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Managing microbial risks of drinking water and sanitation to prevent waterborne infectious diseases

Beheersen van microbiologische risico’s van drinkwater en sanitaire voorzieningen om wateroverdraagbare infectieziekten te voorkomen

(met een samenvatting in het Nederlands)

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Chapter 1

General introduction

Contaminated drinking water and poor sanitation are linked to transmission of diseases such as cholera, dysentery, hepatitis A, typhoid and polio (Howard and Bartram, 2003). Globally, waterborne pathogens and water-related diseases are a major public health concern, not only due to the morbidity and mortality they cause, but also as a result of the high costs that represents their prevention and treatment (Ramírez-Castillo *et al.*, 2015). Unsafe drinking water and sanitation are major causes for the spread of waterborne diseases in a community (WHO and UNICEF, 2021). Therefore, reliable access to clean drinking water and sanitation are the main means to prevent waterborne diseases. Although considerable improvements have been achieved globally, still two billion people lacked safely managed drinking water in their homes and 3.6 billion people lacked safely managed sanitation services in 2020 (WHO and UNICEF, 2021).

Drinking water supply is the provision of drinking water from catchment and source(s) through treatment to the point of consumption (WHO 2017b). Safely managed drinking water is located on premises, available when needed and free of contamination. The drinking water comes from ‘improved’ sources that are potentially capable of delivering safe water by nature of their design and construction (UNICEF and WHO, 2023).

Sanitation is the management of human excreta and wastewater from all steps of the sanitation chain, from toilet capture and containment, through emptying and transport of excreta for treatment and final disposal or end use (e.g. for agricultural purposes) (Tilley *et al.*, 2014; WHO 2018). Sanitation systems include the technologies and services for the management of human excreta and wastewater (Tilley *et al.*, 2014). Safely managed sanitation is the use of improved facilities that are not shared with other households and where excreta are safely disposed of in situ or transported and treated off-site (UNICEF and WHO, 2023).

Human excreta and domestic wastewater contain numerous bacterial, viral, and protozoan pathogens (Leclerc *et al.*, 2002), which can be transmitted to the environment through various pathways. Contamination of water and exposure to contaminated water, can happen within the complete drinking water supply and sanitation chain. If the pathogens survive long enough they may cause diseases when a susceptible host gets exposed through direct contact with contaminated water, ingestion of the water or inhalation of aerosols (Daley *et al*., 2018).

People directly exposed to treated and untreated wastewater are sanitation workers, maintaining the sanitation systems or working at wastewater treatment plants, and farmers using (treated) wastewater for irrigation or sludge as fertilizer. There is suggestive evidence of elevated occupational risk among sanitation workers for a range of health conditions (Oza *et al.*, 2022). They occasionally face serious health and safety issues, which are increased by the lack of adequate protective equipment (Philippe *et al.*, 2022). Inappropriate reuse of wastewater, as end-product of the sanitation chain, has caused viral outbreaks worldwide (Sano *et al*., 2016). Exposure to wastewater through agricultural irrigation resulted in skin irritation, rashes, and dermatitis among farmers (Dickin *et al*., 2016).

Surface waters often become microbially contaminated through discharges of treated wastewater or combined sewer overflows (Demeter *et al*., 2021). The use of such fecally contaminated surface waters for drinking water production, shellfish culture, domestic, irrigation and bathing purposes poses a potential health risk (Figure 1.1). In total, thirteen waterborne outbreaks were reported caused by contaminated surface waters mainly related wastewater (Moreira and Bondelind, 2017). In two large outbreaks in Sweden, 49,400 people were infected due to consumption of drinking water produced from surface water contaminated with wastewater (Moreira and Bondelind, 2017).

Intrusion of wastewater into groundwater supplies due to heavy rainfall or discharge of wastewater may result in contaminated drinking water sources (Fong *et al*., 2007; Moreira and Bondelind, 2017). Contamination of groundwater supplies can be persistent as shown in two waterborne outbreaks in Finland (Kauppinen *et al*., 2018).

Pathogens, such as *Legionella*, can be transmitted via bioaerosols generated during wastewater treatment posing a health risk to workers or to habitants of their surroundings (Korzeniewska, 2011). In the Netherlands, a wastewater treatment plant at a food processing company was identified as the likely source of contamination for an observed local increase of pneumonia caused by *Legionella pneumophila* (Loenenbach *et al*., 2018).

