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Decontamination of enteric pathogens in soil ecosystems irrigated with low quality water for continuous irrigation practice

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Abstract

Recently, management of soil irrigated with low-quality water has become necessary to have a healthy crop. This research work aims to design the best management practices (BMP) to minimize drainage water hazards in the soil ecosystems. Based on the source of irrigation water, a column experiment was implemented on three soil samples that collected from three governorate in Egypt. The subsistence of enteric pathogens in soil irrigated with three types of low-quality water either sole sewage effluent (Giza) or a mix of drainage and industrial effluent (Kafr-el-sheikh) or drainage effluent (Sinai) was monitored periodically for 90 days. The trailed soils were divided as: non inoculated cultivated (C) or inoculated cultivated with sole phosphate dissolving bacteria (T1) or *Acidithiobacillus sp*. (T2) or with a combination of both microorganisms (T3). Three common hyperaccumulator plants (*Brassica napus*, *Plantago psyllium*, and *Plantago major) were cultivated separately in cultivated, inoculated soil trials* in comparison to non-cultivated, non-inoculated control treatments (NC). Results section illustrates the removal pattern of fecal coliforms and *Salmonella* sp. in the trailed soil ecosystems, in response to different treatments during 90 days of experimental monitoring. The trailed remediation amendments, either in the single or combined application, followed by phytoremediation with three different phytoremediation plants, exhibited a positive effect in diminishing pathogenic bacteria in the three tested soil ecosystems, yet at varying degrees. The study concludes that, applied mixture of all treatments represented by choice of *Plantago psyllium* (as best phytoremediator plant) and combination of two remeditative bacterial inoculums (*Acidithiobacillus* and phosphate dissolving bacteria) in contaminated soil was selected as the BMP among the other applied treatments.

1. Introduction

Water is a restricted indispensable resource for agriculture, industry, and human existence. In arid and semi-arid regions, where water resources are inadequate, challenges for achieving the highest possible water use efficiency are not that easy. In association with the increased population and rise in economic and social activities, the anxiety on water resources availability is getting higher. As consequence, decision-makers have adopted several planning tools to secure water allocation and distribution

Low quality water defined as water streams that contains measurable quantities of chemical substances, salts, heavy metals, and harmful microbes that may have direct or indirect effect on crop yield production or soil fertility characteristics. Irrigation waters usually deliver substances from its environment and sometimes it is mixed with waste products of human's activities such as domestic or industrial effluents. These substances usually loaded with organic and inorganic pollutants as well as high intensities of bacterial population and coliforms which existence is deteriorative to human and animal health FAO (2007).

The intense application of low-quality waters for example, sewage effluents, drainage water and industrial waters in Egyptian farming is a decades-old practice that is nowadays gaining increased attention with the depletion of freshwater resources particularly in arid and semiarid countries. Certainly, irrigation with low quality water represents a major threat to public health and environmental quality reported by Saber (2007).

Low quality water had been involved as an important cause of health risk especially for chronic, abdominal, and intestinal diseases as well as eruption of other acute diseases especially when applied in irrigation lacking adequate treatment

(WHO, 2006; Hassanain et al., 2021). The probable transfer of living pathogens from polluted soil ecosystems to human food or drink is of great concern in Egypt because of the existence of broad types of pathogens in the polluted low-quality water and soil ecosystems (Kabary et al., 2021a). Another source of transmission mainly due to, the widespread of machine-less labor in farming which based on farmers who are having a close contact with pathogens and/or possess relatively low standards of hygiene (NygÍrd et al., 2008).

It is worthy to mention that one of the three main arms of Egyptian water strategies is the reuse of treated low water quality in farming. Hence, it is expected that Egyptian farmers might receive in the very near future less irrigation water both in quantity and quality (Saber et al., 2015). low quality water is usually polluted with enteric pathogens, organic toxins as well as certain potential toxic elements that pose an environmental hazard to both human and living biota.

The main target of the current work was to explore the action of certain remediative amendments applied to soil ecosystems irrigation with low quality water on the survival of enteric pathogens such ecosystems for extended periods and to reach the optimum management practices that should be used to minimize such hazards.

