

## BACTERIOLOGICAL ASSESSMENT OF DRINKING WATER OF RAWALAKOT DISTRICT, AZAD KASHMIR, PAKISTAN

Syed Waasif Shoukat<sup>1</sup>, Said Akbar Khan<sup>1</sup>,  
Muhammad Ishtiaq<sup>2✉</sup>, Inayat Shah<sup>3</sup>,  
Syed Umair Ullah Jamil<sup>1</sup>, Tahseenullah Khan<sup>1</sup>

### ABSTRACT

**OBJECTIVE:** To assess the bacteriological quality of drinking water sources in District Rawalakot, Azad Kashmir, Pakistan.

**METHODS:** After approval from the Ethical Review Committee of Bahria University Islamabad; this cross-sectional descriptive study was conducted in Rawalakot District from September 2017 to March 2018. Out of 90 samples, 45 samples (15 each from bore-well, open-well & spring water) were collected in September & October 2017 as pre-rainfall season's samples and 45 samples were collected in post-rainfall season of February & March 2018, exactly from the same 45 sites from which pre-rainfall water samples were collected. All the samples were analyzed for total bacterial, total coliforms, Escherichia Coli, Shigella and Salmonella Species.

**RESULTS:** Overall bacterial growth on Nutrient Agar was more commonly observed in samples from Bore-well (n=11/15; 73.33%) during pre-rainfall season and in open-well samples (n=12/15; 80%) during post-rainfall season. Total coliforms growth on MacConkey agar was more frequent in post-rainfall season in samples from spring-water (n=9/15; 60%). On eosin methyl blue agar, majority (n=5/15; 33.33%) of Escherichia Coli growth was observed during post-rainfall season in spring water samples. Almost all of the post-rainfall season samples revealed less number of Shigella and Salmonella species compared to pre-rainfall water samples.

**CONCLUSION:** The post-rainfall season drinking water samples of bore wells, open wells and spring water showed higher number of bacteriological contaminations except Shigella and Salmonella species which were reduced in post-rainfall samples of bore-wells and revealed equal numbers among open-wells and spring water samples.

**KEY WORDS:** Drinking Water (MeSH); Springs (MeSH); Bacteriological (MeSH); Escherichia Coli (MeSH); Salmonella (MeSH); Shigella (MeSH).

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### INTRODUCTION

Water is considered as one of the most important element on earth surface, maintaining life. Water has covered approximately 70% of the Earth's surface but the fresh water supply is of great concern in the developing and developed countries.<sup>1</sup>

According to World Health Organization report, globally approximately 1.2 billion people are without safe drinking water; and mostly affecting the people of sub-Saharan Africa, South Asia and East Asia.<sup>2</sup> Bacteriological contaminations are reckoned as one of the most threatening problem of drinking and is a major cause of water-borne disease i.e.

1. Department of Earth and Environmental Sciences, Bahria University, Islamabad Campus, Islamabad, Pakistan.
2. Department of Community Medicine, Northwest School of Medicine Hayatabad Peshawar, Pakistan.
3. Institute of Basic Medical Sciences, Khyber Medical University, Peshawar, Pakistan.  
Email: drishtiaq250@yahoo.com  
Contact#: +92-3349121822

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gastroenteritis, diarrhea, dysentery, typhoid, hepatitis A & E and other health related issues.<sup>3,4</sup>

In many international and national studies, each year approximately 0.5 million children die due to diarrheal diseases.<sup>5,6</sup> Moreover, according to research studies, in developing countries approximately 1.8 million children died from biological agents of water and food in year 1998.<sup>6-8</sup> Inefficient sewage system and improper sanitation barriers, direct discharge of waste into the natural reservoirs and water bodies, are considered as the major source of fecal contamination of water.<sup>9</sup> Generally, it is believed that pathogenic bacteria for example Escherichia coli, Thermo-tolerant coliforms, total coliforms, fecal streptococci and Clostridium perfringens, parasites and viruses can contaminate quality of drinking water. Most probable sources of these contaminants in water can be fecal matter, agriculture runoff and domestic waste, urban and pasture runoff.<sup>10,11</sup>

In Pakistan, both surface and ground sources of drinking water are polluted with coliforms, toxic metals and pesticides and it is ranked at number 118 among 190 countries regarding the quality of drinking water.<sup>12,13</sup> Usually, the bacteriological assessment of drinking water is carried out to check the presence of total and fecal coliforms. Coliforms are generally found in the environment and are usually harmless to human beings but their presence in water is used as sign of water polluted with disease causing agents. The presence of Escherichia coli and fecal coliforms in water suggests that water is