Drinking water and sanitation play a role in the transmission of infectious diseases through various transmission routes (Figure 1.1). Drinking water supply and sanitation services face threats, such as aging infrastructure, urbanization and climate changes, which may affect the transmission of microbial hazards. Drinking water suppliers and sanitation providers need to know and understand these hazards and the associated risks these hazards pose to the drinking water supply and sanitation system and they have to manage these risks well. Absent, inadequate, or inappropriately managed drinking water and sanitation services subject individuals to preventable health risks (WHO and UNICEF, 2021). Risk management, including risk assessment and water quality monitoring, plays an important role in safely managing drinking water supply and sanitation services. Water quality monitoring, risk assessment and risk management will be explained in the following paragraphs.

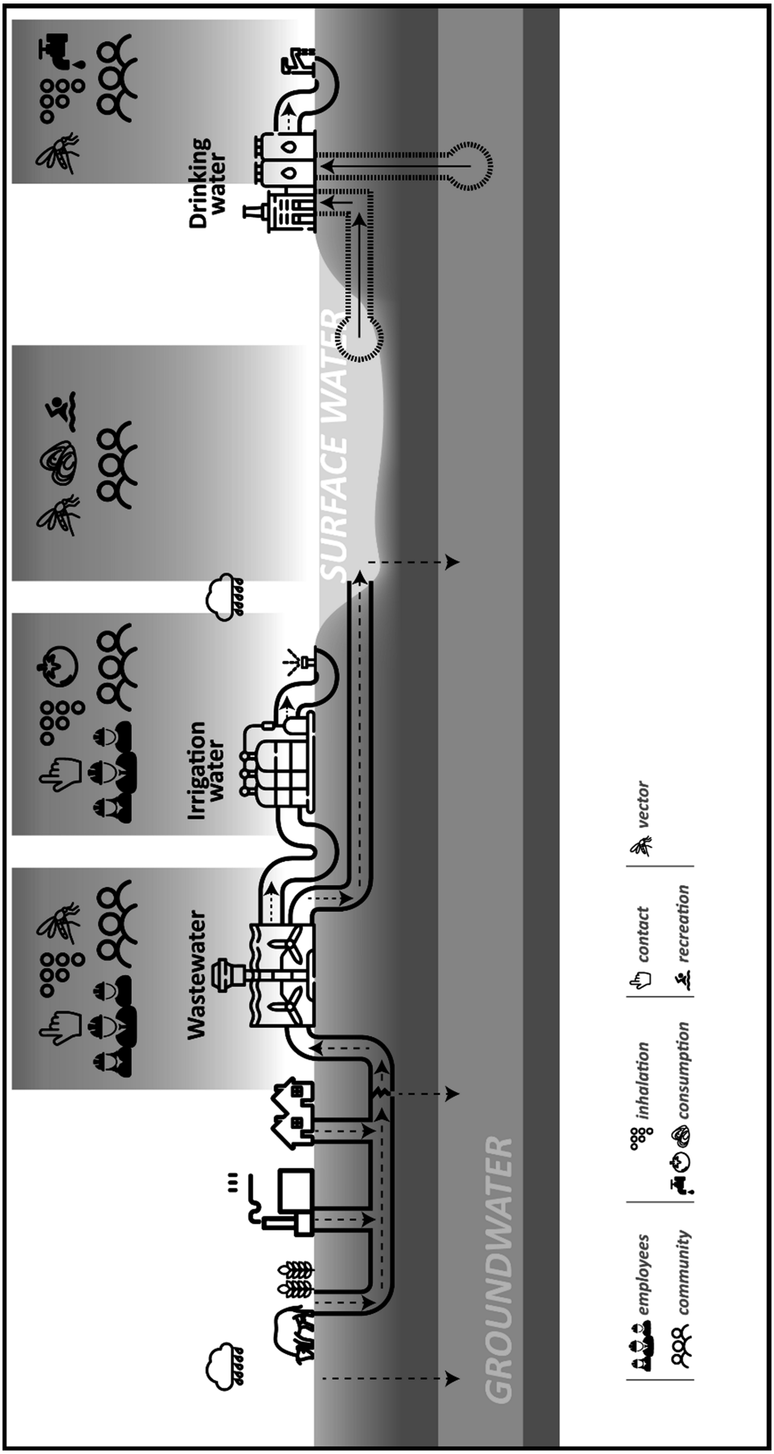

Figure 1.1. *Role of water in transmitting pathogens.*

Water quality monitoring

Monitoring of the microbial quality of drinking water or wastewater is a key element ensuring drinking water and sanitation safety by revealing (early) signals of hazards to initiate remedial actions. In this way, water quality monitoring provides information for risk management and contributes to the prevention of waterborne infectious diseases, and thus protects public health. Different purposes of water quality monitoring are available which are able to effectively support the sustainable supply of safe drinking water and safe sanitation. The different purposes of water quality monitoring are shown in Figure 1.2 and described in more detail in the following paragraphs.

