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Anthropogenic Influences on Microbial Contaminants: A Study of Agra Canal in Haryana Stretch, India

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Abstract

Water serves as an essential resource for irrigation worldwide. The Agra Canal plays a vital role in the irrigation infrastructure of the Faridabad and Palwal districts in Haryana. India. An experimental setup was employed to assess the impact of canal water on locally grown vegetables (Spinach, radish, and tomato). The plants in each pot were irrigated using three different water ratios: 100% groundwater, 50% groundwater + 50% canal water mixing, and 100% canal water. After harvesting, rhizospheric soil was analyzed for similar parameters, followed by fungi characterization and DNA sequencing. Results indicated that location S2 had the highest microbial load, whereas location S6 had the lowest amount detected. It was observed that pots irrigated with 100% canal water exhibited higher microbial load compared to those receiving other watering ratios; this suggests that Agra Canal water contains a substantial amount of fungus, which negatively impacts crop yield, including *Albifimbria verrucaria, Aspergillus novafumigatus, Aspergillus niger, Fusarium equiseti, Altenaria Spp., Chetomeaceae Spp., Humicola Spp.,* and uncultured fungal stains, have also been identified as dominating in rhizospheres soil, as well as in Agra canal water, indicating potential detrimental effects on plant health. In addition, to further explore potential risks, it is necessary to examine and reduce the sewage effluent discharge into Agra Canal.

Keywords Total Plate Count, DNA Sequencing, Microbial load, Crop yield, Sewage, Agra Canal

Introduction

The quality of water resources has become a crucial issue globally, especially in rapidly developing nations such as India, where the combined effects of urbanization and industrialization have resulted in the degradation of water bodies (R. Pal et al., 2017; Sharma et al., 2021). Due to the microbial load, the Yamuna River in the Delhi region is called the sewage canal (Tripathy & Chowdhry, 2017). The Agra Canal, a significant irrigation water source in the Haryana and Uttar Pradesh region, has not been spared from this environmental burden. This case study aims to investigate anthropogenic activities' impact on the Agra Canal microbial loads, providing valuable insights for effective water quality monitoring and management. The Agra canal is a critical water source that serves the needs of the local population for irrigation purposes, and any deterioration in its water quality can have far-reaching consequences, not only for the health and well-being of the surrounding agricultural communities but also for the broader ecosystem and economic viability of the region (Krishan, 2017). Due to unawareness, the impact of water contaminants in this environment, and the vast microbial diversity found in similar tropical ecosystems, many species remain unidentified, and their effects are unassessed. (Ortiz-Vera et al., 2018). Researcher's findings indicate that the discharge of untreated industrial effluents, agricultural runoff, and municipal waste into water bodies can significantly elevate the microbial loads, leading to the proliferation of pathogenic microorganisms and the deterioration of water quality (Mahadikar et al., 2022; Sharma et al., 2021). Azi et al. detected microbial and fungal loads beyond the permissible limits from the contaminated water-irrigated vegetables (Azi et al., 2017), which is unsafe for human health. The seasonal variation in water quality parameters, such as dissolved oxygen, pH, and temperature, can also influence the dynamics of the microbial community within water systems (Pali, 2022). The increasing use of sewage water aggravates the problem of plant fungi in irrigation water as concerns over water scarcity spread (Hong & Moorman, 2005). However, Fungi contamination probably affects the quality of crops either directly or indirectly (Amza, 2018). Fusarium wilt is a severe vascular disease in crop plants; *Fusarium* wilt attacks potatoes, tomatoes, and other Solanaceae plants (Jain et al., 2019). Infections from *Aspergillus niger* species can lead to severe morbidity and mortality of fruits and vegetables, especially in jujube fruit, during growth, harvest, storage, transportation, and processing (Xin et al., 2023). *Fusarium* species cause severe crop degradation in *Maize* (Tiru et al., 2021).

This research extends beyond visible plant health indicators to the rhizospheric soil, one of the most essential microbial hotspots in the soil. Over 19,000 fungi are known to cause diseases in crop plants worldwide. They may remain dormant but alive on both living and dead plant tissues. (Lahlali et al., 2022). Microbial and Fungi analysis of this soil provides valuable insights into the potential hazards of different water sources (Zhang et al., 2019). Surface water (ponds, lakes, rivers, and streams), as well as artificial reservoirs and canals, are all possible sources of plant pathogens that originate from the surrounding soil or plant debris; through these various avenues, the initial water used for irrigation may be the source of plant pathogens (Hong & Moorman, 2005). The fungus's development and ramification throughout the kernel and the creation of metabolites may directly affect grain quality by changing the composition or metabolism of the grain or making it unfit for humans or animals. According to (Rahil et al., 2013), irrigation intervals did not significantly affect tomato yield production. Wastewater reuse in agriculture provides an additional source of water, nutrients, and organic matter (Vazhacharickal & Gangopadhyay, 2014). The flowers and fruits of many crop plants can readily become infected with fungi growing or existing in the soil. The findings of this study highlight the need for proper treatment and management of the water before its use for irrigation to mitigate the potential risks to food safety and public health (Furqana et al., 2015; A. N. Yadav, 2022). The research aims to analyze the microbial load, Yeast, and Mold from Agra Canal water and soil samples of experimentally irrigated pots, molecular sequencing of fungi, and their impact on crops.

