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Detection of Environmental Sources of Infectious Diseases in Groundwater Networks (DESIGN) – Cryptosporidium and VTEC Incidence in the Republic of Ireland.

Carlos Chique[1,2], Paul Hynds[3], Liam Burke[4], Dearbháile Morris[4], Michael Ryan[5], and Jean O'Dwyer[1,2]

[1]School of Biological, Earth and Environmental Science (BEES), University College Cork, Cork, Ireland
[2]Water and Environment Research Group, Environmental Research Institute, University College Cork, Cork, Ireland
[3]Environmental Sustainability and Health Institute (ESIH), Technological University Dublin, Dublin, Ireland
[4]The Antimicrobial Resistance and Microbial Ecology Group, National University of Ireland, Galway, Galway, Ireland
[5]Department of Chemical Sciences, University of Limerick, Limerick, Ireland

Approximately 500 million Europeans use a groundwater source for water consumption on a daily basis. Private (unregulated) groundwater wells are key sources of domestic drinking water in the Republic of Ireland (ROI) with approximately 750,000 users. The distribution of groundwater wells in the ROI is highly concentrated in rural areas in correspondence with the absence of piped infrastructure. The nexus of key (extra-)local factors, including high private groundwater reliance, ubiquity of domestic wastewater treatment systems and pastoral agriculture in rural areas, in conjunction with a temperate maritime climate and distinctive hydro(geo)logical settings, has been linked to a high groundwater susceptibility to contamination and risk of waterborne enteric infection.

DESIGN focuses on the incidence and sources of verotoxigenic-producing Escherichia coli (VTEC) and *Cryptosporidium* spp. – the most prevalent waterborne pathogens inducing enteric illness in the ROI – associated with private groundwater wells. The findings of a systematic literature review focusing on the prevalence of *Cryptosporidium* in domestic groundwater supplies are presented. Calculated detection rates for groundwater wells (19%) and samples (13%) indicate *Cryptosporidium* spp. contamination of domestic groundwater supplies is common, representing a latent health risk of direct concern to groundwater consumers and public health authorities. Presented figures provide unprecedented "baselines" highly applicable in groundwater/catchment management and epidemiology (e.g., QMRA). Several knowledge gaps were identified with the lack of standardized reporting among investigations emerging as a key concern.

The results of temporal (i.e., repeat) sampling regimes analysing the spatio-temporal incidence of *Cryptosporidium* and VTEC in groundwater wells are also presented. Sampling locations (n = 80) were geo-referenced and linked to multiple variables (e.g., land-use, agricultural statistics, hydrogeology) compiled in a novel geo-database. In conjunction with supply infrastructural data, relevant risk factors associated with VTEC and *Cryptosporidium* well contamination were

identified. Furthermore, incorporating previously available data from project partners, stochastic QMRA and Environmental Fate Model(s) were produced to assess the relative risk of VTEC well contamination and seasonal influence. The explorative *Cryptosporidium* sampling regime provides the first national account of (oocyst) incidence in domestic groundwater infrastructure enabling insights into potential environmental sources and (surface-groundwater) transport mechanisms. The results obtained represent a stepping stone towards the development of bespoke groundwater management strategies in the ROI based on the 'One Health' concept.