OPEN ACCESS

International Journal of Applied Sciences and Biotechnology

ISSN 2091-2609

Indexing and Abstracting

CrossRef, Google Scholar, Global Impact Factor, Genamics, Index Copernicus, Directory of Open Access Journals, WorldCat, Electronic Journals Library (EZB), Universitätsbibliothek Leipzig, Hamburg University, UTS (University of Technology, Sydney): Library, International Society of Universal Research in Sciences (EyeSource), Journal Seeker, WZB, Socolar, BioRes, Indian Science, Jadoun Science, Jour-Informatics, Journal Directory, JournalTOCs, Academic Journals Database, Journal Quality Evaluation Report, PDOAJ, Science Central, Journal Impact Factor, NewJour, Open Science Directory, Directory of Research Journals Indexing, Open Access Library, International Impact Factor Services, SciSeek, Cabell's Directories, Scientific Indexing Services, CiteFactor, UniSA Library, InfoBase Index, Infomine, Getinfo, Open Academic Journals Index, HINARI, etc.

CODEN (Chemical Abstract Services, USA): IJASKD

Vol-3(3) September, 2015

Available online at:

http://www.ijasbt.org

&

http://www.nepjol.info/index.php/IJASBT/index

SEM-Biotech Publishing

Impact factor*: **1.422**
Scientific Journal Impact factor#: **3.419**
Index Copernicus Value: **6.02**
IBI Factor 2015**: **4.19**

*Impact factor is issued by Universal Impact Factor. Kindly note that this is not the IF of Journal Citation Report (JCR).
#Impact factor is issued by SJIF INNO SPACE; **Impact factor is issued by INFOBASE INDEX.

For any type of query and/or feedback don't hesitate to email us at: editor.ijasbt@gmail.com

Yehia A. Osman et al. (2015) Int J Appl Sci Biotechnol, Vol 3(3): 528-536
DOI: 10.3126/ijasbt.v3i3.13332

ISSN (Online): 2091-2609
DOI Prefix: 10.3126/ijasbt

International Journal of Applied Sciences and Biotechnology (IJASBT)

Research Article

EFFICIENCY OF TRADITIONAL WATER TREATMENT PLANT AND COMPACT UNITS IN REMOVING VIRUSES

Yehia A. Osman[1], Waled M. El-Senousy[2], Adel A. El-Morsi[1] and Mohammed K. Rashed[2]

[1] Botany Department, Faculty of Science, Mansoura University, Egypt, 35516
[2]Water Pollution Research Department, Environmental Research Division, National Research Centre (NRC), 33 El-Buhouth st., Dokki, Giza P.O. 12622, Egypt.

Corresponding author's email: kimo_micro_h@yahoo.com

Abstract

The fecal bacteria have been taken as the gold standard for water industry. However, the spread of viral gastroenteritis due to drinking water have given a momentum to a recent push by microbiologists to consider viruses as important pollution indicator as fecal bacteria. Therefore, we designed a study to evaluate the efficiency of two types of water purification systems: the traditional water treatment plant and two types compact units. Both systems produced drinking waters free of bacteria, chemical contaminants and mostly viruses free. However, recent advances in molecular biology techniques, such as RT-PCR have detected Rotaviruses in chlorinated drinking waters resulted from all systems. The frequency of Rotaviruses since October 2010 till September 2012 in Shark El-Mansoura WTP in drinking water samples was 12.5% similar to raw water. While the compact unit at Depo Awam (American design) the frequency of Rotavirus was 16.6% in both raw and drinking water samples. On the other hand the virus frequency in the raw and drinking water sample in El-Danabik unit (Egyptian design) were 12.5% and 4.16% respectively. Signifying failure of the chlorination process in removing viruses completely. However, detection of Rotavirus genome in the drinking water samples does not means the presence of its infectivity. The infectious ability of the rotaviruses was confirmed by CC-RT-PCR in all positive samples, where viral RNA was not detected in the collected drinking water samples. In conclusion RT-PCR and CC-RT-PCR techniques high lightened the need to include viruses as mandatory pollution indicator in water treatment plants.

Keywords: Viral gastroenteritis; Drinking water; Rotaviruses; Pollution; Water purification systems; RT-PCR

Introduction

Many health risks are associated with drinking water include infectious diseases caused by bacteria, protozoa, viruses and intestinal helminthes (Ress *et al*., 2000 and Gibson *et al*., 2011). This is dramatized in the 842,000 deaths and billions of cases of diarrheal disease were reported annually due to the inadequate access to either sufficient and/or safe drinking water (Clasen *et al*., 2014). The dependence of water treatment and manufacturing industry on bacterial indicators such as total coliform, fecal coliform (*Escherichia coli*), Streptococci and *Salmonella*) was not enough (Leclerc *et al*., 2002 and Carducci *et al*., 2013). Since the bacterial indicators do not always reflect the risks associated with other important pathogenic bacteria, protozoan parasites (*Cryptosporidium, Giardia*) and enteric viruses (Griffin *et al*., 2001, Jiang *et al*., 2001 and Noble and Fuhrman, 2001).