Material Methodology

Sampling Sites

The Agra Canal originates on the right bank of the Yamuna River at Okhla Baraze (Verma et al., 2022). It irrigates about 138,000 hectares of land, mainly in Uttar Pradesh and some areas of Rajasthan (Baba et al., 2018). Agra Canal is a lifeline for irrigation in Utter Pradesh, Haryana, and some parts of Rajasthan. Six sampling sites were selected based on urban and rural areas with agricultural practices to analyze the microbial load. The first three locations are from Faridabad, and the last three are from Palwal district of Haryana State, India. Location first (S1) was the Meetha Pur border (28°29'45.2 "N 77°18'57.4" E), second (S2) Site was BPTP Bridge sector 81 crossing (28°23'22.7 "N 77°20'19.4" E the third (S3) was Fatehpur-Prahlad Pur crossing (28°14'49.7 "N 77°21'09.5" E), fourth (S4) was Basant Garh Palwal,((28°08'40.2 "N 77°21'48.1" E), fifth (S5) was Bela-Ajijabad crossing (28°01'20.4 "N 77°25'48.4" E, and the last point, sixth (S6), was Bansva village (27°53'48.6 "N 77°26'34.3" E) of Palwal District (Figure 1). Sampling sites were selected after the physical survey, and water samples were analyzed for physicochemical parameters to assess irrigation water quality. However, it was found that all the physicochemical parameters were within the permissible limits, and then research moved toward the fungal plant pathogen.

