**Supplementary Material**

**Conventional wastewater treatment and reuse site practices modify bacterial community structure but do not eliminate some opportunistic pathogens in reclaimed water**

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**Supplementary Text**

Detailed Description of Sampling Sites

Treatment processes utilized at Mid-Atlantic WWTP1, an urban tertiary wastewater treatment plant processing 681,390 m3 of wastewater (including domestic and hospital wastewater) per day (Rosenberg Goldstein et al., 2012; Rosenberg Goldstein et al., 2014), are screens, primary clarifier, activated sludge reactors, secondary clarifier, sand filters, chlorination (2-3 mg/L), de-chlorination and effluent discharge (<0.1 mg/L chlorine residual) (Rosenberg Goldstein et al., 2012; Rosenberg Goldstein et al., 2014). A portion of the treated effluent from this plant is transported, through an enclosed pipe, to Mid-Atlantic SI1 for spray irrigation (Rosenberg Goldstein et al., 2014) where it undergoes screening and ultraviolet (UV) disinfection (>30,000 µW/cm2, 254nm UV wavelength) followed by storage in an open-air pond (peak capacity 15141.65 m3) (Rosenberg Goldstein et al., 2014). Water from the storage pond is then pumped to spray heads as needed (Rosenberg Goldstein et al., 2014). Backpack sprayers are used to apply reclaimed water to locations not covered by spray heads (Rosenberg Goldstein et al., 2014).

Treatment processes at Mid-Atlantic WWTP2, a suburban tertiary wastewater treatment plant processing 7,570 m3 of wastewater (including domestic and hospital wastewater) per day (Rosenberg Goldstein et al., 2012; Rosenberg Goldstein et al., 2014), are screens, primary clarifier, primary aeration tank, secondary aeration tank, secondary clarifier, multimedia filter, chlorination (2-3 mg/L), de-chlorination and effluent discharge (< 0.1 mg/L chlorine residual) with a portion of the effluent being transported to a landscaping site for re-use via spray irrigation (Rosenberg Goldstein et al., 2012; Rosenberg Goldstein et al., 2014).

Midwest WWTP1 is a rural tertiary treatment plant processing 1,363 m3 of wastewater (including domestic wastewater and agriculturally influenced storm-water) per day, with treatment processes being screens, activated sludge lagoons, clarifiers, seasonal chlorination and de-chlorination (4 mg/L in June, July and August) and effluent discharge (chlorine residual of 0 mg/L) (Rosenberg Goldstein et al., 2012; Rosenberg Goldstein et al., 2014). A portion of effluent from this plant is transported to a landscaping site for re-use via spray irrigation (Rosenberg Goldstein et al., 2012; Rosenberg Goldstein et al., 2014).

Midwest WWTP2 is a rural tertiary treatment plant processing 1,439 m3 of wastewater (domestic, food production and agriculturally influenced wastewater) per day with treatment processes being screens, sequencing batch reactor, lagoon cell A, lagoon cell B, lagoon cell C, lagoon cell D, lagoon cell E, and effluent discharge (Rosenberg Goldstein et al., 2012; Rosenberg Goldstein et al., 2014). There is no on-site disinfection and unchlorinated effluent is transported to an agricultural site for irrigation of fodder crops (Rosenberg Goldstein et al., 2012; Rosenberg Goldstein et al., 2014).

**Supplementary Figure Legends**

**Figure S1** Number of observed sequences and Good’s coverage, a metric used to estimate sample coverage, for samples sequenced. Samples with fewer than 100 sequences were filtered.



**Figure S2** Alpha-diversity estimates and observed operational taxonomic unit (OTU) number in influent samples from all four wastewater treatment plants (WWTPs). No statistically significant differences in alpha-diversity estimates were found across influent samples from all four wastewater treatment plants.