Waterborne illness is complicated due to the presence of about 140 different serological types of viruses, found in water *via* sewage contamination. These viruses are capable of causing illnesses to humans such as acute gastroenteritis (AGE) diseases (Taylor *et al*., 2001, Hamza *et al*., 2009, Enserink *et al*., 2015 and Patil *et al*., 2015). These viruses are transmitted from person to person or through contaminated drinking water, food and bathing or recreational water (Rodriguez-Lazaro *et al*., 2012). The poor correlation of bacterial indicators with viruses is of particular concern because it cannot be used as reliable indicators of faecal pollution and viral particles in water (Jurzik *et al*., 2010, Chigor and Okoh, 2012 and Carducci *et al*., 2013). Furthermore, enteric viruses were detected in raw, surface water, ground water and treated drinking water despite meeting quality standards for coliform bacteria (Cho *et al*., 2000 and Pusch *et al*., 2005).

Considering the following virus attributes: low infectious doses, linkage with both acute and chronic disease and frequent implication in swimmer-associated illnesses (Fong and Lipp, 2005). Moreover, Chigor and Okoh (2012) have shown that bacteriological indicators, some human viruses and coliphages may beneficially serve as an index in determining viral contamination and the presence of human fecal waster. Generally, viruses are more resistant to extreme environmental conditions and treatment processes, such as chlorination, UV radiation and filtration compared

to fecal bacterial indicators and other pathogens (Ahmed *et al*., 2010). While, enteric viruses are relatively resistant to heat, disinfectants and pH changes despite the absence of viral envelope (Koopmans *et al*., 2002). Most of the enteric viruses are host specific and thus allow screening of the species which is the source of fecal contamination (Silva *et al*., 2011 and Wu *et al*., 2011). Enteric viruses are shed in extremely high numbers in the feces of infected individuals, 10^5 to 10^{13} virus particles per gram of stool (Hamza *et al*., 2009 and Schultz *et al*., 2011).

Acute gastroenteritis, mainly diarrhea, is one of the most common diseases in human, and remains a leading cause of morbidity and mortality worldwide. It is reported that about 3–5 billion cases of acute gastroenteritis occur each year in children under 5 years, resulting in nearly 2 million deaths (Parashar *et al*., 2003, Mulholland, 2004 and Elliott, 2007). In developing countries, the incidence rate of acute gastroenteritis is 2.1 to 3.8 diarrhea episodes per child between 11 and 48 months of age per year (Kosek, 2003).

This has posed the challenge of finding a suitable indicator of viral contamination of drinking water. In 2008, the World Health Organization has estimated that rotavirus alone caused 453, 000 deaths, accounting for 5% of all deaths in children younger than 5 years old (WHO, 2012). Therefore, we have taken rotaviruses as good indictors production good quality drinking waters. In this study, we detected Rotavirus in raw Nile water and after each step of treatment in Shark El-Mansoura Water Treatment Plant (WTP) and the two compacts units of Depo Awam and El-Danabik villages.

Materials and Methods

Sites of the water treatment plant and compact units

Two types of water treatment plants distributed in three sites were involved in this study. The conventional Shark El-Mansoura water treatment plant (WTP) which supplies residents of Mansoura City, Egypt, with drinking water. While, the two compact units of Depo Awam (American design) and El-Danabik (Egyptian design) supplies drinking water to residents of respective villages. The WTP is supplied by the raw water from El-Mansoria canal and the water treatment in this plant goes through the traditional steps of flocculation, sedimentation, sand filtration and final chlorination. The Depo Awam unit is supplied by fresh waters from Bahr Tnah and El-Danabik unit is supplied by raw water from a small fresh water canal branched from El-Bahr El-Sageer canal.

Water Samples Collection

A total of 192 water samples (20 litters each) were collected from the three sites in the period of October 2010 to September 2012. A total of 96 samples were collected from the different steps of water purification in WTP as follow: 24 samples from raw water (inlet), 24 samples after sedimentation step, 24 samples after sand filtration step and 24 samples from outlet water (drinking). While, 48 water samples were collected from each compact unit: 24 samples from inlet raw water and 24 samples from outlet water. The chlorinated water samples were treated with sodium thiosulfate (0.5% wt/v) to inactivate chlorine followed by 6N aluminum chloride, to increase the stability of the viruses in the concentrated water samples (APHA, 2005).

Physicochemical Analysis of Water Samples

The physiochemical properties of raw water samples: like temperature, pH, turbidity, alkalinity, electrical conductivity, hardness, chloride, dissolved oxygen and consumed oxygen were measured by the standard procedure detailed in the Egyptian standard methods (EMH, 2007) and the American standard methods (APHA, 2005)..

Bacteriological Analysis of Water Samples

Fecal contamination analysis of water samples were performed according to the universally accepted standard methods for bacteriological examination of waters. In these methods we estimated the number of live heterotrophic bacteria on R2A (low nutrient media), total coliform bacteria using membrane filter technique within 24 h at 35°C on an Endo-type medium containing lactose, the fecal coliform using M-FC media at 44.5 ± 0.2°C and detection of fecal *Streptococcus* group on m-enterococci media grown for 48 hr at 35 ± 0.5ºC (APHA, 2005).

