EVALUATION OF THE PRESENCE–ABSENCE (P–A) TEST: A SIMPLIFIED BACTERIOLOGICAL TEST FOR DETECTING COLIFORMS IN RURAL DRINKING WATER OF INDIA

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Abstract. One thousand three-hundred and ninety-four drinking water sources comprising ground water, surface water and piped supplies were tested in order to compare the presence–absence (P–A) test with standard MPN method to detect coliforms as indicators of water quality. Out of 1394 samples, 1074 (77.04%) and 1030 (74.88%) were positive by the MPN and P–A test, respectively. The P–A test detected 96% of the positives detected by the MPN test. The P–A test may be effectively used as a rapid screening method to detect coliform contamination in less polluted sources such as ground water and piped supplies.

1. Introduction

In rural India ground water supplied via hand pumps is becoming an increasingly important source of drinking water. This water, however, varies and is not always free of bacteriological contamination and thus screening for coliforms is very desirable. In addition, rural piped water supply schemes require monitoring for effective removal of coliforms species after treatment. There is, therefore, a great need in India for a cheap and simple method for monitoring drinking water particularly of rural sources for bacteriological contamination. The presence–absence (P–A) test is an inexpensive procedure for rapid qualitative detection of bacterial indicators of faecal pollution, particularly that developed by Clark (1967) as a means for monitoring drinking water systems. This was considered by several workers as an alternative to the multiple tube (MPN) and membrane filtration (MF) method for monitoring water samples (Clark, 1967, 1969, Pipes and Christians, 1984).

The P–A test has certain advantages where resources and time factors are limited. If a significant number of the samples of test water is expected to be free of faecal coliforms (as in the case with ground water or chlorinated water) it could be a waste of resources to mount a quantitative test for each sample.

Under the programme of National Drinking Water Mission (Government of India) remote villages from several regions of India (Figure 1) were surveyed for bacteriological quality of drinking water. In this paper data obtained from standard total coliform (MPN) tests were compared with those from the P–A test.

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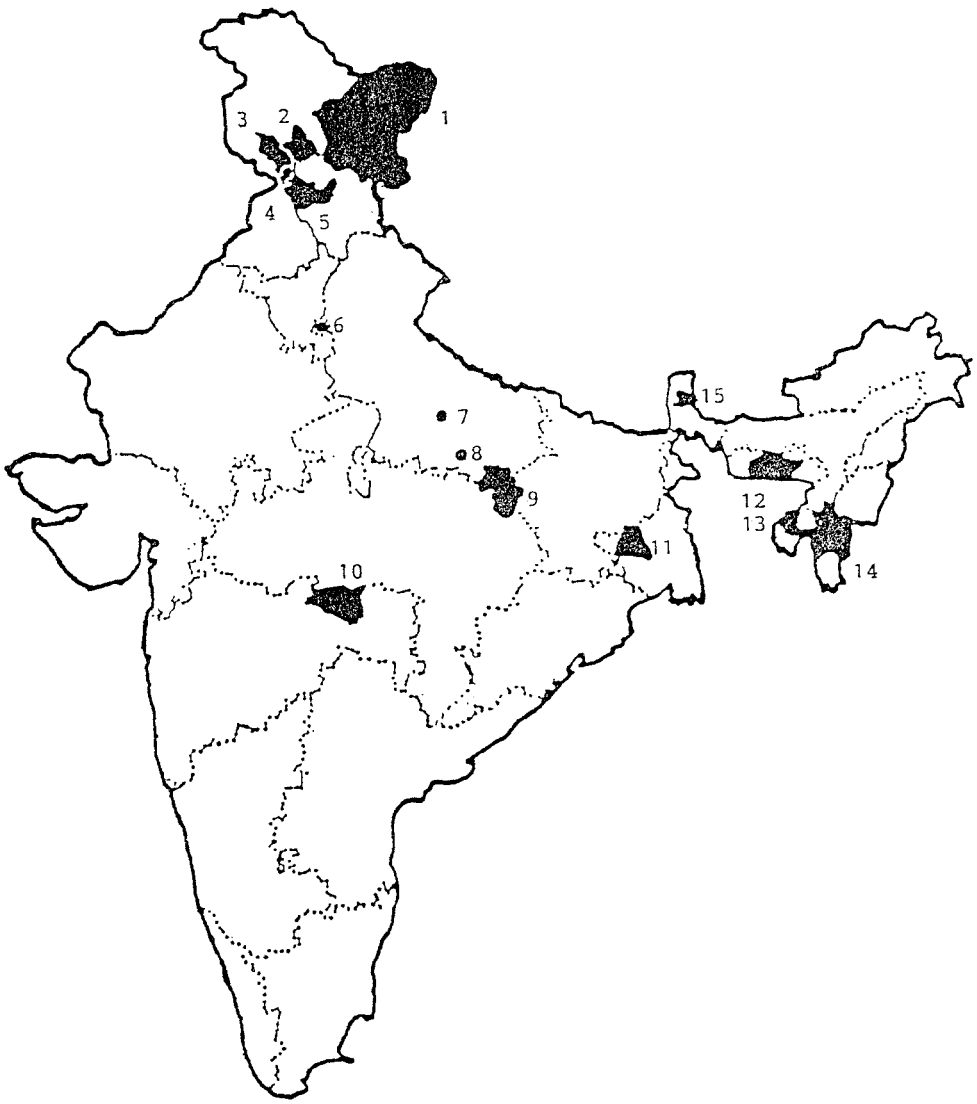

