EFFICACY OF PATHOGEN REMOVAL DURING FULL-SCALE OPERATION OF WATER REUSE FACILITIES IN MONTEREY, CALIFORNIA

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ABSTRACT

The full-scale operation of water reuse facilities—and delivery of recycled water to farms—by the Monterey Regional Water Pollution Control Agency began in April 1998. Depending on demand for the reclaimed municipal wastewater, up to 90,000 m^3/day (24 million gal/d) can be treated and distributed to farmers to irrigate 5,000 hectares (12,000 acres) of food crops, including artichokes, lettuce, broccoli, and strawberries. The treatment train consists of primary sedimentation, biological secondary treatment (trickling filter/solids contact), coagulation, flocculation, dual-media filtration, and chlorine disinfection. Since 1988, the tertiary effluent has been monitored daily for total and fecal coliform bacteria and approximately three times per year for pathogenic organisms of concern, including total culturable virus, *E. Coli* 0157:H7, *Legionella*, *Salmonella*, *Shigella*, *Cyclospora* oocysts, *Giardia* cysts, *Cryptosporidium* oocysts, and helminth eggs. During five years of monitoring, the California Title 22 standard on total coliform has been exceeded only five times. In terms of pathogens, only *Cryptosporidium* oocysts, *Giardia* cysts, and *Cyclospora* oocysts have been found in the final effluent. *Cryptosporidium* oocysts were detected in 39% and *Giardia* cysts and Cyclospora oocysts in 6% of the effluent samples, with maximum concentrations of 2.3, 0.3, and 0.034 cysts/L, respectively. These low concentrations are not believed to represent a health risk.

Keywords: pathogens, reuse, agricultural irrigation

INTRODUCTION

The Monterey Regional Water Pollution Control Agency (MRWPCA) has provided centralized collection and treatment of the wastewater for Monterey County, California, including the communities of Monterey, Castroville and Salinas, since 1989. In 2002, an average flow of 82,000 m^3/d (21.6 million gal/d) was treated by primary sedimentation and biological secondary treatment (trickling filter/solids contact). Full-scale tertiary treatment facilities, consisting of coagulation, flocculation, dual-media filtration, and chlorine disinfection, began operating in the Spring of 1998. In 2002, the tertiary plant supplied an average flow of 62,000 m^3/day (16.3 million gal/d) over a 9-month period to irrigate approximately 5,000 ha (12,000 acres) of vegetable crops, including artichokes, lettuce, broccoli, and strawberries. Twenty years of planning, field research, pilot demonstration, design, and construction of the treatment and distribution facilities preceded the delivery of recycled water.

During the Monterey Wastewater Reclamation Study for Agriculture (MWRSA) in the early 1980s, pilot-scale tertiary treatment facilities were used to produce reclaimed wastewater to irrigate field trial plots. The results of this study provided convincing evidence that tertiary treated reclaimed water was at least as safe as using the well water commonly used for

irrigation of such crops (Sheikh et al., 1990). Nonetheless, as the time drew near to begin full-scale irrigation using reclaimed water, local farmers and shippers of produce became increasingly concerned about the news coverage of a number of food scares caused by contaminated imported vegetables and poorly handled meat products. These news events caused adverse publicity for the food sectors involved, resulting in economic losses. In partnership with the local water agency, MRWPCA collaborated with the local growers and the Monterey County Environmental Health Department to respond to these concerns. To ensure that every precaution was taken before beginning reclaimed water service to the farming community, MRWPCA undertook a three-month Food Safety Study to determine the potential for existence and survival of pathogens of concern in the product water at the full-scale plant (Sheikh *et al*., 1999). During this study, the tertiary effluent was discharged to the outfall (no irrigation took place).