Virus Isolation

Primary and secondary water samples concentrations were performed by adsorption/elution technique (APHA, 2005). Primarily, the water samples were acidified (pH 3.5) before filtration, to enhance the adsorption of virus particles to the negatively charged nitrocellulose membrane filters. Viruses were eluted from the nitrocellulose membranes by a 70 ml of 0.05 M glycine buffer containing 3% beef extract, pH ~9.5 (Smith and Gerba, 1982 and Rose *et al*., 1984). In the secondary concentration**,** the eluate from primary concentrate is again acidified (pH 3.5) to help viruses to be trapped in the flocks of proteins and organic components before being harvested by centrifugation at 3,000 rpm. Each of the harvested pellets was dissolved in 1 ml Na_2HPO_4 (0.14N, pH 9) and kept at -70ºC until used for detection of viruses (Katzenelson *et al.*, 1976).

Extraction of Total RNA from Rotaviruses

The Rotaviruses RNA was extracted by the TRIzol method (BIOZOL Total RNA Extraction reagent, BioFlux, Japan) according to the manufacturer's instructions as detailed by Steyer *et al* (2008). The RNA pellet was dissolved in 50-100μl of RNase-free water and stored at -70 °C for further use.

Reverse Transcriptase-PCR (RT-PCR)

Rotavirus viral protein 6 (VP6), is the gold standard for detection and diagnosis of all Retroviruses. Two pairs of oligonucleotide primers were used to amplify a 379-b region of the VP6 gene: VP6-F: 5'-

GACGGNGCNACTACATGGT-3' and VP6-R: 5'-GTCCAA TTCATNCCTGGTGG-3'. A second pair of primers, VP6-NF: 5'-GCTAGAA ATTTTGATACA-3' and VP6-NR: 5'- TCTGCAGTTTGTGAATC- 3', were used to amplify a 155 b fragments. Extracted water samples (5 μl) were heated to 99°C for 5 min and immediately placed on ice. Salts, nucleotides, primers and 100 U of reverse transcriptase (Fermentas-EU) were added in 10 μl final volume to give a working concentration of 50 mM Tris-HCl, pH 8.3, 40 mM KCl, 5 mM $MgCl_2$, 5 mM DTT, 0.5 mM Tween 20, 0.2 mM of each dNTP·s (Fermentas-EU) and 1 μM of both VP6-F and VP6-R primers. The samples were incubated for 60 min. at 50°C for the RT reaction. Five μl of the RT product were added to a final volume of 50 μl of the PCR reaction mix containing 5 μl of the PCR buffer (Fermentas-EU), 2 mM $MgCl_2$, 0.2 mM of each dNTP·s, 1 μm of each primer and 2.5 U of the *Taq* DNA polymerase enzyme (Fermentas-EU). After a denaturation step of 95°C for 3 min, 40 cycles of amplification at 94°C for 1 min, 50°C for 1 min, and 72°C for 1 min. were performed with a final extension of 72°C for 10 min. The nested PCR involved adding 2 μl of first-round PCR product to a 48μl PCR mix containing 10 mM Tris (pH 8.0), 50 mM HCl, 2.5 mM $MgCl_2$, 0.2 mM of each dNTPs (Fermentas-EU), 1 μM of VP6NF and VP6NR primers, and 2.5 U of *Taq* DNA polymerase (Fermentas-EU). Cycling conditions for VP6NF/VP6NR were 35 cycles of 94°C for 1 min, 42°C for 1 min, and 72°C for 1 min. PCR products (10 μl) were analyzed by electrophoresis on 3% agarose gels (Iturriza-Gömara *et al.*, 2002 and Gallimore *et al.*, 2006).

Results and Discussion

International and local standards were set and established worldwide for drinking water purifications processes which included minimum and maximum limits for contaminants. Water purification processes are intended to remove all sorts of contaminants from natural waters (rivers, canals and/or reservoirs) which are loaded with undesirable chemicals and biological contaminants, that can cause human illness such as bacteria, viruses, fungi, protozoa and some algae.

Two types of water purifications systems do exist in Egypt and worldwide. The traditional methods of water purification (includes physical processes such as flocculation, sedimentation, sand filtration and chlorination) and the pre-packed compact smaller units used for quick water purification mainly in rural areas. The two systems examined in Mansoura and its surroundings proved to be highly efficient in removing all sorts of chemicals and microbial contaminants from raw waters and produced drinking waters meeting all national and international standards. However, their abilities to remove the viral causative agents of human illnesses, such as Rotaviruses. Since Rotaviruses was detected in some drinking water samples from both systems. While seasonal detection of Rotaviruses in winter and autumn was observed.

Water quality through the presence of pathogenic enteric microorganisms may negatively affect human health. Where, coliform bacteria, such as *Escherichia coli*, and coliphages are normally used as indicators of water quality. However, the presence of above-mentioned indicators do not always suggest the presence of human enteric viruses which may be more resistance than the bacterial indicators. Therefore, Lin and Ganesh, (2013) concluded that it is highly important to study human enteric viruses in water to avoid their pathogenic action on children and immune-compromised people. While the current study demonstrated the efficiency and efficacy of the two water treatment systems in removing all microbial and undesired chemical contaminants. It failed to completely remove causative agents of gastroenteritis viruses such as Rotaviruses (Tables 1, 2 and Fig 1).