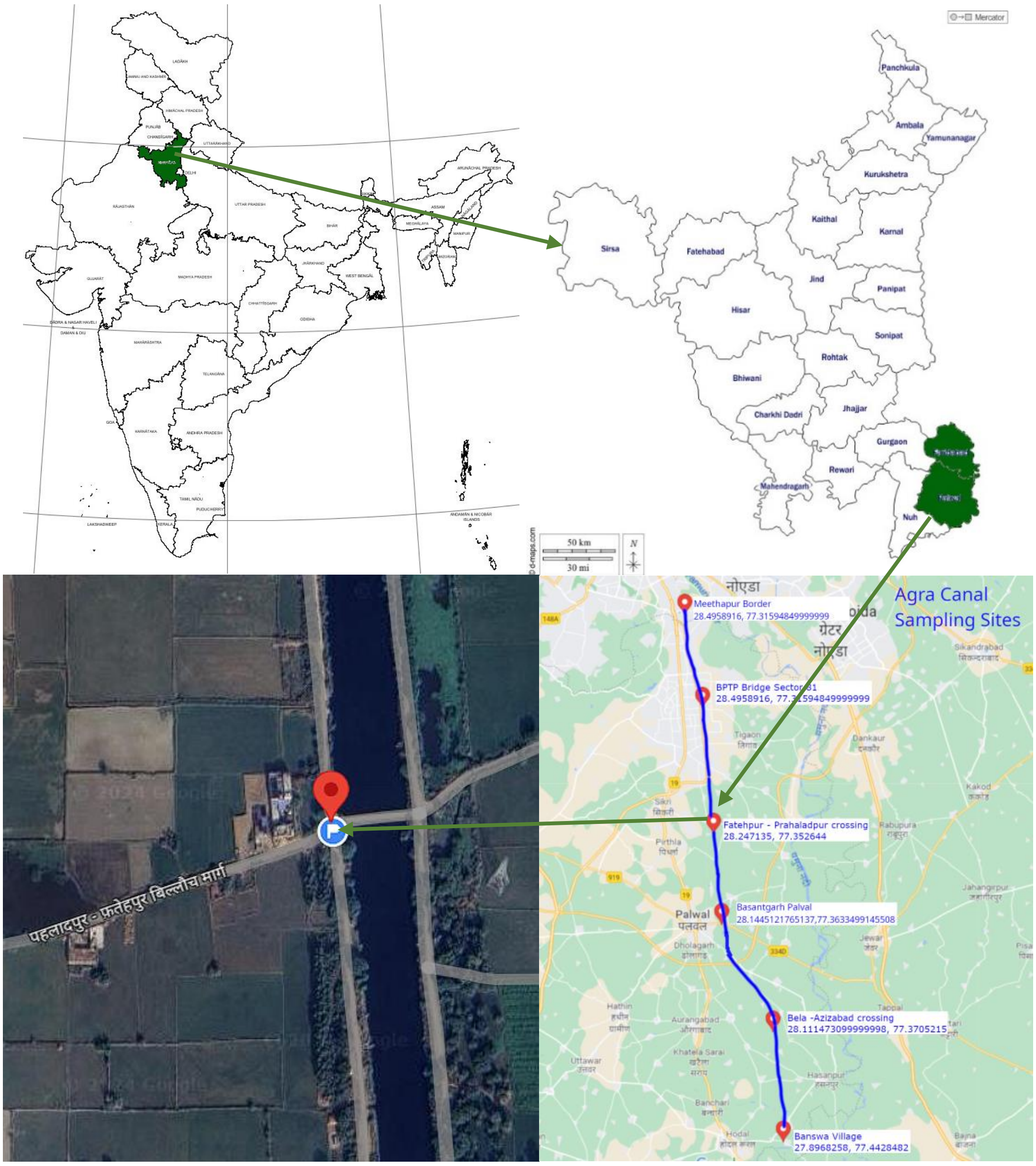


Figure 1 Sampling site of Water sample collection and experimental study (S3).

Experimental Setup

For the experiment, three veggies were selected: Spinach (leafy), tomato (fruit), and radish (root) (Hassan et al., 2024). The soil was taken from the same farm from which soil samples were taken, prepared, and organized in triplicate around the third week of September 2022. Sterilized soil is used to avoid any contamination. Ground Water and canal water for irrigation were also used from the same site (S3). Irrigation was administered to each pot, as per requirement (Petrou et al., 2020). The first set of pots was irrigated with 100% groundwater (GW), the second set with 50% groundwater (GW) + 50% canal water (CW), and the third set with 100% canal water (CW). The pots were kept on the Floor terrace of RPS Palms, sector 88 Greater Faridabad, Haryana, India.

Sample Collection and Analysis

The water samples were collected from six sites along the Agra Canal regularly over one year for three seasons. (2022- 2023). Water samples were collected in a sterilized 250 ml glass bottle, warp the bottle mouth with foil paper kept in the ice box, and sent to the laboratory. Soil samples were collected after the vegetables were harvested and collected in half Kg polythene Ziplock bags (mixed soil from all triplicate pots). The analysis was carried out in an ISO 9001: 2015 certified laboratory. Soil samples were analyzed for Yeast and Mold and the Total plate count. Samples were analyzed using IS 5402 (TPC) and IS 5403 (Yeast and Mold) (S. Pal et al., 2016). Fungi samples for DNA sequencing were isolated after repeated pure culture. Yeast was identified after gram staining, subculture on chromogenic media, and incubated at 25℃. The identification of the fungal cultures was first based on morphological characteristics (Saini et al., 2016). DNA sequencing (Sanger dideoxy method) was used to identify the microbial communities in the samples. For PCR, the QIAMP DNA Mini kit protocol was followed for DNA isolation; denaturation occurred at 94℃ for one min., annealing was done at 48.5℃ for 30 sec., and elongation was done at 72℃ for 1.30 min. Moreover, the final extension was done at 72℃ for 10 min.

Results Discussion

Microbes like bacteria, viruses, and fungi are significant pathogens in irrigation water, which increases the microbial load (Hong & Moorman, 2005). Table 1 presents the microbial load in Agra Canal Water, measured season-wise and location-wise. The data is summarized with each parameter and season's mean and standard deviation (SD).

Table 1 Season-wise and location-wise TPC and Yeast and Mold quantity in Agra Canal Water