Even though the MWRSA and Food Safety Studies demonstrated the ability of the tertiary treatment plant to produce—consistently—a virtually pathogen-free recycled water, MRWPCA has continued periodic pathogen monitoring during its regular operational mode since 1998. The purpose of the monitoring program is to confirm the results of the previous studies and to provide a high level of confidence in the safety of the recycled water produced at the plant. Further, it was reasoned that if the results ever demonstrate the discharge of viable pathogenic organisms, corrective action can be taken. The pathogen monitoring program is in addition to the daily monitoring of total (required by the California Title 22 regulations) and fecal coliform bacteria. To provide an additional margin of safety, MRWPCA also monitors the concentrations of total and fecal coliform in the effluent of the storage pond. In addition to the monitoring conducted by MRWPCA, the Monterey County Environmental Health Department conducts independent monitoring of total and fecal coliform bacteria and *Clostridium perfringens* at the storage pond outlet. All of the results from the monitoring programs are displayed, as soon as they became available, on the website of the MRWPCA (www.mrwpca.org). The goal of this paper is to summarize and discuss the results of the indicator organism and pathogen monitoring of the tertiary and pond effluent at the full-scale plant from 1998 to early 2003.

MATERIALS AND METHODS

Starting in 1998, as required by the California Water Recycling Criteria contained in Title 22 of the state code of regulations (DHS, 2001), daily monitoring of total coliform bacteria was performed by collecting grab samples from the east and west chlorine contact basins. The same samples were also analyzed for fecal coliform bacteria. Daily grab samples near the outlet of the storage pond, an open reservoir where the effluent from both the east and west chlorine contact basins awaits distribution, were also analyzed for total and fecal coliform bacteria. All samples were analyzed on-site in the laboratory by the multiple tube fermentation method (9221B for total coliform and 9221C for fecal coliform; Greenberg et al., 1992).

To monitor for pathogens, samples were collected at least three times a year from one of the chlorine contact basins (alternate each sampling event) and shipped on ice to BioVir Laboratories (Benicia, CA) for analysis. For total culturable virus and *Cyclospora*, up to 1,200 and 100 L, respectively, are filtered after dechlorination with sodium thiosulfate. The rest of the organisms are analyzed in grab samples. The samples were analyzed for the presence of total culturable virus (EPA 600/R-95/178), fecal coliform bacteria (Method 9221C, Greenberg et al., 1992), *E. coli* 0157:H7 (Method 997.11, AOAC, 1995), *Legionella* (Method 9260J, Greenberg et al., 1992), *Salmonella* (Method 9260B, Greenberg et al., 1992),

Shigella (Method 9260E, Greenberg et al., 1992), *Cyclospora* (EPA 600/R 178 modified), *Cryptosporidium* and *Giardia* (Method 1623, EPA821-R-97-023), and helminth ova (EPA/625R-92/013).

RESULTS

The results from five years of daily monitoring of total and fecal coliform bacteria in the effluent of the west and east chlorine contact basins are summarized in Tables 1 and 2. The total number of samples analyzed each year varied depending on the number of days that the tertiary plant was in operation, which was determined by the irrigation demand from the farmers. For both tests, the detection limit was 2 MPN/100 mL. Over the 5-year period, more than 2000 samples from the west and east basins were analyzed for total and fecal coliform bacteria. A total of 173 samples (106 in the west basin and 67 in the east basin) contained detectable total coliform and a total of 16 samples (12 in the west basin and 4 in the east basin) contained detectable fecal coliform bacteria. There were only four occasions on which the 7-d median concentration of total coliform bacteria was ≥ 2 MPN/100 mL (three in west basin and one in east basin); the 7-d median concentration of fecal coliform bacteria was never ≥ 2 MPN/100 mL. The highest concentration of total coliform bacteria was 900 MPN/100 mL, which was measured in only one sample, and the second highest concentration was 23 MPN/100 mL. The highest concentration of fecal coliform bacteria measured was 2 MPN/100 mL.