2. Material and Methods

2.1. Soil sampling

Samples from three different soil areas (Fig. 1) were subjected to irrigation with low quality waters either single, mixed sewage effluent or agriculture drainage water. The 1st soil sample was collected from Sinai Governorate where the soil ecosystem was irrigated for 33 years with mixed Nile and drainage water of El-Salam canal. The 2nd soil sample was collected from northern Delta (Kafr-el-Sheikh Governorate) where the soil ecosystem was irrigated for eighty years with drainage water combined with sewage and industrial effluents from Kitchener drain and the 3rd soil sample was collected from Giza Governorate (Kombera site) where the soil was irrigated for thirty-five years with sewage effluent from El-Libeny drain. Some physical and chemical characterizations of used soils are presented in Tables 1 and 2.

Fig. 1. Location of the three soil samples collected from three Governorates

Table 1
Chemical characteristics of soil samples collected from different locations (oven dry basis)

Location	pH	EC dse.m^{-1}	OM %	Land use	Particle size distribution %					
					Coarse	Fine	Total sand	Silt	Clay	Texture
Konbora	7.53	0.61	3.59	Dill plants	1.4	42.4	43.8	19.2	38	CL
Sinai	7.79	2.7	0.59	Beet	3.5	41.2	44.7	31.6	23.7	loam
Kafr El-Sheik	8.16	2.1	1.96	Common bean	1.3	13.4	14.7	34.1	51.2	Clay

Table 2
Heavy metals analysis of irrigation water samples collected from different locations

Location	Ni	Cu	Zn	Cd	Pb	Cr	Mn	Fe
Safe level	0.2	0.2	2.0	0.01	5.0	0.1	0.2	5.0
Nile water	–	0.01	0.02	nd	nd	0.01	–	–
Sinai-El-Salam Canal	0.066	0.055	0.645	0.08	1.71	0.061	0.612	2.5
Kafr-El-Sheik	0.24	0.4	2.315	0.04	nd	0.06	0.820	0.52
Konbera (Giza)	0.15	0.5	3.4	0.05	3.0	0.35	2.5	0.15

2.2. Treatments' preparation

Twenty four kilograms from each of the soil samples, were collected separately and packaged in ten plastic columns with a diameter of 30 cm, a height of one meter and an active soil height of 90 cm. Five treatments in three replicates were trailed including, non-cultivated control soil (NC), cultivated control soil (C), sulfur treated soil with 1.25 ton/fed and inoculated with *Thiobacillus* sp. (T_1), soil treated with probentonite (bentonite to rock phosphate ratio 1:1) and inoculated with phosphate dissolving bacteria (PDB) at the rate of 1.25 ton/fed (T_2), and soil treated with a mixture of the aforementioned remediative amendments (T_3).

The moisture content of the soil in each column was adjusted with any of the three sources of low-quality water to fifty percent of the soil field capacity and kept at this rate during the experiment by continuous irrigation with the same source. Three seeds of the hyperaccumulator canola plant *(Brassica napus)* inoculated with Arbuscular mycorrhiza AM conidia were sown in each cultivated column. Parallel to the trials, the same treatments were replicated in a pot experiment using two other hyperaccumulator plants, i.e., plantains (*Plantago psyllium)* and blond psyllium (*Plantago major)*. Surface soil samples were collected from each treatment initially and after 30, 60 and 90 days to track the survival of soil pathogens.

2.3. Microbiological Methods

2.3.1. Cultivation and Fortification

All microorganisms used in the bioremediation trails were cultivated in Bioflo and Celligen bioreactor, each in its specific medium, until reach 10^6 CFU. Prior to experimental setup, mixed cultures of (*Bacillus megaterium, Bacillus subtilis, Bacillus cereus, Pseudomonas putida, Pseudomonas fluorescens*) were prepared in nutrient broth medium and mixed equally to serve as Phosphate dissolving bacterial inoculum. On the other hand, *Thiobacillus* cultures were prepared in elemental sulfur salt medium (Staley et al., 1989) for inoculation. Bacterial cultures were obtained from Agricultural Microbiology Department culture collection, National Research Centre, Cairo, Egypt.

The microbial cultures were impregnated on a proper mordant at the rate of 20 ml microbial suspension per 100 g mordant, oven-dried soil. Initially, each inoculated soil column received 100 g mordant inoculated with the targeted microorganisms.

