water is usually not treated at all or treated insufficiently to ensure acceptability according to international guidelines (WHO, 1983).

The assessment of bacterial water quality in such circumstances entails measuring levels (often high) rather than detecting the presence or absence of faecal indicator bacteria, in the hope that some means of reducing these levels as far as possible (if not to zero) can be identified.

One of the factors influencing the levels of faecal bacteria in tropical waters is seasonal change. The transition from a dry to a wet period has been associated with an increase in such levels (Feachem, 1974; Barrell & Rowland, 1979), but whether levels are generally higher in the wet season than the dry season (Barrell & Rowland, 1979), lower (Bagde & Varma, 1982) or similar (Muhammed & Morrison, 1975) seems to vary according to source type and/or geographical location. Comprehensive data are lacking. The most detailed study of this subject so far reported from West Africa (Barrell & Rowland, 1979) was not sustained for a full one-year period and was restricted to an investigation of well-water quality.

The present work was undertaken to identify the seasonal variations in water quality which might occur in tropical developing countries, both in general terms and with respect to specific source types. Such data would assist in the prediction of potential health risks and permit correlation with the observed seasonality of epidemiological data (Chambers *et al.* 1979).

## MATERIALS AND METHODS

*Water sources and sampling procedure*

Each of 28 water sources used by 30 rural settlements situated within a study area of approximately 185 km<sup>2</sup> in the Southern Province of Sierra Leone was sampled monthly over the period August 1979 to July 1980. The sources comprised 1 major river, 1 minor river, 13 streams, 4 springs, 5 swamps and 4 wells. (The wells were hand-dug, unlined and were poorly protected at their heads.) The major and minor rivers were each sampled at two locations, where they were being used by different settlements. Some sources dried completely during the dry season and could not then be sampled; however, where the inhabitants of a settlement dug a shallow well in a dried swamp or stream bed, for their continued supply of water, this was tested and was considered to represent the swamp or stream.

An approximately 100 ml sample was taken and the water temperature recorded at each place and month. All samples were returned to the laboratory for analysis within 6 h.

*Bacterial analysis*

Counts of faecal coliform (FC) organisms, presumptive *Escherichia coli* (indole-positive FC), faecal streptococci (FS), presumptive *Streptococcus faecalis* (FS capable of producing characteristic colonies on tyrosine-sorbitol-thallos acetate agar) and *Clostridium perfringens* spores, and the presence or absence of *Salmonella* spp. in a 10 ml sample were determined by methods previously described (Wright, 1982); these were standard methods (DHSS, 1969) except that tubes set up for the FC test were incubated directly at 44 °C.

The colony count was determined by pour-plating 1 ml each of sample that had been diluted 10 and 100 times in quarter-strength Ringer's solution, with Yeast Extract Agar (DHSS, 1969). Because of the high ambient temperature in Sierra Leone, plates were incubated at 37 °C only, rather than the recommended 20–22 °C and 37 °C. Counts were recorded as colony-forming units (c.f.u.) after 24 h incubation.

*Climatological data*

Rainfall and air temperature data over the period of study were obtained from the meteorological station at Njala, situated within the study area.

## RESULTS

Variations in rainfall and temperature over the period of study (Fig. 1) were typical for Sierra Leone (Odell *et al.* 1974). Rainfall variations were more extreme than in other West African countries (Ojo, 1977). Seasonal transition periods were November–December (wet to dry season) and April–May (dry to wet season). Rain fell on only 14 days during the 5-month period December to April. The average water temperature in the sources examined remained within the narrow range 25.1–27.5 °C throughout the year. The patterns of seasonal variation in different source waters are shown in Fig. 2. The overall pattern consists of a slight