The concentrations of total and fecal coliform bacteria measured at the outlet of the storage pond are summarized in Table 3. More than 1100 samples were analyzed over the five-year period. A total of 108 and 7 samples contained detectable concentrations of total and fecal coliform bacteria, respectively. On five occasions, the 7-d median concentration of total coliform bacteria was ≥ 2 MPN/100 mL. The 7-d median concentration of fecal coliform bacteria was never ≥ 2 MPN/100 mL. The results from periodic monitoring of pathogens are reported in Table 4. Over the five-year period, 18 grab samples were analyzed. No viral (total culturable virus) or bacterial (*E. Coli* 0157:H7, *Legionella*, *Salmonella*, *Shigella*) pathogens or helminth eggs were found in any of the samples. *Giardia* cysts and *Cyclospora* oocysts were found in one sample each, at concentrations of 0.1/L and 0.034/L, respectively. *Cryptosporidium* oocysts were found in 7 samples, with concentrations ranging from 0.3 to 2.3/L.

DISCUSSION

The California Title 22 regulations for irrigation of food crops require that the 7-d median concentration of total coliform bacteria is less than 2 MPN/100 mL, that the concentration in more than one individual sample does not exceed 23 MPN/100 mL in any 30-d period, and that the concentration in any individual sample never exceeds 240 MPN/100 mL (DHS, 2001). During five years of operation at the MRWPCA water reclamation facility, the 7-d median standard was exceeded on four occasions (three times in 1998 and once in 2000) and the maximum allowable concentration was exceeded once (900 MPN/100 mL in 1999) (Table 1). The 900 MPN/100 mL sample was believed to be caused by black bird droppings. To prevent further contamination from birds, the chlorine contact basins have since been covered and fishing line suspended above the railings. Since the covers were installed the standards have not been exceeded. Over the entire five years, there were no instances in which the concentration exceeded 23 MPN/100 mL in more than one sample in a 30-d period. These data illustrate that the plant has consistently and reliably complied with California's strict effluent reuse standards.

Table 1. Summary of data on the concentration of total coliform bacteria in the effluent of the west and east chlorine contact basins

Year	West Chlorine Contact Basin				East Chlorine Contact Basin			
	No. samples	No. ≥ 2 MPN		Maximum conc., MPN	No. samples	No. ≥ 2 MPN		Maximum conc., MPN
		Daily	7-d median			Daily	7-d median	
1998	191	36	3	8	74	6	0	8
1999	185	22	0	900[a]	209	24	0	4
2000	193	25	0	23[b]	202	24	1	13
2001	228	8	0	17	236	6	0	2
2002	247	13	0	4	266	7	0	13
2003	34	2	0	2	24	0	0	0
Summary	**1078**	**106**	**3**	**900**	**1011**	**67**	**1**	**13**

[a] 900 MPN/100 mL was measured in only one sample. The maximum concentration measured in the rest of the samples (in 1999) was 8 MPN/100 mL.
[b] 23 MPN/100 mL was measured in only one sample. The maximum concentration measured in the rest of the samples (in 2000) was 11 MPN/100 mL.

Table 2. Summary of data on the concentration of fecal coliform bacteria in the effluent of the west and east chlorine contact basins

Year	West Chlorine Contact Basin				East Chlorine Contact Basin			
	No. samples	No. ≥ 2 MPN		Maximum conc., MPN	No. samples	No. ≥ 2 MPN		Maximum conc., MPN
		Daily	7-d median			Daily	7-d median	
1998	202	3	0	2	76	0	0	0
1999	186	2	0	2	209	0	0	0
2000	191	2	0	2	201	2	0	2
2001	228	3	0	2	235	2	0	2
2002	246	2	0	2	316	0	0	0
2003	34	0	0	0	24	0	0	0
Summary	**1087**	**12**	**0**	**2**	**1061**	**4**	**0**	**2**

Table 3. Summary of data on the concentrations of total and fecal coliform bacteria at the outlet of the storage pond