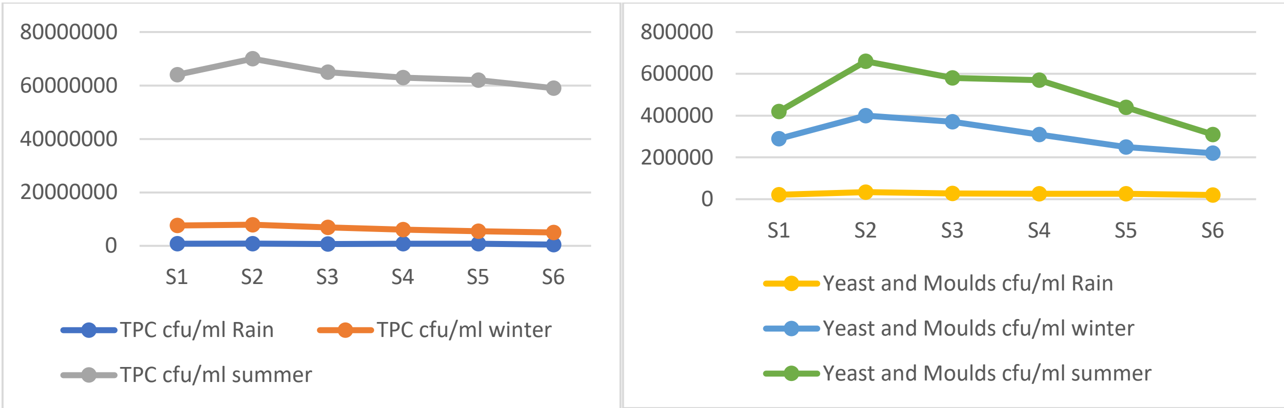
Parameters	Season	**S1**	**S2**	**S3**	**S4**	**S5**	**S6**	**Mean**	**SD**
TPC	Rain	7.40×10^5	8.30×10^5	7.20×10^5	7.60×10^5	7.60×10^5	4.50×10^5	7.10×10^5	1.33×10^5
	winter	7.60×10^6	7.90×10^6	6.90×10^6	6.10×10^6	5.50×10^6	5.00×10^6	6.50×10^6	1.16×10^6
	summer	6.40×10^7	7.00×10^7	6.50×10^7	6.30×10^7	6.20×10^7	5.90×10^7	6.38×10^7	3.66×10^6
Yeast and Moulds	Rain	2.10×10^4	3.40×10^4	2.70×10^4	2.60×10^4	2.60×10^4	2.00×10^4	2.57×10^4	5.01×10^3
	winter	2.90×10^5	4.00×10^5	3.70×10^5	3.10×10^5	2.50×10^5	2.20×10^5	3.07×10^5	6.89×10^5
	summer	4.20×10^5	6.60×10^5	5.80×10^5	5.70×10^5	4.40×10^5	3.10×10^5	4.97×10^5	1.29×10^5

Unit cfu/ml (colony-forming units per millilitre)

TPC estimates the total number of microbial colonies present in the water samples. In the rainy season, the TPC counts ranged from 4.50×10^5 to 8.30×10^5 cfu/ml, with the mean count being

7.10×10^{5} cfu/ml and an SD of 1.33×10^{5} cfu/ml. The Yeast and Mold counts ranged from 2.00×10^{4} to 3.40×10^{4} cfu/ml with a mean count of 2.57×10^{4} and an SD of 5.01×10^{3}. In the Winter Season, the TPC ranged from 5.00×10^{6} to 7.90×10^{6} cfu/ml with a mean value of 6.50×10^{6} cfu/ml and SD 1.16×10^{6} cfu/ml. The Yeast and Mold ranged from 2.20×10^{5} to 4.00×10^{5} cfu/ml. with the mean count was 3.07×10^{5} cfu/ml with SD 6.89×10^{4}. In the Summer Season, the TPC ranged from 5.90×10^{7} to 7.00×10^{7} cfu/ml; the mean count was 6.38×10^{7} with an SD value of 3.66×10^{6} cfu/ml. The Yeast and Mold ranged from 3.10×10^{5} to 6.60×10^{5} with a mean count of 4.97×10^{5} cfu/ml with an SD of 1.29×10^{5}.

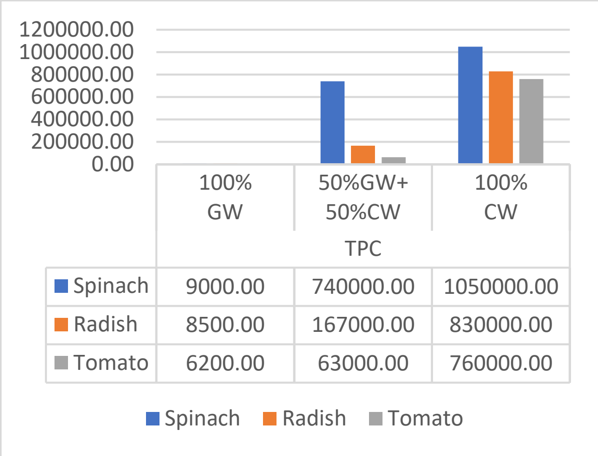
Figure 2 Representation of TPC, Yeast, and Mold Seasonal and location wise