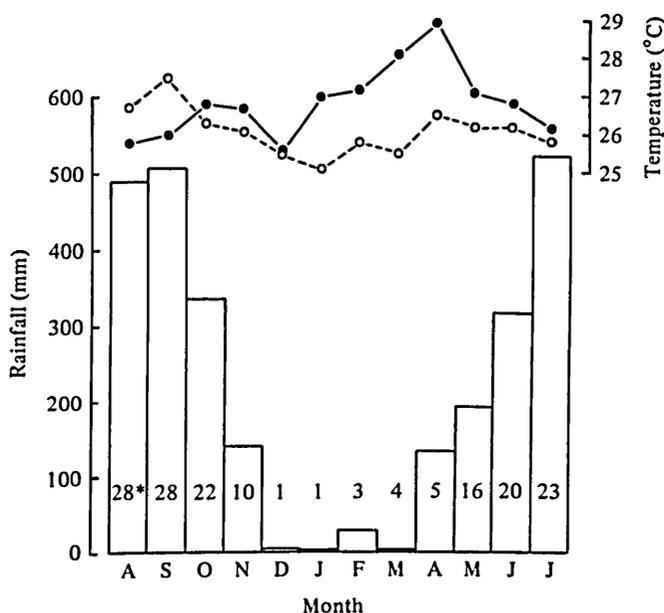


Fig. 1. Monthly rainfall, mean air temperature (●—●) and mean water temperature (○---○) over the period of study.

\*Figures in this line refer to the number of days per month when rainfall exceeded a trace.

rise in counts at the transition from wet to dry season, a progressive rise during the dry season and a further increase at the transition from dry to wet season. Counts were consistent and at their lowest during the wet season.

An analysis of the data for individual source types produced essentially similar patterns of seasonal variation to the overall pattern, with the exception of data for the major river: for this source, the transition from wet to dry season corresponded with a decline in the levels of the bacterial variables examined and wet-season levels were higher than dry-season levels. All source types were prone to faecal contamination and to seasonal fluctuations in contamination.

Some divergence was observed between the counts of the faecal groups, FC and FS, and counts of the presumptively identified specific indicators, *E. coli* and *S. faecalis*, respectively, as these increased during the dry season. A statistical comparison of the counts (Table 1) indicated this divergence to be significant.

It was further observed that, whilst the primary combinations of positive and negative tubes obtained in the FC multiple-tube test (with minerals-modified glutamate medium) were normal throughout the year, the secondary combinations of tryptone-water tubes (derived from the positive glutamate tubes) shown to be indole-positive and -negative were sometimes abnormal, i.e. different from those given in standard tables (e.g. DHSS, 1969; WHO, 1983). This appeared to be a seasonal effect. In particular, when FC counts were highest, during the months April to June (the transition from dry to wet season), 12% of the water samples examined produced abnormal combinations of indole-positive and