2.3.2. Fecal coliforms

Fifty-gram portion of each soil sample were mixed with 450 ml sterile physiological saline water in sterile blender jar and blended for 1 to 2 min at low speed (8000 rpm) to prepare homogenous slurry. Three different ten-fold decimal dilutions were prepared, and fecal coliforms (per 100 ml) were enumerated by most probable number (MPN) technique according to APHA (1995).

2.3.3. *Salmonella* sp.

1 ml from the soil dilutions 10^{-1}, 10^{-2} and 10^{-3} of each soil sample was inoculated onto SS media for the detection of *Salmonella*. All tubes were incubated at 37°C for 48 hr. *Salmonella* sp. were identified by culture characters, morphological characters, and biochemical reactions according to the scheme illustrated by Quinn et al. (2002).

3. Results

3.1. Fecal Coliforms

Results given in Table 3 and Fig. 2 represent the intensities of fecal coliforms in the controls and remediated soil ecosystems collected from Kafr-el-Sheikh, Giza and North Sinai governorates. Results indicated that the initial intensities of fecal coliforms varied in the different experimented soils, reaching a maximum of 2200 CFU g^{-1} in the soil ecosystem irrigated with sewage effluent in Giza governorate and a minimum of 40 CFU g^{-1} in the soil ecosystem irrigated with drainage water at Sinai governorate. Varied behaviors were displayed by fecal coliforms towards the different treatments. The treatment with a combined mixture of the different remediative amendments was generally more effective in reducing the intensities of fecal coliforms in the different soil ecosystems. Yet, the differences in the efficacy of the combined mixture of all trailed remediative amendments (T3) and that of either rock phosphate or elemental sulfur (T1, T2) were not that sensible. As example, In the case of *Brassica napus* trails, combined mixture of all remediative trials (T3) shows decreasing in fecal coliform count (80, 40 and 4) in comparison to sulfur trails T2 (40, 40 and 6) and rock phosphate trails (T1) (120, 36 and 4). In the non-cultivated control trails, fecal coliform survived, and their intensities were consistent in the first month by the extra fecal bacte-

Table 3
MPN of fecal coliforms in soil ecosystems irrigated with different low quality water under bioremediation and phytoremediation trails in the green house (CFU g^{-1})

Treatments		Sewage effluent (Giza)				Mixed drainage water and sewage and industrial effluent (Kafr-El-Sheikh)				Drainage water (Sinai)			
		Time in days											
Bioremediation treatments	Phytoremediation	0	30	60	90	0	30	60	90	0	30	60	90
Non-cultivated Control (NC)	*Brassica napus*	2200	2200	1140	960	660	660	340	280	40	14	28	8
Cultivated Control (C)		2200	5000	940	520	660	460	70	40	40	30	36	4
Rock phosphate (T1)		2200	4400	540	120	660	40	20	36	40	10	8	4
Elemental sulfur (T2)		2200	5600	440	40	660	28	40	40	40	22	16	6
Combined mixture (T3)		2200	440	200	80	660	22	36	40	40	11	13	4
Non-cultivated Control (NC)	*Plantago major*	2200	2200	1140	960	660	660	340	280	40	14	28	8
Cultivated Control (C)		2200	440	600	40	660	40	40	0	40	30	30	4
Rock phosphate (T1)		2200	660	390	136	660	0	0	0	40	36	22	0
Elemental sulfur (T2)		2200	136	40	26	660	0	0	0	40	3	4	0
Combined mixture (T3)		2200	660	30	28	660	0	0	0	40	4	2	0
Non-cultivated Control (NC)	*Plantago psyllium*	2200	2200	1140	960	660	540	340	280	40	14	28	8
Cultivated Control (C)		2200	4800	800	280	660	90	22	0	40	36	40	36
Rock phosphate (T1)		2200	440	236	34	660	40	40	0	40	30	40	0
Elemental sulfur (T2)		2200	122	220	44	660	0	0	0	40	40	36	8
Combined mixture (T3)		2200	40	38	36	660	0	0	0	40	10	9	8

ria added with the water stream, followed by a slight decrease in day 60 till the end of the experimental period. As example, Giza non cultivated control soil trail, the bacterial intensities in days 30, 60 and 90 of the experiment were 2200, 1140 and 960 CFU g^{-1}.