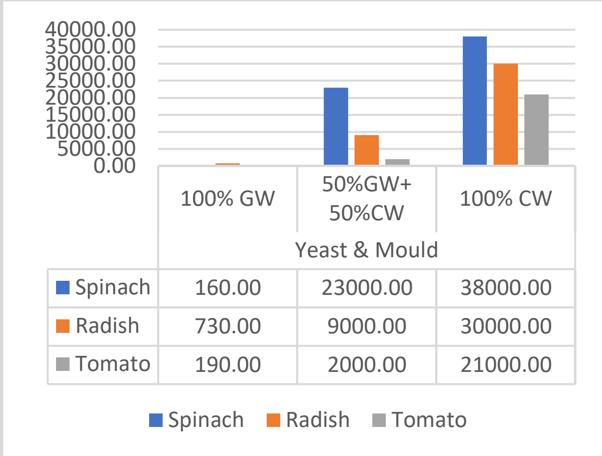
A B

The results from the microbial analysis of the water samples collected from the Agra Canal indicate a significant impact of anthropogenic activities. Microbial counts showed seasonal variation, with the highest levels occurring during summer and winter and the lowest during monsoon; similar results were also obtained by (N. Yadav et al., 2019). The higher microbial counts during the summer (Figure 2A) than in winter seasons can be attributed to the reduced water flow and increased pollutant loading due to sewage dumping (Tripathy & Chowdhry, 2017) and industrial activities (Mondal, 2020). These findings also showed that the microbial and fungal loads were higher at S2, the sites located in the urban area, indicating the significant contribution of anthropogenic activities to the deterioration of water quality and possibly due to colossal sewage dumping between the S1 and S2 sites (Ortiz-Vera et al., 2018). Other study results show that the minimum fungi abundance was in a river with a low concentration of organic matter. However, the highest microbial taxonomic biodiversity was in the waters, abundant in organic matter and rivers and distinguished by higher streamflow quantity. Potentially pathogenic fungi mainly occur in the waters of polluted rivers (Pietryczuk et al., 2018). Concentrations were slightly low at S6 (Figure 2B), which may be due to the absence of sewage source and effluent, and this indicates that the water has a self-clearing capacity when lowing (Verma et al., 2022).

Figure 3 TPC, Yeast, and Mold count in spinach, radish, and Tomato soil irrigated with 100% GW, 50% GW+50% CW irrigated pots.



A



B

Analysis of rhizospheres fungi and their impact on plant health

Root development is enhanced in the presence of water that may be subjected to increased contact with the soilborne inoculum of the pathogens (Lamichhane & Bartoli, 2015). The microbial analysis results reveal the TPC, Yeast and Mold counts have a higher proportion of 100% CW irrigated soil than mixing water irrigated and 100% GW (Fig.3A). TPC for Spinach, radish, and tomato grown with 100% CW were approximately in Spinach 1.5×10^6, in radish 8.3×10^5, and tomato 7.6×10^5 CFU/g, compared to 7.40×10^5, 1.67×10^5 and 6.30×10^4 CFU/g in Spinach (Fig. 3B), radish, and tomato respectively for those grown with 50% GW+50% CW, similar observation obtained by other researchers (Reddy et al., 2017; Sahu & Sirothiya, 2020). The amount of TPC, Yeast, and Mold in 100% GW also indicates that microbes are present in groundwater (Oliveira et al., 2013; Ren et al., 2023). Similarly, the yeast and Mold count for the vegetables grown with 100% CW were around 3.8×10^4, 3.00×10^4, and 2.1×10^4 CFU/g, respectively, while those grown with 50% GW+50% CW were 2.30×10^4, 9.00×10^3, and 2.00×10^3 CFU/g, and least in 100% GW irrigated soil. The Microbial load shows the trend 100% CW> 50% GW+50% CW> 100% GW. These findings suggest that the use of canal water, which is likely more count by various human acts, can lead to a significant increase in the microbial loads of the irrigated crops, potentially be toxic to public health; this statement is supported by (Banach & Van Der Fels-Klerx, 2020). The higher counts observed in the vegetables grown with 100% CW can be attributed to pathogenic and non-pathogenic microorganisms in the canal water, which can affect the plant health, which indicates that the use of canal water can significantly increase the microbial load in the irrigated crops, which have a risk to food safety and public health. As per the Guideline, 1000 cfu/100 mL, even for the strictest standards for unrestricted irrigation (Jeong et al., 2016). Pal et al. found that microorganisms' bacterial and fungal count in vegetables ranges from 8.72-5.08 log cfu/and 6.53-2.3 log cfu/g, respectively (S. Pal et al., 2016). Fungi were identified in rhizosphere soil for further investigation. *Yeast* and *Candida* spp. were found in 100% GW irrigated vegetables (spinach, tomato, and radish); these are non-pathogenic fungi in plants.

In spinach-mixed water, 50% GW+50% CW irrigated soil, *Fusarium equiseti*, and *Aspergillus fumigatus* were present. Spinach soil, rinsed with 100% canal water, contained *Alternaria spp.*, and uncultured fungus clones dominated. In contrast, other fungus species such as *Aspergillus fumigatus*, *Albifimbria verrucaria*, *Chaetomiaceae spp.*, *Aspergillus niger*, *Fusarium equiseti*, were also present.