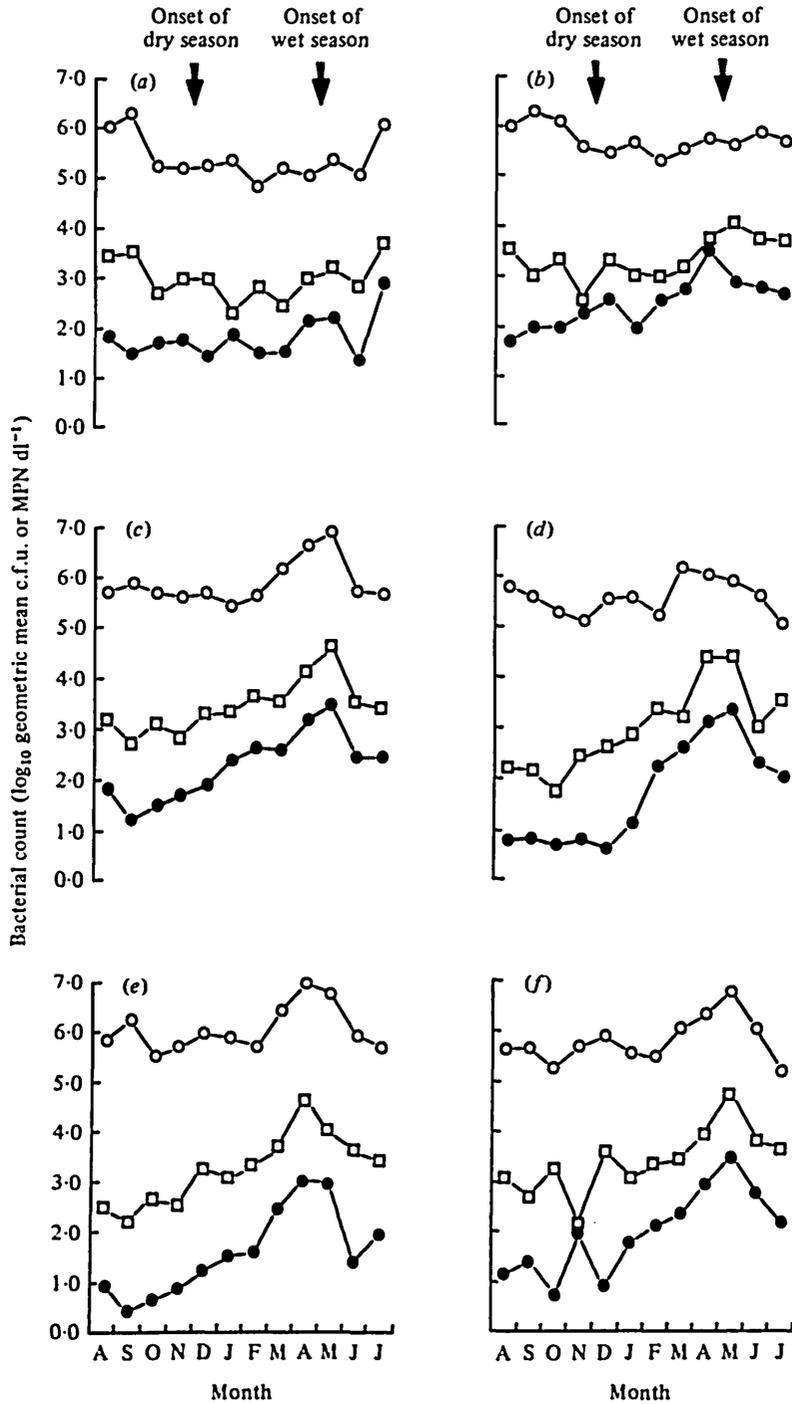


Fig. 2. Seasonality of bacterial water quality in different source types: a, major river; b, minor river; c, stream; d, spring; e, swamp; f, well; ○—○, total bacteria; □—□, FC; ●—●, FS. For clarity, counts of presumptive *E. coli*, presumptive *S. faecalis* and *C. perfringens* spores are not shown.

Table 1. Correlation of monthly mean FC and FS counts with respective confirmation rates as presumptive *E. coli* or presumptive *S. faecalis*

	Month											
	Aug.	Sep.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	July
Log <sub>10</sub> geometric mean FC count dl <sup>-1</sup> (x)	2.89	2.56	2.82	2.62	3.20	3.10	3.43	3.44	4.09	4.37	3.49	3.44
Log <sub>10</sub> geometric mean presumptive <i>E. coli</i> count dl <sup>-1</sup>	NR	NR	2.20	2.24	2.61	2.61	2.83	2.77	3.27	3.41	2.84	2.66
% confirmation rate (y)	-	-	24	40	26	32	25	21	16	11	22	16
Correlation*	$y = e^{5.214 - 0.625x}$ , $r = -0.890$ , $P < 0.001$											
Log <sub>10</sub> geometric mean FS count dl <sup>-1</sup> (x)	1.42	1.07	1.18	1.48	1.51	1.88	2.23	2.47	3.02	3.26	2.24	2.28
Log <sub>10</sub> geometric mean presumptive <i>S. faecalis</i> count dl <sup>-1</sup>	1.16	0.91	0.96	1.31	1.29	1.53	1.50	2.02	2.62	2.66	1.84	1.89
% confirmation rate (y)	54	67	60	67	59	45	19	33	42	26	41	41
Correlation*	$y = 81.92 - 17.85x$ , $r = -0.804$ , $P < 0.01$											

NR, result not recorded.

\*Expressed as the best-fit regression equation, correlation coefficient and level of significance.

Table 2. Frequency of isolation of organisms identified as *Salmonella* spp. from water sources, related to the time of year and to FC counts

	Month											
	Aug.	Sep.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	July
% of sources shown to contain <i>Salmonella</i> spp.	NT	NT	NT	7	18	50	25	36	18	54	54	54
Geometric mean FC count dl <sup>-1</sup>	780	360	660	420	1600	1300	2700	2800	12000	23000	3100	2800

NT, not tested.

-negative tryptone-water tubes; this is more than ten times higher than the recommended level at which the use of the multiple-tube test is invalidated (APHA, 1981).

A similar, although less pronounced, complication was experienced with the multiple-bottle pre-enrichment method for estimating *C. perfringens* counts. Whilst combinations of positive and negative bottles of differential reinforced clostridial medium (DRCM) were normal, combinations of positive and negative tubes of Crossley milk obtained after subculture from positive DRCM bottles were sometimes abnormal. No seasonal variation was observed, but from all the samples examined throughout the year, 2 % produced such abnormal combinations.

