Seasonal Variation in Water Quality: A Study with Reference to River Kuwano, Basti, (U.P.), India

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Received October 08, 2021; Revised November 10, 2021; Accepted November 17, 2021

Abstract Kuwano river is one of the important rivers flowing through the Basti District, (U.P.), India. Recently, district witnessed rapid urbanization and industrialization, in catchment of river, leading to pressure on surface water quality. The present study aimed to find out the effect of seasonal changes on physico-chemical characteristics of river Kuwano water. The seventeen parameters i.e., pH, TDS, EC, DO, BOD, COD, Total Hardness, Ammonia, Nitrate, Nitrite, Phosphate total, Ca, Mg, Fe, E. Coli, Fecal Coliform and Total Coliform, were included in the study. The higher microbial presence in river water is a health concern. The study reveals that river water quality affected seasonally and is only suitable for bathing and irrigation purpose.

Keywords: Kuwano River, seasonal variation, inorganic nitrogen, microbial parameter, water quality

Cite This Article: Gopal ji Kushwaha, Shivendra Mohan Pandey, and Pradip Kumar, “Seasonal Variation in Water Quality: A Study with Reference to River Kuwano, Basti, (U.P.), India.” Applied Ecology and Environmental Sciences, vol. 9, no. 11 (2021): 976-982. doi: 10.12691/aees-9-11-9.

1. Introduction

River waters are complex, metabolically active, ecosystem components. It regulates the influx of nutrients/pollutants in water system [1]. The river does not just mirror their environment, they also reflect the society around them and accumulates all the “sins” of humanity [2]. India is heading towards a freshwater crisis due to the improper management and contamination of surface water resources, particularly river system [3]. Nutrient loading in the system disrupts intrinsic food web and finally modifies ecosystem productivity. Rivers recharge ground water, in the meantime these pollutants may move slowly into the ground and affect’s ground water quality.

Many regions of world suffer from either chronic shortage of fresh water or the pollution of readily accessible water resources [4]. According to recent UNICEF report about 800 million people in Asia & Africa are living without access to safe drinking water. Consequently, this has caused many people to suffer from various diseases [5]. Fecal pollution of drinking water causes water-borne diseases, which whipped out entire population of cities [6,7,8,9]. Bacteria and bacteriophages suggested as potential indicators of water quality [10].

The quality of river water is an area of concern in Uttar Pradesh. The report, of directorate of environment, UP, reveals that between 2004 and 2008, water quality indicators in many cases are negative. While Kanpur and Varanasi exhibit class B (drinking water with treatment) water upstream, the downstream water quality reduces to class D (fishing). The downstream water quality surveyed in most rivers needs attention [11].

Basti district is a part of middle Ganga plain, covering an area of 2771.7 sq. km. It lies between North latitude 26° 23’ & 27° 30’ and East longitude 82° 17’ & 83° 20’, with total population of 2,780,683 as in 2021 (estimated as per Aadhar). The Basti district receives a normal rain fall of 1169.80 mm; with ground water availability 1,06,048 hm. The area lies in sub-humid climatic region. In the middle and southern part of the district, the river Kuwano and Ghaghara are the main rivers, besides these rivers there are numbers of nalas and ponds in the area. The river Kuwano, originate from Bahriach District of U.P. It passes through Gonda and Siddharth Nagar district before entering in Basti District near Chandhokha village of Ramnager block. In the Basti district, Kuwano flows through north-west to south-east and is fed by its tributaries Bisuni, Manvar and Kathinaya. Before draining through, near Banpur in Kudaraha block, it travels over a length of approx. 55 kms within district boundaries. In Gorakhpur district it merges in Ghaghara near Shahpur village. The river is lifeline of the city. While passing through Basti, urban sewage, solid waste and industrial effluents are incorporated in it. Small-scale industries including the manufacturing units of brassware, iron and carpentry goods, agricultural implements, bricks, agro-products, footwear, soaps, candles, industries are placed in neighbourhood of river. These industries along with three sugar mills, discharge their waste directly or indirectly in the river every day. These untreated effluents contain number of harmful chemicals, that can change the water quality of the river and cause health concern.
The studies on the water quality in relation to Kuwano river have not explored in depth. The river water quality may vary from time to time and site to site, due to input waste composition [12]. It is, therefore, imperative to regularly monitor the quality of Kuwano river water, find out its suitability for different uses.