In tomatoes that were irrigated with 50%GW+50%CW, *Chaetomiaceae spp.*, *Aspergillus niger* was dominating. (Bandh et al., 2011), obtained *Aspergillus* spp., dominating in Dal Lake Kasmir. In tomato soil rinsed with 100% canal water, *Fusarium equiseti, Alternaria spp., and Aspergillus niger* were dominating; other fungal species, *Humicola spp., Aspergillus novafumigatus, Alternaria spp., Fusarium equiseti and Albifimbria verrucaria* also obtained. Other researchers also obtained dominant fungi *Fusarium* and *Scolecobasidium humicola* in tomato and cabbage (Mahmoud & Narisawa, 2013). Tomato and spinach's best growth was obtained in 50% GW+50%; it may be due to the mixing of water, which is a balanced amount of nutrition (Ayers et al., 1985). Fungi can be less effective in the best available nutritional conditions, and there were fewer fungi in mixed water than in the 100% CW irrigated plants, although various climatic factors affect the fungal activity.

Radish soil was irrigated with 50% GW+50% CW, and the irrigated area was dominated by *Aspergillus novafumigatus*. At the same time, *Chaetomiaceae spp.* and *Fusarium equiseti* were also identified. Radish soil with 100% CW contained *Humicola spp.* and *Fusarium equiseti* dominatingly, while *Aspergillus novafumigatus*, *Albifimbria verrucaria*, *Chaetomiaceae spp., Alternaria spp.*, *Aspergillus fumigatus*, were present. Radish's best growth and yield were obtained in 100% GW irrigated pots, possibly due to tuber crops being more sensitive to polluted water and without pathogenic fungi. All the obtained rhizospheric fungus also dominated the Agra Canal water.

Other fungal species, such as *Cladosporium cladosporioides*, *Rhizopus spp.*, *Myrothecium verrucaria*, *Trichoderma album*, *Verticillium spp.*, *Cladosporium herbarum*, *Penicillium spp.*, *Chrysosporium indicum*, *Mucor recemosus*, *Penicillium oxalicum*, *Aspergillus sclerotioru*, *Aspergillus candidus*, *Aspergillus flavus*, *A.nidulans*, *A.sydowi*, *Curvularia lunata*, *Mucor recemosus*, *Alternaria alternata*, *Aspergillus niger*, *A.ochraceous*, *A. terreus*, *Penicillium oxalicum*, *Phoma sorghina*, *Synecephalastram racemosum*, *Trichoderma harzianum*, *T. koningi*, *Trichothecium roseum*, *Verticillium alboatrum* and *V. lilacinus*, *Chaetomium globosum*, and *Colletotrichum species*, and others present in Yamuna River, (Tripathy & Chowdhry, 2017) reported 704 different fungal colonies, of which 26 different genera of fungi and 54 different species were identified in the Delhi NCR region, but *Humicola* spp. was not detected in the Yamuna River water by (Tripathy & Chowdhry, 2017). The activity of fungus depends on many factors, such as pH, temperature, irrigation timings and methods, crop rotation, seed quality, the timing of harvesting, availability of nutrients, and environmental factors (Awuchi et al., 2021). The impact of Agra Canal water on crop quality may vary depending on the water composition. According to (Hong and Moorman, 2005), an entire crop can be lost if the water carries undesirable chemicals or microbes, so other chemicals and microbes may also impact crop yields.

Other researchers also obtained some similar fungi from wastewater (Becker & Shaw, 1955). *Pythium, Rhizopus, Rhizoctonia, Aspergillus*, *and Fusarium* were the most frequently isolated genera in irrigation water (Suliman, 2022). Additionally, some fungi in Yamuna River water, such as *Cladosporium cladosporioides,* cause leaf spots and Mold in crops; it is a cosmopolitan fungus that causes leaf and fruit spots (Farooq, 2017). It also can cause discoloration and deterioration of plant tissues, while Trichoderma species can compete with beneficial microorganisms in the soil, potentially disrupting the plant's rhizosphere (Jiménez-García et al., 2018). All plant parts may be subject to additional infection from fungi added to the soil in sewage-polluted waters used as irrigation supplements (Cooke & Kabler, 1957). Such fungi can transfer to edible parts of vegetables and can cause serious health issues in animals and humans. Agra canal water also causes skin infections in farmers.