Year	Total coliform				Fecal coliform			
	No. samples	No. ≥ 2 MPN		Maximum conc., MPN	No. samples	No. ≥ 2 MPN		Maximum conc., MPN
		Daily	7-d median			Daily	7-d median	
1998	191	8	0	240	191	2	0	2
1999	203	19	0	240	206	4	0	6
2000	209	46	2	220	209	1	0	70
2001	236	7	0	11	236	0	0	0
2002	267	26	3	80	267	0	0	0
2003	32	2	0	8	32	0	0	0
Summary	**1138**	**108**	**5**	**240**	**1141**	**7**	**0**	**70**

Table 4. Results of pathogen monitoring of disinfected tertiary recycled water from the Monterey Regional Plant. Samples in which organisms were detected are shown in bold face. Results shown with shaded background are from Sheikh et al. (1999).[a,b,c]

Sampling Date	Total Virus Culturable Virus/100L	Fecal Coliform, MPN /100 mL	*E. Coli* 0157:H7 CFU/100mL	*Legionella* Colonies per liter	*Salmonella* MPN/100mL	*Shigella* MPN/100mL	Cyclospora oocysts/L	*Giardia* cysts with internal structure/L	*Cryptosporidium* oocysts w / internal structure/L	Helminth (*Ascaris*) Ova/L
10/29-10/30/97	-	N.D.	<2	<100	-	-	-	-	N.D.	-
11/12-11/13/97	-	N.D.	<2	<100	-	-	-	-	-	-
11/17-11/18/97	-	N.D.	<2	<100	-	-	<0.011	<0.011	<0.011	-
11/24/97	-	N.D.	<2	<100	<2.2	-	<0.034	0.03	<0.034	-
12/2-12/3/97	-	N.D.	<10	ND	-	-	<0.019	0.08	<0.019	-
12/8-12/9/97	-	N.D.	<2	<100	<2.2	-	<0.011	0.09	<0.011	-
12/15-12/16/97	-	N.D.	<2	<100	<2.2	-	<0.0037	0.05	<0.0037	-
07/20-21/1998	<0.1	<2	<1	<1	<2.2	N.S.	<0.01	<0.01	<0.01	<0.5
06/03-04/1998	<0.1	<2	<1	<1	<2.2	N.S.	<0.001	<0.001	<0.001	<1
05/11-12/1998	<0.1	<2	<1	<1	<2.2	N.S.	<0.01	<0.01	<0.01	<1
05/25/1999	<0.1	<2	<1	<1	<2.2	<2.2	N.S.	<0.1	<0.1	<1
06/15/1999	<0.1	<2	<1	<1	<2.2	<2.2	<0.02	<0.1	<0.1	<1
07/19/1999	<0.1	<2	<1	<1	<2.2	<2.2	<0.008	<0.1	**0.3**	<1
08/17/1999	<0.2	<2	<1	<1	<2.2	<2.2	<0.03	**0.3**	<0.1	<1
09/20/1999	<0.1	<2	<1	<1	<2.2	<2.2	<0.05	<0.1	**1.7**	<1
10/26/1999	<0.1	<2	<1	<1	<2.2	<2.2	<0.02	<0.1	<0.1	<1
05/23/2000	<0.1	<2	<1	N.S.	< 2.2	<2.2	<0.08	<0.1	<0.1	<1
07/25/2000	<0.1	<1	<1	<20	<2.2	<2.2	<0.05	<0.1	**0.2**	<1
09/19/2000	<0.1	<2	<1	<20	<2.2	<2.2	<0.1	<0.1	<0.1	<1
05/30/2001	<0.2	<2	<2	<10	<2.2	<2.2	<0.1	<0.1	**0.2**	<1
07/25/2001	<0.1	<2	<2	<20	<2.2	<2.2	<0.04	<0.1	**2.3**	<1
09/25/2001	<0.1	<2	<2	<3	<2.2	<2.2	<0.03	<0.03	**0.2**	<1
06/26/2002	<0.1	<2	<2	<1	<2.2	<2.2	<0.12	<0.1	<0.1	<1
08/27/2002	<0.1	<2	<2	<0.33	<2.2	<2.2	**0.034**	<0.1	<0.1	<1
10/28/2002	<0.1	<2	<2	<0.33	<2.2	<2.2	<0.018	<0.1	**0.3**	<0.33