Fig. 1. Locations of drinking water tests. 1. Leh (J & K); 2. Doda (J & K); 3. Udhampur (J & K); 4. Jammu (J & K); 5. Kangra (H.P.); 6. Delhi; 7. Lucknow (U.P.); 8. Allahabad (U.P.); 9. Mirzapur (U.P.); 10. Nagpur (Maharashtra); 11. Bankura (W.B.); 12. West Khasi Hillls (Meghalaya); 13. N. Tripura (Tripura); 14. Aizawl (Mizoram); 15. East & South Sikkim (Sikkim).

2. Materials and Methods

2.1. Sampling Area

Water samples were collected from surface water sources in Aizawl district, Mizoram, West Khasi Hills district, Meghalaya, North Tripura district, Tripura, East

and South districts, Sikkim, Udhampur and Doda districts, Jammu and Kashmir and Kangra district, Himachal Pradesh. Leh (Ladakh) in Jammu and Kashmir is a high-altitude, cold, arid region where melting ice makes a major contribution to the surface water used for drinking. Ground water from hand pumps, tube wells, ring wells and dug wells were collected in North Tripura district, Tripura, Mirzapur district, Uttar Pradesh, Bankura district, West Bengal, Nagpur district, Maharashtra, Leh, Jammu and Kashmir and cities of Lucknow, Allahabad and Delhi. Water samples were taken from several rural piped water supply schemes in Sikkim, Meghalaya, Himachal Pradesh and Jammu and Kashmir.

2.2. Bacteriological Analysis

A total of 1394 water samples were collected from 271 hand pumps, 230 dug wells, 52 tube wells, 308 springs, 192 streams, 48 river and 293 piped supplies from areas shown in Figure 1. Sampling was carried out with the assistance of local Public Health Engineering Departments. Water samples were collected in sterile glass bottles and transported to the field laboratory in ice. The total coliform test (MPN) was performed by the multiple tube technique using MacConkey broth (Hi-Media Pvt. Ltd. Bombay) incubated at 37°C for 48 h (APHA, 1985).

The P–A test was performed as described by Clark (1967). Milk dilution bottles (capacity 150 ml) were filled with 50 ml double strength MacConkey broth (Hi-Media Pvt. Ltd., Bombay) and autoclaved. Fifty ml samples were inoculated and the bottles were incubated at 37°C for 48 h. The acid and gas formation in the broth was considered as a positive test.

2.3. Statistical Analysis

McNemar statistics were used for comparing results of P–A with MPN.