2. Methodology

The water samples from the three selected stations: Upstream at Bandhuwa village (26° 81′ N & 82° 69′ E), Midstream at Amhut (26° 77′ N & 82° 71′ E) and Downstream at Lalganj (26° 65′ N & 82° 82′ E) were collected in replicate (Figure 1). The samples were collected round the year in the Monsoon (September), Winter (January) and summer (May), to obtain data with seasonal variation. The river water samples from different sampling stations were collected in duplicate in two liter capacity polythene containers pre-washed with dilute Hydrochloric acid, detergent, tap water & distilled water. Sampling was carried out manually without adding any preservative, after flushing out at least 2 to 3 minutes. Samples were brought immediately to the laboratory and kept in the refrigerator for further analysis.

![Figure 1. Sampling location map of the study area](image)

| S.No | Parameters                  | Symbols | Unit | Analytical Method                  | Instrument/Apparatus                          |
|------|-----------------------------|---------|------|------------------------------------|-----------------------------------------------|
| 1.   | pH                          | pH      | -    | -                                  | Ionix pH meter                                |
| 2.   | Total Dissolved Solid       | TDS     | mg/l | -                                  | Ionix TDS meter                               |
| 3.   | Electrical Conductivity     | EC      | μS/cm| -                                  | HM Digital AP-2 Conductivity Tester           |
| 4.   | Dissolved Oxygen            | DO      | mg/l | Azide Modification                | -                                             |
| 5.   | Biochemical Oxygen Demand   | BOD     | mg/l | 5-Day BOD Test                    | -                                             |
| 6.   | Chemical Oxygen Demand      | COD     | mg/l | Closed Reflux, Colorimetric Method| Aimil Spectrochem NV-201 (Model No 022000 SPL) |
| 7.   | Total Hardness              | -       | mg/l | EDTA Titrimetric Method           | Aimil Spectrochem NV-201 (Model No 022000 SPL) |
| 8.   | Ammonia                     | NH₄⁺    | mg/l | "Colorimetric method             | Aimil Spectrochem NV-201 (Model No 022000 SPL) |
| 9.   | Nitrate                     | NO₃⁻    | mg/l | "Brucin Method                   | Aimil Spectrochem NV-201 (Model No 022000 SPL) |
| 10.  | Nitrite                     | NO₂⁻    | mg/l | "Sulphanilic Acid Method          | Aimil Spectrochem NV-201 (Model No 022000 SPL) |
| 11.  | Phosphate (Total)           | PO₄³⁻   | mg/L| "Ascorbic acid Method            | Aimil Spectrochem NV-201 (Model No 022000 SPL) |
| 12.  | Calcium                     | Ca      | mg/l | EDTA Titrimetric Method           | -                                             |
| 13.  | Magnesium                   | Mg      | mg/l | Calculation Method                | -                                             |
| 14.  | Iron                        | Fe      | mg/l | "Phenanthroline Method           | Aimil Spectrochem NV-201 (Model No 022000 SPL) |
| 15.  | E. Coli                     | -       | MPN/100ml| "MPN of coliform             | -                                             |
| 16.  | Fecal Coliform              | -       | MPN/100ml| "MPN method for fecal coliform  | -                                             |
| 17.  | Total Coliform              | -       | MPN/100ml| "MPN of coliform             | -                                             |

*APHA, 2005. [14]. †Trivedi and Goel, 1984. [13].
The collected water samples were used for analysis of seventeen important parameters. The parameters studied were, pH, TDS, EC, DO, BOD, COD, Total Hardness, Ammonia, Nitrate, Nitrite, Phosphate total, Ca, Mg, Fe, E. Coli, Fecal Coliform and Total Coliform. pH, Temperature, TDS, and EC were analysed on spot by Digital portable meters, the remaining parameters were analysed in the laboratory within twenty-four hours of sample collection (Table 1), following the standard procedure [13,14].

3. Results and Discussion

3.1. pH

The pH is a measure of the acidity of water: the lower the pH, the more acidic is the water. In most natural waters pH values are dependent on carbon dioxide, carbonate, and bicarbonate equilibrium. The pH of water is influenced by geology of the area, buffering capacity of water [15]. The water having pH less < 7 may cause tuberculation and corrosion, while higher the values may produce in crustation, sediment deposit and difficulties in chlorination for disinfection of water [16]. In the study pH in all the sampling locations varied between 7.62 to 7.70 during monsoon, 7.8 to 8.0 during winter and 6.2 to 6.74 during summer (Table 2, Table 3 & Table 4). The fluctuations may be due to variation in water flow and photosynthetic activity of aquatic plants [17].