Organisms identified as *Salmonella* spp. were isolated often during the investigation (Table 2). Seasonally, there appeared to be two peaks in isolation frequency, the first in January and the second covering a 3-month period at the beginning of the wet season, May to July. There were no significant relationships between

the frequency of isolation of these organisms and the levels of any of the faecal indicators tested for.

#### DISCUSSION

It appears from the seasonality of bacterial water quality demonstrated that the population in the study area would be increasingly exposed to the possibility of acquiring water-borne, faecally derived infections as the dry season progressed, with the highest risk occurring at the beginning of the wet season. Presumably, because of the lack of sanitation development in the area (Wright, 1982), much human as well as animal faecal material was deposited on the soil surface. The action of rainfalls during the dry season and the first rainfalls of the wet season then flushed accumulated faecal matter into water sources.

Additionally, as water sources diminished in volume during the dry season (and often changed from a flowing state to stagnancy), the faecal contamination which undoubtedly occurred whilst inhabitants were collecting water would have been diluted to a lesser extent.

The increasing level of faecal contamination indicated during the dry season supports the hypothesis of a 'concentration effect' proposed by Drasar, Tomkins & Feachem (1981), to explain dry-season increases in diarrhoeal disease in the tropics. However, 'concentration' is not considered an appropriate word as this implies that faecal bacteria are surviving for long periods whilst the volume of water in a source decreases. It is rather the decreasing extent of dilution of a continual input of faecal bacteria that leads to the observed increase in their levels and an 'accruing-input effect' would thus be a better expression.

Feachem (1974) and Barrell & Rowland (1979), whilst also identifying an increase in indicated faecal contamination at the transition from dry to wet periods, did not show any trend(s) within particular seasons. Barrell & Rowland (1979) did, however, suggest that dry-season levels of contamination were lower (in wells) than wet-season levels. In the present study, average wet-season levels of contamination were clearly lower than dry-season levels, with the exception of data for the major river. These latter data can be explained by the action of ephemeral, tributary streams, which would have carried pollution from areas removed from the main river channel during the wet season, but which dried up in the dry season.

Having implicated the transition period from dry to wet season as a time of increased health risk, it must be pointed out that it is precisely at this time that care should be taken over the interpretation of faecal indicator counts, as confirmation rates were at their lowest. This was probably due to the differential survival patterns of the various constituent organisms of the faecal indicator groups in the faecal matter which had accumulated during the dry season. One consequence of this is to cast doubt on the use of the FC/FS ratio as an indication of the origin of pollution (Geldreich & Kenner, 1969; Feachem, 1975). This ratio applies to fresh faecal contamination. It could well be that FS survive for longer in human faeces after defaecation (but before entering a water source) than FC; if such faecal matter then entered water, a FC/FS ratio indicating non-human pollution would be observed.

Another aspect of low confirmation rates is that direct subculture from

positive tubes of minerals-modified glutamate medium (MMGM) to tryptone water for the demonstration of indole production (DHSS, 1982) cannot be recommended for testing waters such as those encountered herein. The high numbers of FC other than *E. coli* which might be present in these waters could produce positive tubes of MMGM containing mixed cultures of small numbers of *E. coli* and large numbers of other FC organisms; on subculture to tryptone water, the other FC organisms might then competitively inhibit the growth of *E. coli*, producing a false-negative indole reaction. This is proposed as the explanation for the observed production of unacceptably high numbers of abnormal combinations of indole-positive and -negative tubes of tryptone water. For similar reasons, with respect to the *C. perfringens* count, direct subculture from a potentially mixed culture in a differential medium to a non-selective medium for confirmation is not considered good practice.

Wherever possible, therefore, it is recommended that, for the analysis of raw tropical water for faecal indicator bacteria, membrane filtration should be used in preference to the multiple-tube test, the former producing relatively pure cultures for confirmatory subcultures to be made directly. Such subcultures are required because of the probable presence of closely related organisms (of less sanitary significance) in water samples, other than the primary faecal indicator which is being investigated.

The lack of a demonstrable, significant correlation between faecal indicator counts and the incidence of *Salmonella* spp. supports earlier observations (Wright, 1982) and might be considered to preclude the drawing of anything other than tentative conclusions about the relative 'safety' of waters containing different levels of faecal indicator bacteria, at least over the range of faecal indicator counts described herein. This requires further investigation.

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