Although, fecal coliforms follow similar consistency pattern in the control cultivated soil with no added amendments, there were a further decrease in total fecal count compared to the non-cultivated trials. Applying the same *Brassica napus* example in Giza soil, the decreasing rate of fecal bacterial count in day 30, 60 and 90 were 5000, 940 and 520 CFU g^{-1}. Furthermore, results in Table 3 and Fig. 2 also showed that, fecal coliforms did not totally disappear from the soil ecosystems after three months under *Brassica napus* treatments in all soils trails. Meanwhile, they were entirely undetected in case of Kafr-el-Sheikh and Sinai phytoremediated soils with either *Plantago major* or *Plantago psyllium* particularly when proceeded with bioremediation with either (T_1, T_2 or T_3).

Generally, the disappearance of fecal coliforms in the different soil under study, was achieved in the soil trials collected from Sinai governorate (irrigated with mixed drainage water) and Kafr-el-Sheikh governorate (irrigated with mixed drainage water and effluents) which may be explained by natural low intensities of these bacteria in the irrigation water sources. In contrary, complete decontamination of fecal microbes hadn't achieved in Giza governorate (irrigated with sewage effluent) that might ascribed to the higher fecal coliform intensity of the irrigation water used.

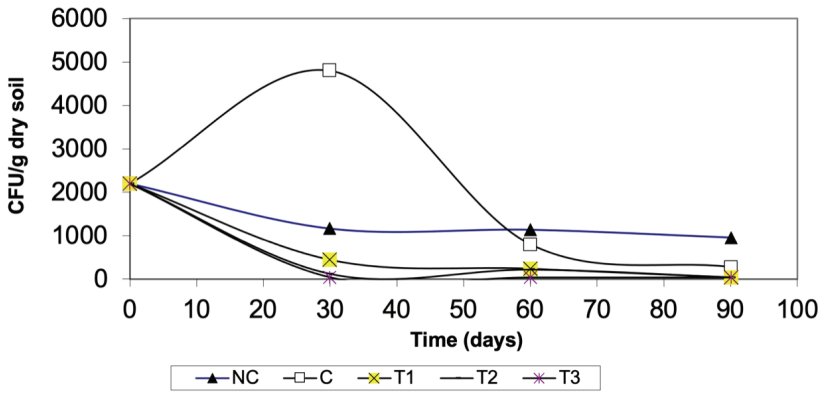

Fig. 2. Kinetics of fecal coliforms appeared in sewage soil cultivated with *Plantago psyllium* under different treatments applied. (Non-Cultivated control (NC), Cultivated control (C), cultivated inoculated with phosphate dissolving bacteria (T1), cultivated inoculated with *Acidithiobacillus* (T2) and cultivated mixed inoculated the two cultures (T3)

3.2. *Salmonella* sp.

Results given in Table 4 and Fig. 3 represent the existence and behavior of *Salmonella* sp. in three trailed soil ecosystems after different treatments. Results indicated that the *Salmonella* sp. was initially detected in both soil ecosystems irrigated with sewage effluent (Giza governorate) or with mixed drainage water and sewage and industrial effluents (Kafr-el-Sheikh governorate). However, *Salmonella* sp. was totally absent in the soil ecosystem irrigated with mixed drainage water from Sinai governorate, this result may be due to increasing of soil salinity causing a lethal osmotic pressure to *Salmonella* cells, illustrated in Table 1 and 2.

The response of *Salmonella* sp. to the various remediation treatments varied according to the soil source. In Kafr-el-Sheikh soil, *Salmonella* sp. proliferated in both control soils, whether cultivated on non-cultivated, at a higher rate in the former, and was detectable till the end of 90 days experiment period (96 and 67 CFU g^{-1}) in *Brassica napus* cultivated trials.