3.2. Total Dissolved Solid

TDS measure the combined content of all inorganic and organic substances contained in a liquid in molecular, ionized, or micro granular suspended form. Water with high solid content is of inferior palatability and may produce unfavourable physiological reaction in the transient consumer [18]. In the present study the TDS fluctuated between 60.3 to 98.3 mg/l (Table -3). Higher values in the monsoon season any be due to riverfront activities, leaching of soil, sewage discharge etc. Moniruzzaman et al. [19] reported an elevated TDS in summer season due to low volume of water. The observed value of the TDS is within the permissible range as prescribed by BIS [20]. TDS content is usually the main factor, which limits or determines the use of groundwater for any purpose [21].

Table 2. Variation in Physico-chemical and biological parameters of the Kuwano river water in Monsoon season

| September 2020 | Upstream | Midstream | Downstream | Mean |
|----------------|----------|-----------|------------|------|
| pH 7.70        | 7.65     | 7.62      | 7.66       |
| Temperature    | 28.1     | 28.2      | 28.5       | 28.27|
| TDS 88         | 95       | 112       | 98.33      |
| EC 171         | 171      | 186       | 176.00     |
| DO 3.8         | 4.2      | 4.0       | 4.00       |
| BOD 0.80       | 1.2      | 1.2       | 1.07       |
| COD 3.0        | 3.4      | 3.4       | 3.27       |
| Total Hardness | 84       | 88        | 87.00      |
| Ammonia 0.05   | 0.05     | 0.04      | 0.05       |
| Nitrate 0.22   | 0.48     | 0.64      | 0.45       |
| Nitrite 0.21   | 0.22     | 0.58      | 0.34       |
| Phosphate total| 0.30     | 0.36      | 0.33       |
| Ca 12.6        | 12.8     | 12.8      | 12.73      |
| Mg 17.42       | 18.34    | 18.59     | 18.12      |
| Fe 1.22        | 1.65     | 1.60      | 1.49       |
| E. Coli 62     | 230      | 113       | 135.00     |
| Fecal Coliform | 220      | 384       | 347.33     |
| Total Coliform | 340      | 384       | 397.33     |

Table 3. Variation in Physico-chemical and biological parameters of the Kuwano river water in Winter season

| January 2021 | Upstream | Midstream | Downstream | Mean |
|--------------|----------|-----------|------------|------|
| pH 7.8       | 7.8      | 8.0       | 7.87       |
| Temperature  | 16       | 15.5      | 15.3       | 15.60|
| TDS 82       | 88       | 85        | 85.00      |
| EC 145       | 136      | 122       | 134.33     |
| DO 6.6       | 6.5      | 6.5       | 6.53       |
| BOD 2.1      | 2.4      | 2.3       | 2.27       |
| COD 3.4      | 3.5      | 3.8       | 3.57       |
| Total Hardness| 103     | 114       | 112.00     |
| Ammonia 0.34 | 0.35     | 0.35      | 0.35       |
| Nitrate 0.82 | 0.90     | 1.04      | 0.92       |
| Nitrite 0.64 | 0.68     | 0.81      | 0.71       |
| Phosphate total| 0.36    | 0.38      | 0.37       |
| Ca 27.2      | 25.6     | 25.0      | 25.93      |
| Mg 18.9      | 22.2     | 21.6      | 20.90      |
| Fe 0.81      | 0.88     | 0.95      | 0.88       |
| E. Coli 111  | 117      | 114       | 114.33     |
| Fecal Coliform| 204     | 215       | 211.67     |
| Total Coliform| 302     | 312       | 312.00     |
Table 4. Variation in Physico-chemical and biological parameters of the Kuwano river water in summer season