**TABLE I: RESULTS OF PRE & POST- RAINFALL WATER SAMPLES SOURCES OF BORE WELLS, OPEN WELLS & SPRING WATER (n=90)**

	Pre-rainfall season			Post-rainfall season		
	Bore well (n=15)	Open well (n=15)	Spring water (n=15)	Bore well (n=15)	Open well (n=15)	Spring water (n=15)
Nutrient Agar ( $10^7$ )	11 (73.33%)	9 (60%)	6 (40%)	7 (46.67%)	12 (80%)	9 (60%)
MacConkey Agar ( $10^7$ )	5 (33.33%)	1 (6.67%)	3 (20%)	6 (40%)	8 (53.3%)	9 (60%)
Eosin Methyl Blue Agar ( $10^7$ )	2 (13.33%)	0 (0%)	1 (6.67%)	3 (20.00%)	3 (20%)	5 (33.33%)
Salmonella and Shigella Agar ( $10^7$ )	6 (40%)	1 (6.67%)	1 (6.67%)	0 (0%)	0 (0%)	1 (6.67%)

Pre Rainfall Season: September & October Samples; Post Rainfall Season: February & March Samples

contaminated with animals or human waste.<sup>14</sup>

Pakistan being a developing country and thus have high burden of communicable and non-communicable diseases. Moreover, most of the water sources in Pakistan i.e. rivers, lakes, tube-wells, dug-wells and ground water aquifers are highly contaminated with bacteria, therefore this cross-sectional study was carried out in the nearby villages of District Rawalakot, Azad Kashmir, Pakistan; to assess the bacteriological assessment of drinking water resources; and to communicate findings to the local communities and concerned departments, for relevant measures to reduce the burden of water borne diseases.

## METHODS

After taking approval from the Ethical Review Committee of Bahria University Islamabad, Pakistan, a cross-sectional descriptive study was carried out, from September 2017 to March 2018, in the Rawalakot District, Azad Kashmir, Pakistan.

In this study, a total of ninety samples (30 each from bore well, open well & spring water) were collected through convenience technique from the drinking water sources and were analyzed for bacteriological assessment. Out of 90 samples, 45 samples (15 each from bore well, open well & spring water) were collected in September & October 2017 as pre-rainfall seasons samples and 45 samples were collected in post rainfall season of February &

March 2018, exactly from the same 45 sites from which pre-rainfall water samples were collected. Almost all of the drinking water sources used by the local communities of District Rawalakot were included in the sampling frame i.e. bore-wells, open-wells and spring water; and were analyzed for bacteriological assessment. The bacterial analysis of the drinking water samples were carried out by plate count method; and thus on Nutrient-Agar showed the total bacterial load, Eosin Methyl Blue Agar showed *Escherichia coli*, SS-Agar showed presence of *Salmonella* and *Shigella* whereas MacConkey agar showed growth of total coliforms in the collected drinking water samples. Finally a total of 360 parameters were assessed via four different plate counts in the pre- and post-rainfall water samples. Finally the data was analyzed and presented in form of tables against the qualitative water sources variables of bore-wells, open-wells and spring water.

## RESULTS

During pre-rainfall season, samples from bore-wells showed total bacterial count on Nutrient Agar in 11 (73.33%) cases and isolation of *Salmonella* and *Shigella* in 6 (40%) samples (Table I). Similarly, the water samples from open-well and spring water showed bacterial growth on nutrient agar in nine (60%) and six (40%) samples respectively.

During post-rainfall season, water samples from bore-wells showed total bacterial load on Nutrient Agar in 7 (46.67%) samples and growth of total

coliforms on MacConkey agar in 6 (40%) samples. While in samples from open-well revealed total bacterial count in 12 (80%) samples and total coliforms on MacConkey agar in 8 (53.3%) samples. Spring water samples in post-rainfall season showed total bacterial count and total coliforms in 9 (60%) samples each on Nutrient agar and MacConkey agar respectively.

Overall bacterial growth on Nutrient Agar was more commonly observed in samples from Bore-well (n=11/15; 73.33%) during pre-rainfall season and in open-well samples (n=12/15; 80%) during post-rainfall season. Total coliforms growth on MacConkey agar was more frequent in post-rainfall season in samples from spring-water (n=9/15; 60%). Majority (n=5/15; 33.33%) of *E. Coli* growth on eosin methyl blue agar, was observed during post-rainfall season in spring water samples. Almost all of the post-rainfall season samples revealed less number of *Shigella* and *Salmonella* species compared to pre-rainfall water samples.