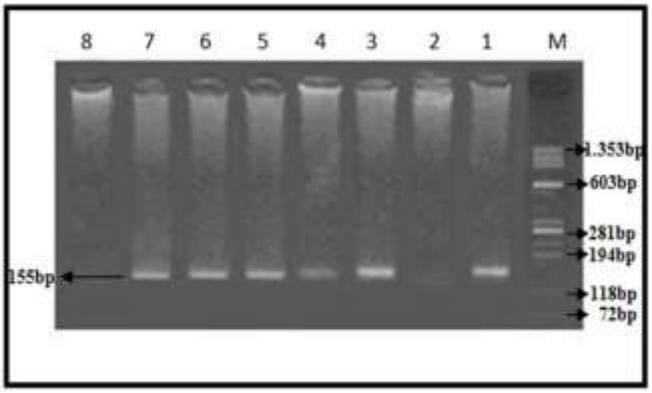

Fig. 1: Agarose gel electrophoresis (3%) in TBE buffer stained with ethidium bromide showing RT-PCR product profile of VP6 gene characteristic of Rotaviruses in examined water samples, lane 1: raw water of Shark El Mansoura (Jan.2011), lane 2: after sedimentation water of Shark ElMansoura (Jan.2011), lane 3: after sand filtration of Shark El Mansoura (Jan.2011), lane 4: chlorinated effluents of Shark El Mansoura (Jan.2011), lane 5: raw water of DepoAwam CU (Nov.2011), lane 6: chlorinated effluents of Depo Awam CU (Nov.2011), lane 7: raw water of El-Dnabik CU (Nov.2011), 8: chlorinated effluents of El-Dnabik CU (Nov.2011), all bands of positive samples were appeared with a size about155b. Marker: ØX 174 / HaeIII (Bio labs).

RT-PCR analysis (using the highly conserved the sixth viral structural proteins, VP6) of water samples collected from Shark El-Mansoura traditional water treatment plant showed a frequency of Rotaviruses in the monthly collected water samples (October 2010 – September 2012) of 3/24 (12.5%), 3/24 (12.5%), 4/24 (16.6%) and 3/24 (12.5%) in raw water, after sedimentation, after sand filtration and in chlorinated effluents (drinking water samples), respectively (Table 1). During the same period, the frequency of Rotaviruses in monthly collected water samples of Depo Awam was 16.60% (4/24) and 16.6% (4/24) in raw water and chlorinated effluents, respectively. While, the frequency of Rotaviruses in El-Dnabik station was 12.50%

(3/24), and 4.16% (1/24) in raw water and Chlorinated effluents, respectively. Only water samples collected in the months of January 2011 and September 2011 showed positive results for the existence of Rotaviruses. While, RT-PCR positive Rotavirus were detected in raw and drinking water samples collected from the compact units of Depo Awam and Danabik in the months of November 2010 and Septemebr 2011, suggesting a failure in the systems in the mentioned months only.

Table 1: Rotavirus in water samples collected from Water treatment plants.

Year	WTPs	Shark El Mansoura[1]				Depo Awam[2]		El Dnabik[3]	
	Months	Raw water	After Sedimentation	After Sand Filtration	Tap water	Raw water	Tap water	Raw water	Tap water
2010	October	-	-	-	-	-	-	-	-
	November	-	-	-	-	+	+	-	-
	December	-	-	-	-	-	-	-	-
2011	January	+	-	+	+	-	-	-	-
	February	-	-	+	-	-	-	+	-
	March	-	-	-	-	-	-	-	-
	April	-	-	-	-	-	-	-	-
	May	-	-	-	-	-	-	-	-
	June	-	-	-	-	-	-	-	-
	July	-	-	-	-	-	-	-	-
	August	-	-	-	-	-	-	-	-
	September	+	-	-	+	-	+	+	+
	October	-	+	-	+	-	-	-	-
	November	-	+	-	-	+	+	+	-
	December	+	-	+	-	+	-	-	-
2012	January	-	-	-	-	+	+	-	-
	February	-	+	+	-	-	-	-	-
	March	-	-	-	-	-	-	-	-
	April	-	-	-	-	-	-	-	-
	May	-	-	-	-	-	-	-	-
	June	-	-	-	-	-	-	-	-
	July	-	-	-	-	-	-	-	-
	August	-	-	-	-	-	-	-	-
	September	-	-	-	-	-	-	-	-

1- Traditional water treatment plant
2- American design compact unit
3- Egyptian design compact unit
+ : Presence
- : Absence

Table 2: Total coliforms, fecal coliforms and fecal streptococci of selected raw water and drinking samples