[a] The variation in detection limits was caused by differences in the original sample volume as well as the volume of concentrate resulting from any concentration steps.
[b] N.D. = Not Detected (but detection limit was not determined)
[c] N.S. = Not Sampled

Although the California Title 22 regulations are based solely on total coliform bacteria, most reuse guidelines outside of California are based on fecal coliform bacteria. For example, for non-processed food crops the USEPA recommends that the 7-d median concentration of fecal coliform bacteria is 0/100 mL, and that no sample exceeds 14 MPN/100 mL (USEPA, 1992). During five years of operation, neither of these criteria was ever exceeded at the MRWPCA reclamation plant.

The regulatory permit for the MRWPCA plant only requires monitoring of the chlorine contact basin effluent; nevertheless, the plant monitors the outlet of the storage pond to ensure that the water quality has not degraded before being sent to farmers for irrigation. The number of samples with detectable total and fecal coliform bacteria was similar in the storage

pond as in the chlorine contact basin effluent. However, the concentrations measured in the storage pond were often higher. The source of the bacteria is believed to be birds. Efforts to discourage birds from entering the pond or residing near it have also been taken.

Since startup, the plant has been operated to minimize the chance of discharge of any water that does not comply with the Title 22 standards. If a total coliform sample from the chlorine contact basin was found to contain more than 23 MPN/100 mL at the presumptive stage of analysis, the outlet gate at the storage pond was closed and the water was recirculated while increasing the chlorine residual by 2 mg/L. Extra samples were also collected near the pond outlet and tested. If the sample was confirmed to contain *less* than 23 MPN/100 mL total coliform bacteria, then the pond gate was reopened. If the sample was confirmed to contain *more* than 23 MPN/100 mL, however, the entire storage pond was drained, the pond bottom treated with hypochlorite, and the pond was refilled with water containing a higher chlorine residual. If a sample from the storage pond outlet was found to contain more than 23 MPN/100 mL, additional samples were taken and the chlorine residual was increased. The pond was only drained if a series of samples contained more than 23 MPN/100 mL.

The only pathogens that were detected in the tertiary effluent during the five-year monitoring effort were protozoan cysts (Table 4); one of 18 samples (5%) was positive for *Cyclospora*, one for *Giardia*, and seven of 18 (39%) for *Cryptosporidium*. These extremely low concentrations are not believed to represent a health risk given that the ingestion of 100 mL of water containing 5 *Giardia* or *Cryptosporidium* cysts (50 cysts/L) has been found to cause a risk of illness of 1 in 10,000 (Walker-Coleman et al., 2002). Furthermore, it is likely that many of the protozoan cysts in the tertiary effluent were not infective. In a study of eight water reclamation plants in Los Angeles County, all using filtration systems meeting the tertiary treatment definition of California Title 22 regulations, Garcia et al. (1999) reported a 99.8% removal rate of *Giardia* cysts, with final effluent samples containing an estimated average of 6 cysts/100 mL. Of these cysts, however, it was estimated that 97% of the cysts were non-viable based on morphology and vital stain exclusion. Follow-up infectivity tests were conducted by inoculating Mongolian gerbils, and it was determined that even a lower fraction of the cysts were infective (Garcia et al., 2002). Gerbils that were fed 1000 cysts recovered from tertiary effluent did not become infected.