3. Results and Discussion

Using the total coliform (MPN) test, 1074 (77.04%) out of 1394 of the samples were found positive for coliforms while 1030 (73.88%) were positive using the P–A test. The P–A test detected 96% of the positive detected by the MPN test (Figure 2). Around 108 (7.74%) disagreements were detected, of which 76 (5.45%) were positive by the MPN test but were negative for the P-A test, whereas 32 (2.28%) were positive by the P–A test but were negative for the MPN test (Table I). The values of disagreements between various water types ranged between 4.34 and 10.23% (Table I). The statistical analysis clearly indicates that the P–A test is comparable with the MPN test. (McNemar statistical values lie in the range of 0.043–2.84; Table I). Out of the total 76 tests which were positive by the MPN test but negative for the P–A test, 53 (76.32%) had coliform counts 10 per ml (Table II).

TABLE I

Comparison of the results of MPN test with the P–A test in different water sources.

Source	Total samples	P–A + MPN-	P–A – MPN+	Disagreement between test (%)	McNemar statistics
Ground water					
Hand pump	271	7	15	8.11	1.49
Dug well	230	0	10	4.34	2.84
Tube well	52	3	2	9.61	0.44
Spring	308	9	19	9.09	0.43
Surface water					
Stream	192	2	8	5.20	1.58
River	48	1	2	6.25	0.57
Piped supplies	293	10	20	10.23	1.64
Total	1394	32	76	7.74	1.28

TABLE II

MPN values of samples found negative to P–A test.

Source	Number of of samples	MPN counts per 100 ml (%) 1–5	6–10	> 10
Hand pump	15	6	3	6
Dug well	10	1	6	3
Tube well	2	2	0	0
Springs	19	6	7	6
Stream	8	1	2	5
River	2	0	0	2
Piped supplies	20	13	6	1
Total	76	29 (38.15)	24 (31.57)	26 (30.26)

Figures in parenthesis indicate percentage.

From the data presented here it is evident that the P–A test using 50 ml water samples is comparable to the standard MPN test used in the detection of coliforms and the P–A test failed to detect coliforms when water samples contained coliforms of 10 per ml.

The data presented here demonstrate that a higher number of hand pumps, tube wells and piped supplies were found free of coliform contamination (Figure 2). Similar findings were also observed in our earlier studies (Ramteke *et al.*, 1990; Ramteke *et al.*, 1992). As a large number of these sources were free of fecal