On the other hand, remediation with either the single or combined tested remediative amendment inhibited the growth of *Salmonella* sp. in the soil ecosystem, which disappeared totally after one month in soils remediated with rock phosphate, elemental Sulfur, or mixture of the trailed remediative amendments with the action of the three tested Phyto accumulator plants. The behavior of *Salmonella* sp. in the soil irrigated with

Table 4
MPN of *Salmonella* sp. in soil ecosystems irrigated with different low quality water under bioremediation and phytoremediation trails in the green house (CFU g^{-1})

Treatments		Sewage effluent (Giza)				Mixed drainage water and sewage and industrial effluent (Kafr-El-Sheikh)				Drainage water (Sinai)			
		Time in days											
Bioremediation	Phytoremediation	0	30	60	90	0	30	60	90	0	30	60	90
Non-cultivated Control (NC)	*Brassica napus*	300	250	200	170	200	190	140	96	0	0	0	0
Cultivated Control (C)		300	350	200	180	200	220	80	67	0	0	0	0
Rock phosphate (T1)		300	100	50	40	200	0	0	0	0	0	0	0
Elemental sulfur (T2)		300	90	50	40	200	0	0	0	0	0	0	0
Combined mixture (T3)		300	150	50	20	200	0	0	0	0	0	0	0
Non-cultivated Control (NC)	*Plantago major*	300	250	200	180	200	190	140	96	0	0	0	0
Cultivated Control (C)		300	220	190	200	200	100	40	0	0	0	0	0
Rock phosphate (T1)		300	80	40	15	200	0	0	0	0	0	0	0
Elemental sulfur (T2)		300	60	40	20	200	0	0	0	0	0	0	0
Combined mixture (T3)		300	40	30	19	200	0	0	0	0	0	0	0
Non-cultivated Control (NC)	*Plantago psyllium*	300	250	200	180	200	190	140	96	0	0	0	0
Cultivated Control (C)		300	380	80	20	200	60	40	0	0	0	0	0
Rock phosphate (T1)		300	110	40	30	200	0	0	0	0	0	0	0
Elemental sulfur (T2)		300	90	20	25	200	0	0	0	0	0	0	0
Combined mixture (T3)		300	90	19	17	200	0	0	0	0	0	0	0

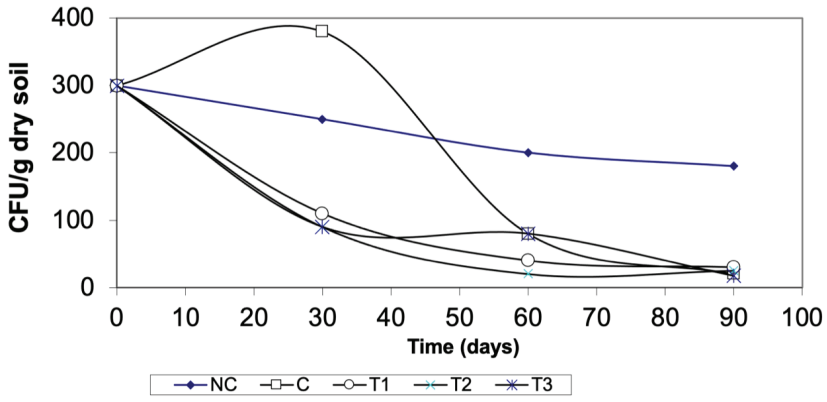

Fig. 3. Kinetics of MPN of *Salmonella* sp. in soil irrigated with sewage LQW cultivated by *Plantago psyllium* under different treatments. (Non-Cultivated control (NC), Cultivated control (C), cultivated inoculated with phosphate dissolving bacteria (T1), cultivated inoculated with *Acidithiobacillus* (T2) and cultivated mixed inoculated the two cultures (T3)

sewage effluent (Giza) was different. The bacteria were detectable, at varied intensities, under the various remediation treatments however, the intensities of *Salmonella* sp. in control soil were always higher. At the end of the experiment, *Salmonella* count reached 40, 40 and 20 CFU g^{-1} without achieving complete decontamination, which may be explained by high intensities of *Salmonella* already existed in the irrigation water source.