| Variable | May 2021 | Upstream | Midstream | Downstream | Mean |
|----------|----------|----------|-----------|------------|------|
| 1 pH     | 6.2      | 6.74     | 6.7       | 6.55       |      |
| 2 Temperature | 28.8 | 28.7     | 28.8      | 28.77      |      |
| 3 TDS    | 60       | 55       | 66        | 60.33      |      |
| 4 EC     | 112      | 108      | 110       | 110.00     |      |
| 5 DO     | 3.1      | 3.2      | 4.0       | 3.43       |      |
| 6 BOD    | 1.8      | 1.6      | 1.8       | 1.73       |      |
| 7 COD    | 2.8      | 2.5      | 3.2       | 2.83       |      |
| 8 Total Hardness | 78 | 76        | 77        | 77.00      |      |
| 9 Ammonia | 0.40     | 0.32     | 0.32      | 0.35       |      |
| 10 Nitrate | 0.45     | 0.58     | 0.70      | 0.58       |      |
| 11 Nitrite | 0.34     | 0.48     | 0.65      | 0.49       |      |
| 12 Phosphate total | 0.24 | 0.27     | 0.26      | 0.26       |      |
| 13 Ca    | 17.2     | 16.12    | 16.9      | 16.74      |      |
| 14 Mg    | 14.8     | 14.6     | 14.6      | 14.67      |      |
| 15 Fe    | 0.43     | 0.32     | 0.26      | 0.34       |      |
| 16 E. Coli | 70       | 136      | 128       | 111.33     |      |
| 17 Fecal Coliform | 246 | 264      | 324       | 278.00     |      |
| 18 Total Coliform | 258 | 378      | 388       | 341.33     |      |

Table 5. Mean ±SD of physicochemical and bacteriological parameters of Kuwano river in seasons

| Variable | Rainy Season (September 2020) (Mean ±SD) | Winter Season (January 2021) (Mean ±SD) | Dry Season (May 2021) (Mean ±SD) |
|----------|----------------------------------------|----------------------------------------|---------------------------------|
| 1 pH     | 7.7±0.04                               | 7.9±0.09                               | 6.5±0.30                       |
| 2 Temperature | 28.3±0.21                            | 15.6±0.29                               | 28.8±0.06                      |
| 3 TDS    | 98.3±12.34                             | 85.0±2.45                               | 60.3±5.51                      |
| 4 EC     | 176.0±8.66                             | 134.3±9.46                              | 110.0±2.00                     |
| 5 DO     | 4.0±0.20                               | 6.5±0.05                                | 3.4±0.49                       |
| 6 BOD    | 1.1±0.23                               | 2.3±0.12                                | 1.7±0.12                       |
| 7 COD    | 3.3±0.23                               | 3.6±0.17                                | 2.8±0.35                       |
| 8 Total Hardness | 87.0±2.65                  | 112.0±5.10                             | 77.0±1.00                      |
| 9 Ammonia | 0.04±0.01                             | 0.3±0.00                                | 0.3±0.05                       |
| 10 Nitrate | 0.41±0.21                             | 0.9±0.09                                | 0.6±0.13                       |
| 11 Nitrite | 0.3±0.21                             | 0.7±0.07                                | 0.5±0.16                       |
| 12 Phosphate total | 0.3±0.03                           | 0.4±0.01                                | 0.3±0.02                       |
| 13 Ca    | 12.7±0.12                              | 25.9±0.93                               | 16.7±0.56                      |
| 14 Mg    | 18.1±0.62                              | 20.9±1.44                               | 14.7±0.12                      |
| 15 Fe    | 1.5±0.24                               | 0.9±0.06                                | 0.3±0.09                       |
| 16 E. Coli | 135.0±86.13                        | 123.7±5.80                              | 111.3±36.02                    |
| 17 Fecal Coliform | 315.3±113.55                   | 230.0±22.23                             | 278.0±40.84                    |
| 18 Total Coliform | 370.3±65.03                 | 312.7±15.17                             | 341.3±72.34                    |

3.3. Conductance

Electrical Conductivity (EC) depends upon temperature, ionic concentration and types of ions present in the water [22]. The conductivity of water samples under study varies between 110 to 176 µS/cm (Table 3), which qualify the BIS standard. The downstream sampling stations has higher EC during monsoon, which attributes higher wastewater discharge from residential area, and run-off from agricultural lands [23]. In monsoon season, water level increases, contains more electrolytes, therefore greater value. However lower values recorded in the summer. In the entire study area EC values don’t exceed permissible limits.

3.4. Oxygen (DO, BOD & COD)

The amount of oxygen in water, shows its overall health. That is, higher oxygen levels suppose that pollution levels in the water are low. Lower oxygen is generally associated with heavy contamination by organic matter. In the present study values of DO fluctuates between 3.4 to 6.5 mg/l (Table-3). The fluctuation may be due to oxygen demanding wastes [23]. Mean value shows that health of water body is not met with the BIS slandered. The high DO in winter is due to decrease in temperature and duration of bright sunlight has influence on the percent of soluble gases (O2 & CO2).