Most fungi are phytopathogens that infect the crops in the field (Kehri et al., 2019). Some fungi act as plant pathogens, causing severe crop losses and postharvest food spoilage (Bandh et al., 2011). Yeast is a plant nonpathogenic fungus (Riesute et al., 2021). The Yeast has a positive impact on increasing vegetative growth (number of plant leaf area, fresh and dry weight of biomass in plants), which may be due to many essential nutrient elements (N, P, and K) present in Yeast (Natsheh et al., 2022). Some *yeasts* have been reported to multiply rapidly and abundantly in the presence of pollution and to be predominant in grossly polluted waters. *Candida spp.* was present in reasonably polluted water and absent in badly polluted water (Tan & Lim, 1984). *Chaetomiaceae spp.* causes seed mold and decreases corn's leaf spot (Soytong et al., 2021), but according to (Hong & Moorman, 2005), It is Present in a water reservoir, and the effect is not specified. *Chaetomiaceae spp* is one of the largest genera of saprophytic ascomycetes. Chaetomium is a biocontrol agent because it is a potent antagonist for various plant pathogens (Soytong et al., 2021).

Albifimbria verrucaria, formerly it was *Myrothecium verrucaria,* has an herbicidal effect on *Conyza canadensis* by reducing the inflorescence growth and necrosis in leaves (Hoagland et al., 2023). It is a phytopathogenic fungus that causes necrotic leaf spots and plant collapse in leafy vegetables, including Spinach and ornamental hosts; it can cause 33 types of leaf necrosis and plant decay, which is found in 34 crop vegetables from Italy (Matić et al., 2019). *Albifimbria verrucaria* strain, a causal agent of several plant diseases, produces mycotoxins trichothocenes, which are capable of exerting a cytotoxic effect on animal cells such that they may interfere with protein synthesis, the immune system, and alveolar macrophages, infection in human caused by the plant matter (Moreno-Flores et al., 2020).

Fusarium spp. is a plant-pathogenic fungus (Arie, 2019); it presents in effluent, well, stream, pond, ebb and flow, nutrient film, and harms the fruit, vegetable, and ornamental plants (Hong & Moorman, 2005; Hussain et al., 2022). The presence of certain fungi, such as *Aspergillus* and *Fusarium spp.,* in vegetables irrigated with contaminated canal water, is a significant concern for human health. On the other hand, *Fusarium equisetum* is an emerging human pathogen that can cause localized or disseminated infections, particularly in immunocompromised patients, and cause wilt in crops like tomato, potato, and spinach. It is present in the rhizosphere of plants irrigated with canal water, which may cause infections due to the development of antifungal resistance in *Fusarium spp.* (Arie, 2019; Sáenz et al., 2020).

Alternaria spp. is present in canal water and effluents and degrades fruits and vegetables (Hong & Moorman, 2005). According to (Mourou et al., 2023), it is the most virulent plant pathogen. *Alternaria* and *Cladosporium* species are ubiquitous fungi that can cause diseases in various crops and reduce produce quality and shelf life. *Alternaria spp.* causes leaf spots in crops, seeds, fruit, and vegetables to decay. Uncultured fungi could potentially have unknown effects on plant health and productivity (Abarenkov et al., 2022).

According to (Xu et al., 2021), *Aspergillus flavus* contamination usually begins during crop development when the crop is associated with aflatoxin production, a toxic and carcinogenic group. *Aspergillus spp.,* notably *A. fumigatus* and *A. novafumigatus* are known to be opportunistic human pathogens that can cause a range of respiratory infections, such as aspergillosis, in immunocompromised individuals (Loreto & Tondolo, 2019). Inhalation of *Aspergillus* spores can lead to allergic reactions, hypersensitivity pneumonitis, and invasive infections in susceptible populations (Awuchi et al., 2021). *Aspergillus spp.* is in the water due to the discharge of organic effluents. *Aspergillus niger* is known to produce ochratoxin A and other mycotoxins, which are harmful to plants, animals, and humans. According to (Prasad & Chaudhar, 1974), *Aspergillus novafumigatus* is known to produce gliotoxin, an immunosuppressive compound commonly found in soil, water, and air, and produces mycotoxins that can harm living beings. The presence of these pathogenic and toxigenic fungi in the canal water and the rhizosphere of the irrigated plants is a serious concern, as it can lead to the contamination of the agricultural produce, posing a threat to food safety and public health, (Nagar et al., 2020; A. N. Yadav, 2021). Consuming vegetables contaminated with these pathogenic fungi impacts the health of the immune systems, such as the elderly, the very young, and those with underlying medical conditions. The findings of this study highlight the need for stringent water quality monitoring and treatment of the Agra Canal water before its use for irrigation purposes.