Water Treatment System	Type of Water	Bacterial Colony Forming Unit (CFU) / 100 ml														
		Total coliforms					Fecal coliforms					Fecal streptococci				
	Month/year	Oct 2010	Jan 2011	Apr 2011	Jul 2011	Sep 2011	Oct 2010	Jan 2011	Apr 2011	Jul 2011	Sep 2011	Oct 2010	Jan 2011	Apr 2011	Jul 2011	Sep 2011
Shark El-Mansoura[1]	Raw water	$59x10^3$	$30x10^3$	$53x10^3$	$73x10^3$	$56x10^3$	$69x10^2$	$55x10^2$	$12x10^2$	$25x10^2$	$45x10^2$	44x10	35x10	22x10	35x10	19x10
	Drinking water	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill
Depo Awam[2]	Raw water	$57x10^3$	$29x10^3$	$41x10^3$	$70x10^3$	$45x10^3$	$77x10^2$	$34x10^2$	$10x10^2$	$19x10^2$	$26x10^2$	57x10	39x10	37x10	37x10	29x10
	Drinking water	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill
Danabik[3]	Raw water	$56x10^3$	$26x10^3$	$43x10^3$	$68x10^3$	$47x10^3$	$76x10^2$	$30x10^2$	$9x10^2$	$17x10^2$	$29x10^2$	58x10	37x10	33x10	39x10	21x10
	Drinking water	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill	nill

1- Traditional water treatment plant
2- American design compact unit
3- Egyptian design compact unit

Table 3: The physico-chemical parameters of selected water samples in different seasons

Seasons	Autumn 2010						Winter 2011						Spring 2011						Summer 2011					
WTPs	Shark[1]		Depo[2]		ElDnabik[3]		Shark[1]		Depo[2]		ElDnabik[3]		Shark[1]		Depo[2]		ElDnabik[3]		Shark[1]		Depo[2]		ElDnabik[3]	
Water types	Raw	Tap	Raw	Tap	Raw	Tap	Raw	Tap	Raw	Tap	Raw	Tap	Raw	Tap	Raw	Tap	Raw	Tap	Raw	Tap	Raw	Tap	Raw	Tap
Temp	23.2	24.7	23	23.5	23	23.4	18	18	18.6	18.4	18	18.7	24	24.5	24.5	24.4	24.4	24	31.7	31.5	30.3	30	30	30.4
R.CL	0	2	0	2	0	1.8	0	1.8	0	1.7	0	1.8	0	1.8	0	1.5	0	1.7	0	2	0	1.8	0	1.7
Turbidity	8.2	0.17	14	0.4	12	0.4	8.5	0.15	6.8	0.96	10	0.25	9.4	0.14	15.4	0.95	11.1	0.34	8.4	0.15	11	0.82	12	0.8
PH	7.68	7.32	7.63	7.34	7.7	7.37	7.74	7.25	7.78	7.32	7.75	7.38	7.7	7.3	7.75	7.45	7.78	7.25	7.75	7.3	7.72	7.38	8	7.4
T.D.S	267	277	265	273	265	287	295	316	276	289	373	319	237	240	231	236	235	251	207	211	209	212	195	200
Alkanility	146	132	144	138	148	132	148	132	142	136	148	132	126	118	128	118	128	124	134	114	138	128	122	116
TotalHardnes	142	140	138	138	140	142	144	138	146	140	138	148	118	114	114	118	112	130	120	114	124	124	120	120
Ca Hardnes	88	88	88	88	86	82	86	82	88	80	82	88	82	80	78	84	78	82	64	66	82	82	80	76
Mg Hardnes	54	52	50	50	54	60	58	56	58	60	56	60	36	34	36	34	34	48	46	48	42	42	40	44
Sulphate	20	25	20	25	20	24	38	42	28	34	37	34	27	36	23	32	28	31	24	30	30	33	28	34
Chlorides	30	38	28	38	26	32	38	46	30	38	36	48	26	30	22	30	24	30	18	26	20	30	18	24
Amonia	0.13	0	0.13	0	0.2	0	0.17	0	0.15	0	0.15	0	0.12	0	0.15	0	0.16	0	0.16	0	0.19	0	0.18	0
Nitrite	1.7	2.7	2.1	3.8	2	3.8	3.7	5	2.8	3.9	2.4	3.2	3.81	4.4	2.5	5.3	3.4	5.5	3.9	5.7	2.8	5.3	3.1	5.5
Nitrate	0.017	0	0.006	0	0.008	0	0.031	0	0.08	0	0.015	0	0.021	0	0.021	0	0.022	0	0.005	0	0.023	0	0.021	0
Iron	0.05	0.02	0.02	0.01	0.01	0.01	0.03	0.01	0.01	0	0.002	0.01	0.04	0.01	0.01	0	0.02	0	0.02	0.01	0.01	0	0	0
Mnganes	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Almonium	0.038	0.065	0.038	0.078	0.044	0.069	0.014	0.021	0.01	0.08	0.017	0.061	0.008	0.014	0.02	0.034	0.006	0.019	0.009	0.022	0.021	0.51	0.02	0.04
(DO)	6.4	7.9	6.2	8.2	5.6	7.9	5.7	8	6.4	7.8	5.6	5.3	6	8	6	7.4	6	7.4	5.2	6.5	5.6	7.2	6.2	7.5
(COD)	3.9	0	4	0	3.9	0	4	0	3.9	0	4	0	3.9	0	3.5	0	3.9	0	4.6	0	3.2	0	4	0

1- Traditional water treatment plant (Shark El Mansoura)
2- American design compact unit (Depo Awam)
3- Egyptian design compact unit (El Dnabik)