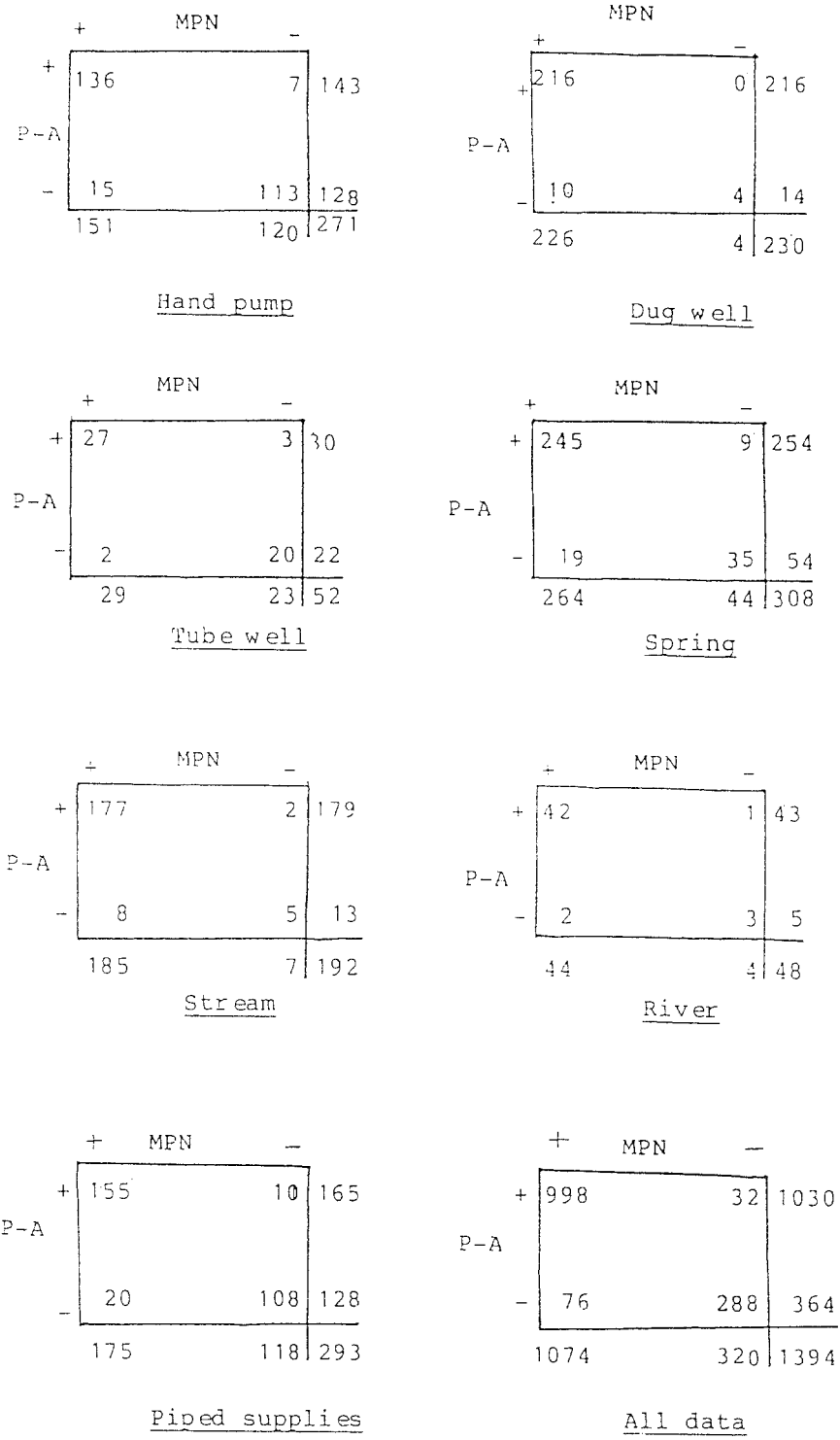

Fig. 2. Contingency table for P–A and MPN in different drinking water types.

contamination, the P–A test may be employed as a rapid screening method to detect the presence of contamination. Only sources with positive P–A test require further testing for fecal coliforms. Thus the results of this study confirm the applicability of the P–A test for testing raw ground water sources.

To make the test more appropriate for the rural environment the test bottle may be filled directly from the pump or tap precalibrated 100 ml mark and transported back to the laboratory for incubation at ambient temperature (30–37°C). The P–A test bottles can be prepared at a base laboratory and distributed to the appropriate public health workers and persons responsible for rural water supplies. Very little technical knowledge is required for performance of the test, and with a minimum of facilities and trained man-power to screen the highly contaminated water samples. The test can be used to assist in initiating more widespread and frequent bacteriological testing of rural water supplies which is essential in overcoming the widespread incidence of water-borne diseases caused by consumption of contaminated water. The test will economize the water quality surveillance programme in developing countries like India.

In addition to coliforms, other indicators such as fecal streptcocci, *Ps. aeruginosa, Aeromonas* spp. and sulfite reducer clostridia (SRC) can be detected by the P–A test (Clark, 1967, 1969, 1980; Clark and Vlassoff, 1973). In a study by Martins *et al.* (1991) all the above indicators were detected by the P–A test using the single medium of lactoslauryl tryptose tryptone (STM) broth. This finding makes the P–A test a very promising tool for the evaluation of the bacteriological quality of drinking water especially in the tropics where the coliforms indicators may not be adequate (Martins *et al.*, 1991).

The P–A test was found to be the most sensitive and cost-effective means of testing potable water supplies for bacteriological contamination from a study that covered three continents (Dutka, 1990). It is easily portable and requires less effort and minimum storage facilities, and is thus ideal for testing potable water qualty anywhere in the world (Dutka 1990).

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