Remedial measures for removal of pathogenic fungus from canal water

A good quality water is required for sustainable irrigation. Crop irrigation with sewage-mixed or otherwise contaminated water can lead to the proliferation of plant-pathogenic fungi, resulting in reduced crop yields; various researchers applied different techniques. According to Pal, multiple factors affect the growth of fungus in irrigation water, such as Irrigation source, and water quality can worsen after periods of heavy rainfall or flooding if water has been in contact with other sources of contamination (S. Pal et al., 2016). The irrigation regime and the amount of water used for irrigation influence the availability of niches where pathogens may persist or multiply under field and postharvest conditions (S. Pal et al., 2016). Researchers used mycofilteation to remove the pathogenic fungus from irrigation water (Mnkandla & Otomo, 2021), which was effective and improved plant health. Jeon says the filtration method may effectively remove zoospores, but it depends on the filter media used in greenhouse settings. They concluded that an Iron Oxide coated sand filter (IOCS) was most effective in removing the spore of Pythium and reducing the root rot; in other filters, an activated carbon filter was also used by Jeon in greenhouse conditions; however, more research is needed in this segment (Jeon et al., 2021) Researchers also proposed treating fungi-contaminated water using advanced techniques like peroxyacetic acid, UV, and filtration, which are eco-friendly and more effective in reducing the microbiological hazards in the water. (Castillo et al., 2013; FAO & WHO, 2023; Tiru et al., 2021). UV treatment can inactivate fungal spores by damaging their DNA, thus preventing them from growing and spreading. UV systems are commonly used in water treatment to ensure that irrigation water is pathogens-free (Mnkandla & Otomo, 2021). Researchers used peroxyacetic acid, a green chemistry solution that effectively eliminates microbiological contaminants in protected horticulture settings (Carrasco & Urrestarazu, 2010). Importantly, this method is more environmentally friendly than traditional chemical disinfection techniques (FAO & WHO, 2023). In chemical treatment, fungicides like etridiazole can treat irrigation water. These chemicals effectively reduce the presence of fungal pathogens and prevent their spread to crops (Jeon et al., 2021).

According to (Castillo et al., 2013), biological control strategy involves antagonistic microorganisms, such as *Bacillus spp*. Various bacteria are recognized as highly effective biocontrol agents because they inhibit the growth of different plant pathogens, including fungal genera like *Pythium, Rhizoctonia, Fusarium,* and *Phytophthora* (Castillo et al., 2013). Using *Bacillus spp.* as a biocontrol agent against various soil pathogens has reported positive responses through observing a negative effect on pathogen growth, for example, against *Alternaria sauce* and *Rhizoctonia solani*, foliage and soil pathogens, respectively, some impact on pathogen mycelia inhibition by the action of *Bacillus*, up to 50% compared to treatment control. Furthermore, in the case of *Aspergillus dauci,* greater control was observed with biocontrol agents compared to chemical treatment. (Castillo et al., 2013; Ntushelo et al., 2019). Another study showed that using crude plant extracts from some Oaxacan flora could inhibit the mycelial growth of two phytopathogenic fungi, *Fusarium oxysporum* and *Penicillium expansum* (Suheri et al., 2014). The natural bioactive compounds from plants could be an effective and eco-friendly alternative to synthetic fungicides. Bacillus spp. Microsphaeropsis spp. *Trichoderma harzianum* is a microbial species identified as an antagonist of *Fusarium species* through the mechanism of antibiosis elicited by a wide range of antifungal metabolites, including antibiotics (Tiru et al., 2021). Appropriate irrigation water management should be one of the integrated disease management practices to prevent seedling infections and losses (Marčiulynas et al., 2020). Regular monitoring of irrigation water for fungal pathogens can help in early detection and management. Using molecular identification methods, such as metagenomic approaches, can provide detailed insights into the diversity and presence of fungal pathogens in irrigation water (Calderon et al., 2023). In conclusion, while various treatment options exist to mitigate fungal pathogens in irrigation water, selecting and implementing the most appropriate approach will depend on factors such as water source, crop type, and local resource availability.

Conclusion and Recommendations

In conclusion, the reduced production of crops and vegetables is primarily due to microbial contamination, particularly fungi, in the Agra Canal's irrigation water. The highest levels of Total Plate Count and Yeast and Mold were found at the S2 location. In contrast, the lowest levels were observed at the S6 location, indicating that the microbial load is primarily attributable to sewage and effluent dumping near the S2 site. The highest value was obtained in Summer> winter> monsoon. In pot results, the highest load obtained was 100% CW > 50% GW +50% CW >100% GW, showing 100% CW having more fungus load and decreasing crop yield. Various plant pathogenic fungi obtained in the rhizosphere soil of spinach, radish, and tomato may impact the crop quality. Adopting best management practices and implementing strict water quality monitoring can help address the issue of microbial contamination in the Agra Canal and ensure the safety of the agricultural produce. There is a need to reduce the sewage and effluents pouring into the Agra Canal stream to improve the yielding of crops and vegetables and adopt suitable remedial measures to enhance crop yielding.

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Abbreviations: Groundwater (GW), Canal water (CW), Total Plate Count (TPC), Species (Spp.), Minutes (min.), Seconds (sec.).

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