Biochemical Oxygen Demand (BOD) levels can be used as measure of organic load. In the present study, the BOD varied between 1.1 – 2.3 mg/l (Table 3) and within the permissible limits of 3 mg/l [31]. High BOD values during winter season correlate with increased microbial activity with increased oxygen availability. In the study BOD:COD ratio 0.33, 0.63 and 0.60 in monsoon, winter, and summer seasons respectively, reflects greater pollution lode in monsoon [24]. Chemical Oxygen Demand (COD) is closely related to BOD. The higher value of COD revels that discharged effluents has more oxygen stripping capacity, and potential to damage aquatic life. The higher COD/BOD ratio in monsoon, correlate with higher influx of effluents in this season.

3.5. Calcium & Magnesium Hardness

Natural hardness of water depends upon the geological nature of the drainage basin and mineral levels in natural water. It is characterized by content of calcium and...
magnesium salts. The total hardness ranged between 84 to 89 mg/l in monsoon, 105 to 117 in winter and 76 to 78 in summer season (Table 2, Table 3 & Table 4), the calcareous bed rock of the city, and domestic sewage could be the possible cause of hardness in winters.

The principal source of calcium and magnesium in groundwater is the silicate mineral groups like Plagioclase, Pyroxene and Amphibole among Igneous and Metamorphic rocks and limestones and dolomite and gypsum among sedimentary rocks. The calcium hardness is recorded 12.7 to 25.9 mg/l (Table 5) with in study area, highest in winter. The lower values of calcium were observed during the monsoon season due to dilution effect of rainwater. The concentration of the magnesium hardness was observed in the range of 14.7 and 20.9 mg/l (Table 5). The seasonal fluctuation revealed that it was higher in winter season due to enrichment of metal. The increase of magnesium is quite proportionate with calcium in all seasons.

3.6. Inorganic Nitrogen

Nitrogen and phosphorus are essential for plant and animal growth and nourishment, but the overabundance of certain nutrients in water can cause several adverse health and ecological effects. Nitrogen, in the forms of nitrate, nitrite, or ammonium, is a nutrient needed for plant growth. Nitrate can get into water directly as the result of runoff of fertilizers containing nitrate. Ammonia and organic nitrogen can enter water through sewage effluent and runoff from land where manure has been applied [25]. Nitrogen contamination is more of a problem in shallow aquifers. Inorganic nitrogen pollution in aquatic ecosystems create three major environmental problems: (1) it can increase the concentration of hydrogen ions in freshwater ecosystems without much acid-neutralizing capacity, resulting in acidification of those systems; (2) it can stimulate or enhance the development, maintenance and proliferation of primary producers, resulting in eutrophication of aquatic ecosystems; (3) it can reach toxic levels that impair the ability of aquatic animals to survive, grow and reproduce [26]. In the study concentration of various nitrogen compounds viz. ammonia (0.3 - 0.04 mg/l), Nitrate (0.4- 0.9 mg/l) and nitrite (0.3 – 0.7 mg/l) found under permissible limit.

3.7. Phosphorus

The major source of anthropogenic phosphorus is sewage, detergents, agricultural effluents, and fertilizers [27,28]. Higher concentration of PO₄ is an indicative of pollution. The concentration of phosphate in the study is also quite low at all the locations, varied between 0.3 to 0.4 mg/L (Table 5). Phosphate gets adsorbed or fixed as aluminium or iron phosphate in acidic soils or as calcium phosphate in alkaline or neutral soils [29]; as a result, the concentration of phosphate in surface water is usually low.