These results were similar to the previously published reports on Rotaviruses in raw Nile water, treated drinking water and ground waters in Egypt (El-Senousy *et al*., 2004, El-Senousy and El-Mahdy, 2009, El-Senousy *et al*., 2013a&b and El-Senousy *et al*., 2014), Tunisia (Sdiri-Loulizi *et al*., 2008) and France (Gratacap-Cavallier *et al*., 2000**).** The French report emphasized that the winter epidemics of Rotavirus infections was associated with a high level of interhuman transmission, after they have analyzed drinking waters in homes of children suffering from Rotaviral gastroenteritis by RT-PCR. Moreover, they have detected in the children's feces rotavirus genome different from human Rotaviruses, three of them were of animal origin (porcine or bovine). On the other hand, Grassi, *et al* (2010) showed the widespread viral contamination in different water samples collected from Italy and Rotaviruses peaked in spring. On the contrary, Verheyen, *et al* (2009) have concluded that no seasonal pattern for viral contaminations was found after comparisons of water samples obtained during the dry and wet seasons from Benin, West Africa. The detection of genome in the drinking water samples did not mean the capability of the virus to cause diseases. It does not confirm the infectivity of the virus (He *et al*., 2009). Liu *et al*., (2006) attributed the higher frequency of detection of rotaviruses (for example) was due to an outbreak of diarrheal in Beijing 2006 where Rotavirus was detected in 60% of all diarrheal patients. The traditional water treatment is extensive process and involves several steps subjective to humans interferences, compact units do everything inside without such interference. Although the disinfecting power of chlorine is well documented in the literature against all types of microbes including viruses, the detection of Rotaviruses in the drinking water sample in Shark El-Mansoura WTP suggested a failure in the chlorination process which needs attention. Chlorination was reported denature the proteins and causes breakage in the nucleic acid molecules (Ogata, 2007). These should be sufficient to remove all forms of microbes and viruses. Moreover, the detection of viruses in drinking waters produced by the two compact units indicate an inherited problems with these units which requires more and thorough investigation.

References

Ahmed W, Goonetilleke A and Gardner T (2010) Human and bovine adenoviruses for the detection of source specific fecal pollution in coastal water in Australia. *Water Research* **44**(16): 4662 -4673. DOI: 10.1016/j.watres.2010.05.017

American Public Health Association. (APHA) (2005) Standard methods. 24th Edition. American Public Health Association, Washington, DC,

Carducci A, Federigi I and Verani M (2013). Virus occupational exposure in solid waste processing facilities. *Annals of occupational hygiene* met **57**(9): 1115-1127. DOI: 10.1093/annhyg/met043

Chigor VN and Okoh AI (2012) Quantitative RT-PCR detection of hepatitis A virus, rotaviruses and enteroviruses in the Buffalo River and source water dams in the Eastern Cape Province of South Africa. *International Journal of Environmental Research and Public Health* **9**: 4017-4032. DOI: 10.3390/ijerph9114017

Cho H, Lee S, Cho J and Kim S (2000) Detection of adenoviruses and enteroviruses in tap water and river water by reverse transcription multiplex PCR. *Canadian journal of microbiology* **46**(5): 417-424. DOI: 10.1139/w00-014

Clasen T, Pruss-Ustun A, Mathers C, Cumming O, Cairncross S and Colford J (2014) Estimating the impact of unsafe water, sanitation and hygiene on the global burden of disease, evolving and alternative methods. *Tropical Medicine and International Health* **19**(8): 884-893. DOI: 10.1111/tmi.12330

Egyptian Ministry of Health (EMH) (2007) Standards and specifications of water quality for drinking and domestic uses. Internal Report. **8**.

Elliott EJ (2007) Acute gastroenteritis in children. *British Medical Journal* **334**(7583): 35. DOI: 10.1136/bmj.39036.406169.80

El-Senousy W, Pintó R and Bosch A (2004) Epidemiology of human enteric viruses in the Cairo water environment. The 1st International Conference of Environmental Research Division on Sustainable Development Environmental Challenges Facing Egypt. National Research Centre, Cairo, Egypt.

El-Senousy WM and El-Mahdy EM (2009) Detection and genotyping of rotaviruses in water treatment plants of El-Dakahlia Governorate. *Egyptian Journal of Biotechnology* **31**: 25-34.

El-Senousy WM, Barakat AB, Ghanem HE and Kamel MA (2013a) Molecular epidemiology of human adenoviruses and rotaviruses as candidate viral indicators in the Egyptian sewage and water samples. *World Applied Sciences Journal* **27**: 1235-1247. DOI: 10.5829/idosi.wasj.2013.27.10.81200

El-Senousy WM, El-Gamal MS, Mousa AA, El-Hawary SE and Fathi MN (2014) Prevalence of Noroviruses among Detected Enteric Viruses in Egyptian Aquatic Environment. *World Applied Sciences Journal* **32** (11): 2186-2205. DOI: 10.5829/idosi.wasj.2014.32.11.91108

El-Senousy WM, Sidkey NM, Abu Senna AS, Abed NN and Hasan SF (2013b) Prevalence of rotaviruses and noroviruses in ground water of some rural areas in El-Giza Governorate, Egypt. *The new Egyptian Journal of Medicine* **48**(3): 18-25.