The results from the pathogen monitoring are consistent with previous monitoring efforts. During the pilot-scale MWRSA experiments it was found that a 5-log removal of enteric virus could be achieved, mostly during the disinfection step (Sheikh *et al.*, 1990); thus, it is not surprising that no culturable viruses have been detected in the tertiary effluent at the full-scale plant. In the Food Safety Study, the concentrations of *E. Coli* O157:H7, *Legionella, Salmonella*, *Cyclospora*, *Giardia*, and *Cryptosporidium* were measured in the raw wastewater (plant influent) and tertiary effluent (Sheikh *et al*., 1999). No *E. Coli* O157:H7 or *Legionella* were detected in any influent (or effluent) sample, and only *Giardia* cysts were consistently present in the influent. The only organism detected in the effluent was *Giardia* cysts (see Table 4). These results are slightly different from what was observed during the present study, in which *Cryptosporidium* was found more frequently than *Giardia*. Based on the influent concentrations of the cysts, it was concluded that a 5 to 6-log removal of cysts occurred at the treatment plant.

Further evidence of cyst removal is provided by the independent monitoring of *Clostridium perfringens* conducted by the Monterey County Environmental Health Department (MCEHD). *Clostridium* endospores have been proposed as an indicator for protozoan cysts,

such as *Giardia* and *Cryptosporidium* (Bonadonna et al., 2002; Rose et al., 2001; Payment et al., 1993). Since 1999, MCEHD has collected grab samples of the secondary effluent and either the tertiary effluent or the storage pond every two weeks; the results are reported in Table 5 (MCEHD, 1999, 2000). A 5 to 6-log removal has been observed, consistent with the removal of *Giardia* cysts. MCEHD also measured the total coliform concentration in the storage pond outlet starting in 1998, and the results have been consistent with the MRWPCA data.

Table 5. Concentration of *Clostridium perfringens* in secondary, tertiary, and storage pond effluents, and calculated removal rate through the treatment process (MCEHD 1999, 2000)

Year	Secondary effluent*, (CFU/100mL)	Tertiary effluent (CFU/100mL)	Pond (CFU/100mL)	Log Reduction (Tenth Power)	Reduction Calculated through
1999	900 – 14,800	<0.01	Not Sampled	4.3 – 5.4	Tertiary
2000	1,700 – 1,200	<0.03	Not Detected	4.4 – 5.4	Tertiary
2001	1,900 – 5,600	<0.03 – 2.3	<0.03	3.1 – 4.9	Tertiary
2002	2,800 –37,500	Not Sampled	<0.03	4.9 – 6.1	Pond
2003	40,000 – 48,000	Not Sampled	<0.03	6.1 – 6.2	Pond

* In August, 2002, the sampling was switched to the primary effluent so that higher removal could be demonstrated.

CONCLUSIONS

After five years of full-scale operation, it is concluded that the MRWPCA water reclamation plant consistently and reliably produced water meeting the California Title 22 standards. In addition, no viral or bacterial pathogens have been detected in the tertiary effluent, and the intermittent concentrations of protozoan cysts detected present a negligible health risk. These data contribute to a growing body of evidence (e.g., York, 1998) illustrating that recycled water is at least as safe, and in some cases safer, as other sources of irrigation water for growing food crops eaten raw.

Additional research is now being conducted at the MRWPCA water recycling facility to determine if adequate treatment can be provided with filter loading rates above the current maximum rate of 12.2 m/hr (5 gal/ft^2-min). The study began in May 2003 and is expected to take three years to complete. In the first phase, pilot filters will be loaded at several rates using identical influent, and the effluent will be tested for a variety of parameters, including indicator organisms. In the second phase, full-scale filters at the MRWPCA Treatment Plant and four other treatment plants in California will be loaded at higher rates and tested in a similar fashion. The ultimate criterion of optimum rate of filter flux will be the ability of the combined filtration-disinfection system to remove and inactivate pathogens consistently and reliably.

REFERENCES

AOAC, 1995. Official Methods of Analysis of the Association of Official Agricultural Chemists, 16th edition, Arlington, Va.