3.8. Iron

The concentration of iron in natural water is controlled by both physico-chemical and microbiological factors. The weathering of rock and discharge of waste effluents on land are generally considered the main source of iron in ground water. Dissolved carbon dioxide, pH and conductance of water affect the nature of aqueous iron species present in the water [29]. Higher levels of iron are attained by waters with low pH (acid waters). Its higher concentrations generally cause inky flavour, bitter, and astringent taste. It can also discolor clothes, plumbing fixtures and cause scaling which encrusts pipes. Excessive concentration may promote bacterial activities in pipe and service mains, causing objectionable odour and red-rod disease in water. However, a reduction in the iron content can be brought about by aeration of waters containing ferrous iron. In the present investigation, the iron content (0.3 – 1.5 mg/l) was within the desirable limit (Table 5 & Table 6), in all the seasons.

| S.No | Parameters | Unit | CPCB (1979) (IS 2296:1992 & IS10,500:1991) WHO Drinking Water specification | Indian Standard Drinking Water Specification (IS 10500 : 2012) |
|------|------------|------|-----------------------------------------------------------------|---------------------------------------------------------------|
|      |            |      | Drinking | Outdoor bathing | ** | Acceptable | Permissible | No relaxation | No relaxation | No relaxation | No relaxation |
| 1.   | pH         |       | 6.5-8.5 | 6.5-8.5            | ** | 6.5-8.5 | 6.5-8.5 | No relaxation | No relaxation | No relaxation | No relaxation |
| 2.   | TDS        | mg/l  | 500     | 2000               | ** | 500     | 2000 | No relaxation | No relaxation | No relaxation | No relaxation |
| 3.   | EC         | μS/cm | <250    | 250-750*           | -  | -       | -    | -              | -              | -              | -              |
| 4.   | DO         | mg/l  | 6       | 5                  | ** | -       | -    | -              | -              | -              | -              |
| 5.   | BOD        | mg/l  | 2       | 3                  | -  | -       | -    | -              | -              | -              | -              |
| 6.   | COD        | mg/l  | -       | -                  | -  | -       | -    | -              | -              | -              | -              |
| 7.   | Total Hardness | mg/l | 200    | -                  | -  | -       | -    | 200            | 600           | No relaxation | 45               |
| 8.   | Ammonia    | mg/l  | -       | -                  | 1.5 | 0.5     | 0.5  | No relaxation | No relaxation | No relaxation | No relaxation |
| 9.   | Nitrate    | mg/l  | 20      | -                  | 50.0 | 45      | 45   | No relaxation | No relaxation | No relaxation | No relaxation |
| 10.  | Nitrite    | mg/l  | -       | -                  | 3.0  | -       | -    | -              | -              | -              | -              |
| 11.  | Phosphate total | mg/L | -       | -                  | -    | -       | -    | -              | -              | -              | -              |
| 12.  | Ca         | mg/l  | 75-200  | -                  | -   | -       | -    | 75             | 200            | 100            |
| 13.  | Mg         | mg/l  | 30-75   | -                  | -   | -       | -    | 30             | 100            |
| 14.  | Fe         | mg/l  | 0.3     | -                  | **  | 0.3     | 0.3  | No relaxation | No relaxation | -              | -              |
| 15.  | E. Coli    |       | -       | 0                  | -   | -       | -    | -              | -              | -              | -              |
| 16.  | Fecal Coliform |       | -       | 0                  | -   | -       | -    | -              | -              | -              | -              |
| 17.  | Total Coliform |      | 50      | 500                | 0   | 500     | 0    | 500            | 0              | 500            | 0              |

* Good for Irrigation. **No health-based guideline value is proposed
***Shall not be detectable in any 100 ml sample
3.9. Microbial Parameter

The presence of coliforms in water is an indicator of contamination by human or from animal excrement. Presence of coliform also indicates the possibility of presence of other pathogenic microorganisms and further indicates the possibility of contamination of the water source with sewage [30]. The samples showed seasonal variation in number of E. Coli (111.3 to 135.0) and Fecal coliform (278.0 to 335.3). The Total count also show seasonal variation, minimum in winter (312.7) and maximum in monsoon (397.3). This may be attributed to higher discharge from contamination sources (surface runoff, domestic sewage, and septic tank) into the shallow aquifers of city. This indicates that the river water is contaminated and there is possibility and risk of the presence of other enteric pathogens.

The presence of fecal coliforms in water indicates a potential public health problem because fecal matter is a source of pathogenic bacteria and viruses. The indiscriminate disposal of domestic waste, improper disposal of solid waste, leaching of wastewater from landfill areas, further aggravate the chances of bacterial contamination of river water. So, it is recommended that water drawn from such sources should be properly disinfected before being used for domestic applications.

4. Conclusion

The study finds correlation in seasonal variation and Kuwano river water quality. The water quality is not suitable for drinking but qualifies for outdoor bathing (organised), Propagation of wildlife and Fisheries, and irrigation purpose [31].

Statement of Competing Interests

The authors have no competing interests.

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