3.3. Kinetic study of fecal coliforms retention in soil ecosystems irrigated with different low quality water as affected by remediation amendments

Results in Table 5 represent the efficiency of remediation material in decontamination of soil applied in three soils ecosystem, using kinetic approach and modified Freundlich equation in the form:

$q = bt^a$, the linear form of the ***model is log q = b + a log t***

Where:
q: the rate of fecal coliforms retention in treated or untreated soil
t: Time (days)
a and b: constants

Increasing of coefficient of determination and Standard error meaning the succession of model to describe the kinetic data. The model is the best empirical models used in describing the kinetic data under Egyptian conditions (Saber et al., 2016). This model as shown high coefficient of determination R^2 and low standard error SE, meaning high efficiency in describing the phenomenon. Results in table imply application of T3 was the best treatment in decreasing the rate of fecal coliform retention in treated soils. The numerical values of rate constant *a* in all locations studied were the lost values compared to other treatments applied or even control cultivated (C) and non-cultivated (NC). For example, in Kafr El-Sheik, the (a) constant of T2 was -2.11, by application T3, the rate constant decreased to -2.26, means that the rate of fecal chloroform presence was significantly decreased in treated soil by application T3. It should be mentioned that the same trend was also observed in other locations studied for the same treatment.

4. Discussion

Miscellaneous types of pathogens constantly exist in the soil ecosystems irrigated with low quality water for extended periods. Most common diseases in the world are caused by water and food borne pathogens. For instance, cholera which caused by bacterium (*Vibrio cholera*), diarrhea, dysentery that caused by *Escherichia coli* bacterial infection, typhoid due to *Salmonella typhi* infection. Most of these microbial pathogens have known to cause dangerous diseases that ultimately leading to morbidity and mortality in developing countries. Liu et al., 2013a stated that, about 88% of diarrhea disease is related to unsafe water supply and hygiene practice. Globally, recent reports indicated that higher than 1.3 million children's deaths are due to diarrhea every year (Liu et al., 2013a; Ma et al., 2014). The main microbial pathogens coupled with farming with low quality water are salmonellosis (*Salmonella* sp.), cholera (*Vibrio cholerae*), dysentery (*Shigella* sp.), *Yersinia* sp., *Campylobacter* sp. and *E. coli*, which

Table 5
Rate constants of fecal coliforms retained in soil ecosystems irrigated with different low quality water under bioremediation and phytoremediation trails in the green house (CFU g^{-1} day^{-1})

Giza Governorate				
	a	b	R^2	SE
NC	–0.62	4.19	0.99**	0.03
C	–1.88	6.17	0.99**	0.07
T1	–2.53	6.60	0.95**	0.27
T2	–2.55	6.20	0.92**	0.34
T3	–2.58	5.75	0.97**	0.07
Kafr El-Sheikh				
NC	–0.29	3.06	0.92**	0.08
C	–1.69	4.15	0.95**	0.38
T1	–1.70	4.09	0.9288	0.51
T2	–2.11	4.12	0.96**	0.43
T3	–2.26	4.24	0.97**	0.36
Saini Governorate				
NC	–0.51	2.03	0.91**	0.16
C	–0.10	1.71	0.93**	0.04
T1	–0.38	1.97	0.95**	0.33
T2	–0.53	1.84	0.98**	0.21
T3	–0.57	1.96	0.97**	0.10

were verified in their ability to survive in the soil ecosystem for several months (Santamarý´ and Toranzos, 2002; Housing and Building National Research Center, 2004; Saber 2007). Kabary et al., 2021a revealed the existence of different genera of water borne bacterial pathogens *(Salmonella, Shigella, Listeria, Campylobacter,* fecal *Streptococci*, and pathogenic *E. coli* O157:H7) in two agriculture drains Belbeis and El Rahawy in Egypt which were commonly used for irrigation.