Enserink R, van denWijngaard C, Bruijning-Verhagen P, van Asten L, Mughini-Gras L, Duizer E, Kortbeek T, Scholts R, Nagelkerke N, Smit H, Kooistra-Smid M and van Pelt W (2015) Gastroenteritis attributable to 16 Enteropathogens in children attending day care. Significant effects of Rotavirus, Norovirus, Astrovirus,

Cryptosporidium and Giardia *The Pediatric infectious disease journal* **34**(1): 5-10. **DOI:** 10.1097/inf.0000000000000472

Fong T and Lipp E (2005) Enteric Viruses of Humans and Animals in Aquatic Environments: Health Risks, Detection, and Potential Water Quality Assessment Tools. *Microbiology and Molecular Biology Reviews* **69**(2): 357–371. DOI: 10.1128/MMBR.69.2.357-371.2005

Gallimore C, Taylor C, Gennery A, Cant A, Galloway A, Iturriza-Gomara M and Gray J (2006) Environmental Monitoring for Gastroenteric Viruses in a Pediatric Primary Immunodeficiency Unit. *Journal of clinical microbiology* **44**(2): 395–399**. DOI:** 10.1128/JCM.44.2.395-399.2006

Gibson K, Opryszko M, Schissler J, Guo Y and Schwab K (2011) Evaluation of human enteric viruses in surface water and drinking water resources in Southern Ghana. *The American Journal of Tropical Medicine and Hygiene* **84**(1): 20-29. DOI: 10.4269/ajtmh.2011.10-0389

Grassi T, Bagordo F, Idolo A, Lugoli F, Gabutti G and De Donno A (2010) Rotavirus detection in environmental water samples by tangential flow ultrafiltration and RT-nested PCR. *Environmental monitoring and assessment* **164**(1-4): 199-205. DOI: 10.1007/s10661-009-0885-x

Gratacap-Cavallier B, Genoulaz O, Brengel-Pesce K, Soule H, Innocenti-Francillard P, Bost M, Gofti L, Zmirou D and Seigneurin J (2000) Detection of human and animal rotavirus sequences in drinking water. *Applied and Environmental Microbiology* **66**(6): 2690-2692. DOI: 10.1128/AEM.66.6.2690-2692.2000

Griffin DW, Lipp EK, McLAUGHLIN MR and Rose JB (2001) Marine Recreation and Public Health Microbiology: Quest for the Ideal Indicator This article addresses the historic, recent, and future directions in microbiological water quality indicator research. *Bioscience* **51**(10): 817-825. DOI: 10.1641/0006-3568(2001)051[0817:MRAPHM]2.0.CO;2

Hamza I, Jurzik L, Stang A, Sure K, Überla K and Wilhelm M (2009) Detection of human viruses in rivers of a densly populated area in Germany using a virus adsorption elution method optimized for PCR analyses. *Water Research* **43**(10): 2657-2668. DOI: 10.1016/j.watres.2009.03.020

He X, Cheng L, Zhang D, Li W, Xie X, Ma M and Wang Z (2009) First molecular detection of group A rotaviruses in drinking water sources in Beijing, China. The Bulletin of Environmental Contamination and Toxicology 83: 120-124. DOI: 10.1007/s00128-009-9708-6

Iturriza-Gömara M, Wong C, Blome S, Desselberger U and Gray J (2002) Molecular characterization of VP6 genes of human rotavirus isolates: correlation of genogroups with subgroups and evidence of independent segregation. Journal of virology 76(13): 6596–6601. DOI: 10.1128/JVI.76.13.6596-6601.2002

Jiang S, Noble R and Chu W (2001) Human adenoviruses and coliphages in urban runoff-impacted coastal waters of southern California. Applied and Environmental Microbiology 67(1): 179–184. DOI: 10.1128/AEM.67.1.179-184.2001

Jurzik L, Hamza I, Puchert W, Überla K and Wilhelm M (2010) Chemical and microbiological parameters as possible indicators for human enteric viruses in surface water. International journal of hygiene and environmental health 213(3): 210-216. DOI: 10.1016/j.ijheh.2010.05.005

Katzenelson E, Fattal B and Hostovesky T (1976) Organic flocculation: an efficient second-step concentration method for the detection of viruses in tap water. Applied and Environmental Microbiology 32: 838-839.

Koopmans M, von Bonsdorff K, Vinje J, de Medici D and Monroe S (2002) Food borne viruses. Federation of European Microbiological Societies Microbiology 26(2): 187–205.

Kosek M, Bern C and Guerrant RL (2003) The global burden of diarrhoeal disease, as estimated from studies published between 1992 and 2000. Bulletin World Health Organization 81(3): 197-04.