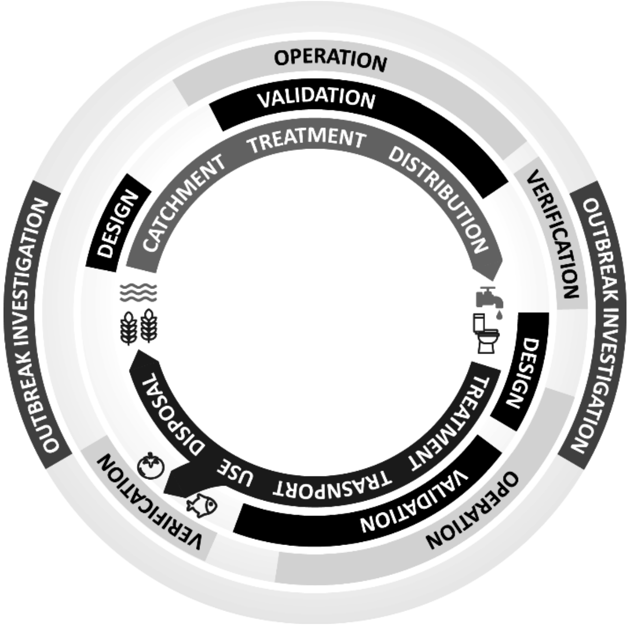

Figure 1.2. *Different purposes of water quality monitoring in the drinking water supply and sanitation chain.*

Design

For the purpose of designing water treatment processes for drinking water supply or sanitation services, it is important to understand source water quality and its variations, as well as pathways of contamination. To protect public health, reliable information on microbial hazards and possible risks affecting source water quality is needed to select or prioritize drinking water sources, develop adequate control measures or design treatment facilities (WHO 2016). Data from source water quality monitoring, also called raw water monitoring, can also identify existing hazards or provide an early warning of source water contamination (Schilderman *et al.*, 1999; Lodder *et al.*, 2010; WHO 2016).

Validation

Validating existing control measures is needed to determine whether a control measure is capable of effectively controlling the hazard or hazardous event. Expert judgement or validation monitoring may be carried out for validation purposes (WHO 2023). Validation monitoring can be done by reviewing existing water quality monitoring data for example, through analysis of online monitoring data that show the history of non-compliant water quality at the outlet of a treatment plant (WHO 2023). If existing data are not available, validation monitoring could be carried out to collect specific information on the performance of control measures, for example through monitoring of the removal of specific pathogens (Van den Berg *et al*., 2005), or laboratory and/or pilot scale experiments (Torkzaban *et al*., 2006; Schijven *et al*., 2013) using both indicators and pathogens.

Operation

Operational monitoring is a planned and routine set of activities used to determine that those steps or processes in the water supply chain that directly affect water quality (control measures) continue to work effectively (WHO 2023). In operational monitoring, the drinking water supplier monitors each control measure in a timely manner to enable effective system management, including taking corrective actions to bring the control measure back to proper operation when necessary. For example, wherever chlorination is practiced as control measure frequent operational monitoring of residual chlorine is recommended (WHO 2017b).

Verification

Compliance monitoring is a final check of the water to verify that the water supplied to consumers or discharged is in compliance with its quality requirements. It confirms whether or not the health-based targets are met and should be conducted by an independent surveillance agency, such as inspectorates, or the water supplier itself with the agreement of the surveillance agency (WHO 2023). Compliance monitoring of the microbial quality of drinking water typically includes testing for *E. coli* as an indicator of fecal pollution, which must be absent in 100 ml of the drinking water (WHO 2017b). Drinking water surveillance is the continuous and vigilant public health assessment and review of the safety and acceptability of drinking water (WHO 2019). Drinking water surveillance should be performed for drinking water supplies operated by drinking water utilities as well as supplies managed by communities (WHO 2019).

Outbreak investigation

If disease outbreaks occur, water quality testing can be applied to investigate the source of the outbreak and the effectiveness of measures taken to control the outbreak. Improving the understanding of the contribution of various fecal sources to contamination, the possible source of pathogens of concern and their transmission routes may aid the prevention of infectious disease outbreaks in the future. A gastroenteritis outbreak was observed in a new housing estate in the Netherlands (Fernandes *et al*., 2007). The outbreak could be associated with the consumption of drinking water contaminated with greywater due to human error in the exposed area. Based on this outbreak, the use of grey water was banned (Fernandes *et al*., 2007). Cases of Legionnaires' disease were reported which were linked to a specific area in the Netherlands. Outbreak investigation showed that a biological wastewater treatment plant was identified as the likely source for this outbreak, and measures could be taken to protect public health (Loenenbach *et al*., 2018).