## DISCUSSION

Among the bore-well water samples; in pre-rainfall seasons n=11 (73.3%) showed total bacterial count (TBC) and n=5 (33.33%) showed total Coliforms. Moreover, only n=2 (13.33%) showed *E. Coli* and n=7 (46.67%) showed *Salmonella* & *Shigella* (SS). In post-rainfall seasons, n=7 (46.67%) showed TBC, n=6 (40%) showed total Coliforms and none of the samples showed SS. Many international studies found increased numbers of *Coli*, total coliforms, fecal coliforms, *Shigella* and *Salmonella* species in post-rainfall samples.<sup>1,15,16</sup> Moreover, many studies revealed that increased prevalence of water borne diseases was due to increased number of total bacterial count and total coliforms as was confirmed by our study results of post-rainfall water samples.<sup>17,18</sup>

In open-well pre-rainfall samples; n=9 (60%) showed total coliforms whereas in post-rainfall n=12 (80%) were found positive; in pre-rainfall samples, n=1 (6.67%; OW10) while n=8 (53.33%) showed total coliforms in post-rainfall samples. Moreover, in none of the pre-

rainfall samples showed E.Coli whereas n=3 (20%) showed E.Coli in post-rainfall samples. Approximately 6.67% (n=1; OW7) showed SS in pre-rainfall samples whereas in none of the post-rainfall water samples revealed SS. Furthermore, many international studies revealed that numbers of E.Coli and TBC increased after rainfall<sup>19,20</sup> as was confirmed in this study among the open-wells water samples. Interestingly, in a study conducted by Hsu TT showed reduced levels of E.Coli and TBC after rainfall and thus these findings were consistent to this study in which Salmonella and Shigella were in less number in pre-rainfall samples of Bore-wells and open-wells.<sup>21</sup> Many international studies; also revealed that microbial contamination was lesser in post-rainfall water samples.<sup>22,23</sup> Although the findings of the above study was contradictory to the results of open well and spring water samples whereas findings of bore-well supported the international studies findings.

In a study conducted by Onuoha SC in Nigeria; found that 23.1% showed positive results for E.Coli, 11.5% for Shigella; and 3.8% for Salmonella.<sup>24</sup> In spring, pre-rainfall water samples; n=6 (40%) showed TBC, n=3 (20%) showed Total Coliforms and only one (n=1; 6.67%) showed E.Coli; and Salmonella & Shigella species in S1 & S3. In post-rainfall water samples; n=9 (60%) showed TBC, n=5 (33.33%) showed E.Coli and only one (6.67%; n-S6) showed Salmonella & Shigella (Table I). Furthermore, in an international study, conducted by Sadik NJ, et al., in 2017; showed that E.Coli, Shigella and salmonella were present both in the pre-rainfall and post-rainfall water samples.<sup>25</sup>

From this study it was recommended that drinking water sources be protected from the man-made factors resulting in contamination. Moreover, the community needs to be educated; and appropriate measures to be adopted by the concerned departments to reduce the man-made factors in addition to the natural causes of bacteriological contamination of drinking water sources of District Rawalakot, Azad Kashmir Pakistan.

## CONCLUSION

From this study, it was concluded that the post-rainfall season drinking water samples of bore-wells, open-wells and spring water showed higher number of bacteriological contaminations except Shigella and Salmonella species which were reduced in post-rainfall samples of bore-wells open wells, and also among the pre-rainfall water samples of bore wells which showed higher concentration of total bacterial count as compared to post-rainfall water samples. Moreover, the Shigella and Salmonella species also revealed equal numbers among open-wells and spring water samples.

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## AUTHORS' CONTRIBUTIONS

Following authors have made substantial contributions to the manuscript as under:

**SWS & SAK:** Conception and study design, drafting the manuscript, final approval of the version to be published

**MI & SUUJ:** Acquisition, analysis and interpretation of data, critical review, final approval of the version to be published

**IS:** Analysis and interpretation of data, critical review, final approval of the version to be published

**TK:** Acquisition, analysis and interpretation of data, drafting the manuscript, final approval of the version to be published

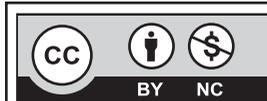
*Authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.*

## CONFLICT OF INTEREST

Authors declared no conflict of interest

## GRANT SUPPORT AND FINANCIAL DISCLOSURE

NIL



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