Leclerc H, Schwartzbrod L and DeiCas E (2002) Microbial agents associated with water borne disease. Critical Reviews in Microbiology 28(4): 371–409. DOI: 10.1080/1040-840291046768

Lin J and Ganesh A (2013) Water quality indicators, bacteria, coliphage, enteric viruses. International Journal of Environmental Health Research 3: 1-23. DOI: 10.1080/09603123.2013.769201

Liu C, Grillner L, Jonsson K, Linde A, Shen K, Lindell A, Wirgart B and Johansen K (2006) Identification of viral agents associated with diarrhea in young children during a winter season in Beijing, China. Journal of Clinical Virology 35: 69-72. DOI: 10.1016/j.jcv.2005.04.007

Mulholland EK (2004) Global control of rotavirus disease. Advances in Experimental Medicine and Biology 549: 161–168. DOI: 10.1007/978-1-4419-8993-2_22

Noble RT and Fuhrman JA (2001) Enteroviruses detected by reverse transcriptase polymerase chain reaction from the coastal waters of Santa Monica Bay, California: low correlation to bacterial indicator levels. Hydrobiologia 460(1-3): 175-184. DOI: 10.1023/A:1013121416891

Ogata N (2007) Denaturation of protein by chlorine dioxide: oxidative modification of tryptophan and tyrosine residues. Biochemistry 46(16): 4898-4911. DOI: 10.1021/bi061827u

Parashar UD, Hummelman EG, Bresee JS, Miller MA and Glass RI (2003) Global illness and deaths caused by rotavirus disease in children. Emerging Infectious Diseases 9(5): 565–572. DOI: 10.3201/eid0905.020562

Patil P, Chitambar S and Gopalkrishna V (2015) Molecular surveillance of non-polio enterovirus infections in patients with acute gastroenteritis in Western India: 2004– 2009. Journal of medical virology 87(1): 154-161. DOI: 10.1002/jmv.23992

Pusch D, Oh D, Wolf S, Dumke R, Schröter-Bobsin U, Höhne M, Röske I and Schreier E (2005) Detection of enteric viruses and bacterial indicators in German environmental waters.

Archives of virology 150(5): 929-947. DOI: 10.1007/s00705-004-0467-8

Rees G, Bartram J, Pond K and Goyet S (2000) Introduction. Monitoring bathing waters - A practical guide to the design and implementation of assessments and monitoring programmes. In: Bar¬tram J and Rees G (Eds) 1st ed., E and FN Spon, London, 337. DOI: 10.4324/9780203462171.pt1

Rodriguez-Lazaro D, Cook N, Ruggeri F, Sellwood J, Nasser A, Nascimento M, D'Agostino M, Santos R, Saiz J, Rzetutka A, Bosch A, Girones R, Carducci A, Muscillo M, Kovac K, Diez-Valcarce M, Vantarakis A, von Bons-dorff C, de Roda Husman A, Hernandez M and van der Poel W (2012)Virus hazards from food, water and other contaminated environments. Federation of European Microbiological Societies Reviews 36(4): 786–814. DOI: 10.1111/j.1574-6976.2011.00306.x

Rose JB, Singh SN, Gerba CP and Kelley LM (1984) Comparison of microporous filters for concentration of viruses from wastewater. Applied and Environmental Microbiology 47(2): 989-992.

Schultz A, Perelle S, Di Pasquale S, Kovac K, De Medici D, Fach P, Sommer H and Hoorfar J (2011) Collaborative validation of a rapid method for efficient virus concentration in bottled water. International Journal of Food Microbiology 145(1): 158–166. DOI: 10.1016/j.ijfoodmicro.2010.07.030

Sdiri-Loulizi K, Gharbi-Khélifi H, de Rougemont A, Chouchane S, Sakly N, Ambert-Balay K, Hassine M, Guédiche M, Aouni M and Pothier P (2008) Acute infantile gastroenteritis associated with human enteric viruses in Tunisia. Clinical Microbiology Reviews 46:1349-1355. DOI: 10.1128/JCM.02438-07

Silva H, García-Zapata M and Anunciação C (2011) Why the use of adenoviruses as water quality virologic marker? Food and Environmental Virology 3(3-4): 138–140. DOI: 10.1007/s12560-011-9069-2

Smith EM and Gerba CP (1982) Development of a method for detection of human rotavirus in water and sewage. Journal of Applied and Environmental Microbiology 43(6): 1440–1450.

Steyer A, Poljšak-Prijatelj M, Barli Maganja D and Marin J (2008) Human, porcine, and bovine rotavirus in Slovenia: Evidence of interspecies transmission and gene reassortment. Journal of General Virology 89(7): 1690-1698. DOI: 10.1099/vir.0.2008/001206-0

Taylor L, Latham S and Woolhouse M (2001) Risk factors for human disease emergence. Philosophical Transactions of the Royal Society of London B: Biological Sciences 356(1411): 983-989. DOI: 10.1098/rstb.2001.0888

Verheyen J, Timmen-Wego M, Laudien R, Boussaad I, Sen S, Koc A, Uesbeck A, Mazou F and Pfister H (2009) Detection of adenoviruses and rotaviruses in drinking water sources used in rural areas of Benin, West Africa. Applied and Environmental Microbiology 75(9): 2798-2801. DOI: 10.1128/AEM.01807-08

World Health Organization (WHO) (2012) Immunization surveillance, assessment and monitoring. Online. http://www.who.int/immunization_monitoring/en/ (accessed 10.01.2013).

Wu J, Long S, Das D and Dorner S (2011) Are microbial indicators and pathogens correlated? A statistical analysis of 40 years of research. Journal of Water and Health 9(2): 265–278. DOI: 10.2166/wh.2011.117