Bonadonna, L., R. Briancesco, C. Cataldo, M. Divizia, D. Donia, and A. Pana, 2002. “Fate of bacterial indicators, viruses and protozoan parasites in a wastewater multi-component treatment system.” *Microbiologica*, 25(4), 413-420.

DHS, 2001. “Title 22 Code of Regulations, Division 4, Chapter 3: Water Recycling Criteria” in *California Health Laws Related to Recycled Water: Excerpts from the Health and Safety*

Code, Water Code, and Titles 22 and 17 of the California Code of Regulations ("The Purple Book"). Available at the California Department of Health Services website: http://www.dhs.ca.gov/ps/ddwem/publications/waterrecycling/purplebookupdate6-01.PDF (accessed on 5/19/03).

Greenberg, A. E., L. S. Clesceri, and A. D. Eaton, eds., 1992. *Standard Methods for the Examination of Water and Wastewater*. 18th Ed. American Public Health Association, American Water Works Association, and Water Environment Federation.

Garcia, A., Walker, A., Yanko, W. 1999. "Removal and Fate of Giardia Cysts in Water Reclamation Plants", *Proceedings of the 72nd Annual Water Environment Federation Technical Exposition and Conference* [CD-ROM], New Orleans, Louisiana, Oct 10-13; Water Environment Federation, Alexandria, Virginia.

Garcia, April, William Yanko, Glenda Batzer, Giovanni Widmer 2002. "Giardia Cysts in Tertiary-Treated Wastewater Effluents: Are They Infective?" Water Environment Research, 74 (6): 541-544.

MCEHD, 2000. "Annual Tertiary Monitoring Report for 2000", Monterey County Environmental Health Department unpublished internal report, available from the Department upon request.

MCEHD, 1999. "Annual Tertiary Monitoring Report for 1999", Monterey County Environmental Health Department unpublished internal report, available from the Department upon request.

Payment, P., and E. Franco. 1993. "Clostridium perfringens and somatic coliphages as indicators of the efficiency of drinking water treatment for viruses and protozoan cysts." *Appl. Environ. Microbiol.*, 59(8), 2418-2424.

Rose, J. B., D. E. Huffman, K. Riley, S. R. Farrah, J. O. Lukasik, and C. L. Hamann, 2001. "Reduction of enteric microorganisms at the Upper Occoquan Sewage Authority Water Reclamation Plant." *Water Environment Research*, 73(6), 711-720.

Sheikh, B., Cooper, R.C. Israel, K. E. 1999. "Hygienic Evaluation of Reclaimed Water Used to Irrigate Food Crops—A Case Study", *Water Science and Technology*, **40**(4-5):261-267.

Sheikh, B., Cort, R. P., Kirkpatrick, W. R., Jaques, R.S., Asano, T., 1990. "Monterey Wastewater Reclamation Study for Agriculture." *Research Journal WPCF*, **26**(3): 216-226.

USEPA, 1992. "Guidelines for Water Reuse." *EPA/625/R-92/004*, U.S. Environmental Protection Agency, Center for Environmental Research Information, Cincinnati, Ohio.

Walker-Coleman, L., York, D. W., Menendez, P., 2002. **"**Protozoan Pathogen Monitoring Results For Florida's Reuse Systems", Proceedings, WateReuse Symposium XVII, September 8-11, 2002, p. 2, Orlando, Florida

Yanko, William A., Analysis of 10 Years of Virus Monitoring Data from Los Angeles County Treatment Plants Meeting California Wastewater Reclamation Criteria, Water Environment Research, Vol. 65, No. 3, May/June 1993, pp. 221-226

York, David W., "Protozoan Pathogens: A Comparison of Reclaimed Water and Other Irrigation Waters", in Conference Proceedings of Water Reuse '98, Joint AWWA-WEF sponsored conference in Orlando, Florida, February 1-4, 1998.