Different biotic and abiotic factors are controlling the existence and survival of enteric microbes in soil. For example, survival of *E. coli* and other fecal microbes in the soil ecosystem usually regulated by the existence of other saprophytic soil microflora. The antibiotic producing characteristics of some of PGP (plant growth promoters) including the applied types of PDB by which restricting the survival of plant pathogens may also affect the survival of fecal microbes in same environment. Moreover, since there is always unknown reservoir of natural plants that may have medicinal properties not yet discovered, the antimicrobial potency of various plant root exudates and organic residues may also restrict the growth of enteric pathogens. In study suggested by Gutierrez-Gines et al., 2021, native New Zealand plants were capable of removing 90% *E. coli* existence in 14 days greenhouse experiment. Like our study, control cultivated trials showed considerable removal of fecal coliforms and *Salmonella* sp. in comparison to non-cultivated trials which reached 98% of total fecal coliforms removal in *Plantago major* cultivated Giza soil after 90 days of the trial. Abiotic factors of the surroundings environment also exert another effective parameter that regulate the existence of enteric pathogens in soil ecosystems. Soil colloids content, pH, cations, moisture, temperature, salinity, organic matter, and heavy metal concentration all these factors have indirect multidimensional effect on enteric pathogens survival. As our results detected, the existence of enteric bacteria was significantly limited with high salinity characteristics of Sinai soil followed by Kafr-El-sheikh soil which may be due to the source of irrigation water. Gerba and Bitton, 1984 stated that, colloids might adsorb enteric pathogens and eventually decrease their death rates. Soil pH and cations concentrations affect the adsorption characteristics of the microbial cells which reflected on bacterial cell survival. Also, Soluble organics and temperature have a strong effect on the proliferation of microorganisms in soil (Straub et al., 1992).

It is broadly steady that it is not feasible to detect the sustenance of the whole pathogenic community in low quality water irrigated soils. For this reason, the reference microbe concept was highly conserved for years. Laboratory examination for fecal coliforms bacteria was convinced by technical institutions to designate microbial contamination. Although, fecal coliforms are not considered as pathogens they are more like ensigns that pathogens could be present and may flourish and proliferate in the soil ecosystem. EPA *endorsed E. coli* as a receptive indicator of fecal contamination. Yet, no single reference microorganism detection could foresee the existence of all kinds of enteric pathogens for all soil types and various host-related fecal contamination. (Tyagi et al., 2006).

Usually, it is denied for any fecal coliform pathogens to be detected at any amount. However, WHO (1989) estimated that fecal coliforms viable count <10 CFU (colony forming unite)/g or ml might be considered as a safe usage level. In the current research study, *Salmonella* count was also put into consideration besides total fecal coliform count as indicator for soil contamination with wastewater because many microbes like *Aerobacter*, *Candida* and *Klebsiella* etc., can enumerate on MacConkey medium (Kabary et al., 2021a) producing the same reaction (gas and acid production) at 44°C after 24hrs of incubation. Saber et al. (2011) stated that decontamination of soil pathogens with various chemical and microbial remediated treatments is a common practice. In this study, different remediated amendments were applied including clay minerals, rock phosphate, elemental sulfur, and exogenous zymogenic microorganisms. Enhancing the growth and activity of *Thiobacillus* sp. in the soil ecosystem by sulfur element led to better and faster removal of pollutants that might be ascribed to the oxidation of elemental sulfur to H_2SO_4 by sulfur-oxidizing bacteria then decreasing soil pH (Tabak et al., 2020).

Clays are hydrous aluminum silicates that are a main component of the colloid fraction of soil ecosystems (Zhao et al., 2014). Different studies have reported the rule of Clay minerals as a natural contaminants' scavengers, sequestering anions and cations either by ions exchange and/or adsorption (Bhattacharyya and Gupta 2009; El-Korashy et al. 2016). Kabary et al. (2021b) reported the rule of bentonite amalgamed with mixture of *Thiobacillus* and PDB cultures in removal of fecal coliform from drainage water samples in percentage 61% compared to control samples.

The worldwide prevalence of microbial pathogens contamination is a serious issue and focusing on the understanding of major techniques and best management practices should be applied to minimize the hazards of pathogen and their significant impacts.

5. Conclusions

This study investigated the possibility of removing Enteric Pathogens in contaminated soils varied in their physiochemical characteristics and source of polluted irrigation water either industrial effluents, sewage effluents or mixture of both. The suggested remediation technique based on application of combined integrated methods: biochemical (sulfur and rock phosphate) and biological (phytoremediation) with fortified clay minerals. Results imply the application of soil treated with combined mixture of bentonite, sulfur and phosphate and inoculated with *Thiobacillus* and phosphate dissolving bacteria (PDB) at the rate of 1.25 ton/fed (T_3) with using *Plantago psyllium* as phytoremediation plant was the BMP to be applied for remediation of such polluted ecosystems and decrease the existence of fecal coliforms.

Abbreviations
BMP: Best Management Practices
C: Cultivated Control soil
NC: Non-cultivated Control soil
MPN: Most Probable Number
WHO: World Health Organization

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