Limitations of water quality monitoring

Many different pathogens can be present in water. Even low numbers of pathogens though difficult to detect can still cause undesired levels of infections through exposure to water. Measurements of pathogens in water require many resources such as human capacity, disposables and funding. Therefore other microorganisms are used as indicators of fecal contamination that are easier to detect. Mostly non-pathogenic bacteria that occur in large numbers in feces are used. *E. coli* and thermotolerant coliforms are the most common indicators. Testing for fecal indicators is useful for knowing that water quality complies with legal standards (compliance monitoring). To guarantee safe drinking water supply and sanitation services, providers and competent authorities mainly rely on operational and compliance monitoring. Also regulations focus on the detection of indicators, e.g. the absence of *E. coli* in 100 ml drinking water (EU 2020) or the *E. coli* concentration for the reuse of treated wastewater (EU 2022).

Although monitoring of water as an end-product has been the standard to determine if drinking water is safe and clean, over the past decades it has become clear that this monitoring is often too little and too late (WHO 2015). While bacterial fecal indicators play an important role in verifying compliance with water-quality standards, overreliance on microbiological compliance monitoring has its limitations (WHO 2019), such as:

- compliance monitoring (or end-product testing) tests only a small amount of water as compared to the total amount of water supplied;
- pathogens can be more resistant to disinfection and might be more persistent in the environment compared to indicators;
- waterborne disease outbreaks still occur even when no fecal indicators are present in drinking water;
- compliance monitoring is not capable of detecting short term fluctuations in water quality; and
- compliance monitoring is reactive and has no early warning capability.

To avoid overreliance on compliance monitoring and better protect human health, a holistic and proactive approach is needed, such as risk assessment and risk management (WHO 2017b, 2018). This approach moves from detecting risk to preventing risk. Risk assessment and risk management have become more predominant in legislation. The European drinking water directive prescribes a risk based approach for water safety (EU 2020). A survey from WHO in 2017 showed that out of 100 countries, 46 countries reported having policy or regulatory instruments that promote or require risk assessment and risk management and in another 23 countries such instruments are under development (WHO 2017a). Water quality monitoring is still an important component and is part of risk management. The different purposes of water quality monitoring are able to effectively support these risk management approaches to sustainably supply safe (drinking) water and sanitation.

Risk assessment

Risk assessment is an important tool to manage risks and hazardous events in drinking water supplies and sanitation systems since it identifies microbial hazards and risks timely and is therefore an essential part of public health protection (WHO 2017b, 2018). Drinking water suppliers and sanitation providers need to understand these hazards and the associated risks these hazards pose to drinking water supplies and sanitation systems. For many years, principles and methods have been developed to conceptualize, assess and manage risks. The risk assessment methodologies identify significant risks and their drivers. A significant number of tools are currently available for risk analysis in water systems, both for qualitative and quantitative methods (Kombo Mpindou *et al.*, 2022).

Sanitary and sanitation surveys can be used as a **qualitative** risk assessment tool. These surveys are visual on-site inspections to identify risk factors that can pose a threat to the wellbeing and health of sanitation workers and drinking water consumers. Sanitary inspection is applied in low-, medium- as well as high-income settings to assess the risk of microbial contamination of water sources. In a sanitary inspection a short, standard form is used to evaluate the physical structure and operation of the system and external environmental factors that may contribute to contamination in water supply systems (WHO 2020). For example, a sanitary inspection may reveal a broken pump or pipeline, or the presence of animals with access to the source. Resulting actions serve to improve or protect the water supply (Bacci and Chapman, 2011; Mushi *et al.*, 2012). Sanitation inspections can be used to assess risk factors at or near sanitation facilities and identify appropriate actions to safeguard public health (WHO 2022a). Sanitary inspections have some limitations, as the short, standard form is not comprehensive, and may not include every factor that might contribute to microbial contamination of the source (Kelly *et al.*, 2020). Another limitation is that the risk score is based on the presence or absence of risk factors and the risk factors are not weighed to determine the risk scores (Howard *et al*., 2003).

Risk assessment is a key step to assess the infrastructure condition as part of asset assessment for drinking water, wastewater or stormwater infrastructure (Harvey *et al*., 2017). Asset assessment provides the basis for lowering infrastructure renewal costs, identifying approaches to extend asset life, and identifying funding options for sustained growth (Harvey *et al*., 2017). Different risk analysis methods can be applied in asset assessment, such as failure mode and effects analysis (FMEA), failure mode effect and criticality analysis (FMECA), hazard analysis critical control point (HACCP) and hazard and operability study (HAZOP). In a recent study in Kenya, FMECA was successfully used to assess drinking water supply systems and identify risks from source to point-of-use for different water sources: hand-dug wells, boreholes and public water supply sources (Odjegba *et al*., 2023). Also in sanitation systems, managers adopt proactive and preventive maintenance (proactive asset management) to reduce disturbance to public health and environment (Baah *et al*., 2015). Risk assessment of sewer pipes requires integration of the likelihood and consequences of failure to reflect the perception of risk (Salman and Salem, 2012). By identifying high failure risk areas, inspections can be implemented based on the system status and thus can significantly increase the sewer network performance (Salman and Salem, 2012; Anbari *et al*., 2017). Proactive sewer asset management assesses the vulnerability of the infrastructure and predicts the risk of failure (Noshahri *et al*., 2021).

A **quantitative method** to assess the microbial risks is quantitative microbial risk assessment (QMRA). QMRA is a structured, systematic, science-based approach to quantitatively estimate risk of infection or illness based on the level of exposure to microbial hazards (Teunis and Schijven, 2018). QMRA combines multiple aspects such as the presence of pathogens, the health effects that may result from exposure and the effect of natural and engineered barriers. QMRA can be applied in drinking water and sanitation systems. The performance of conventional drinking water treatment can be improved using QMRA (Hadi *et al*., 2019). The Dutch Drinking Water Decree (I&W 2011) prescribes that the index pathogens enterovirus, *Cryptosporidium*, *Giardia* and *Campylobacter* should not exceed an infection risk of one infection per 10,000 individuals per year. To demonstrate the microbial safety of drinking water, Dutch drinking water companies must conduct a QMRA at least every four years for these so-called index pathogens (Schijven *et al*., 2011). For sanitation, a fully developed QMRA exists to support decision-makers in selecting appropriate wastewater treatment system designs, quantifying and prioritizing public health risks (Daley *et al*., 2018). Finally, QMRA can be used to assess microbial risks from consumption of raw or slightly cooked vegetables irrigated with undiluted, disinfected wastewater effluent (Hamilton *et al*., 2006a; Hamadieh *et al*., 2021).

Risk management

Managing risks has been important in different areas. To ensure safe food with an extended shelf life for space travel the first HACCP concept was developed in the 1960s by the U.S. National Aeronautics and Space Administration (Weinroth *et al*., 2018). This remains the principal management concept to reduce risks of foodborne illness (Weinroth *et al*., 2018). In 1994, Havelaar explored the application of HACCP to drinking water supply systems (Havelaar, 1994). In some countries, for example Switzerland, the drinking water supply was also regulated through the law for food protection and therefore already required HACCP. Westrell *et al*. (2004) used HACCP to identify and control exposure to pathogens at a wastewater treatment plant. Between 1999 and 2001, an international group of experts discussed the potential to increase consistency in approaches of assessment and management of water-related microbial hazards, which led to the 'Stockholm Framework' (Fewtrell and Bartram, 2001). This framework further explored the possible application of HACCP to the drinking water supply. The World Health Organization (WHO) established a 'Framework for Safe Drinking water' that encompasses setting health-based targets, a risk assessment and risk management approach, and a system of independent surveillance monitoring (WHO 2011). A water safety plan (WSP) is the risk assessment and risk management approach recommended by WHO (WHO 2017b). WSPs encompass all steps in a drinking water supply system from catchment to consumer and their use should ensure continual and sustainable provision of water that is safe for human consumption and meets regulatory water standards relating to human health (WHO 2023). At the same time, the International Water Association (IWA) published the Bonn Charter for Safe Drinking Water, which provides a high-level framework describing the operational and institutional arrangements that are basic requirements for managing water supplies from catchment to consumer (IWA 2004). Microbial contamination of a drinking water system in Walkerton (Canada) had a huge impact on a small community, with seven people who died and more than 2,300 who became ill (O'Connor, 2002). After this outbreak risk management for safe drinking water was recommended (Hamilton *et al*., 2006b; Hrudey *et al*., 2006). Similar to the framework for safe drinking water, the risk management approach is preventive rather than reactive to identify and to manage risks to public health. Water quality monitoring and risk assessment are both highlighted as part of the risk management approaches (Hamilton *et al.,* 2006b; WHO 2011).

The WHO Guidelines on Sanitation and Health provide comprehensive advice on maximizing the health impact of sanitation interventions and ensure universal access to safe sanitation systems (WHO 2018). Safe management of the sanitation service chain is essential for protecting human health and water resources. A sanitation safety plan (SSP), based on the same concept as the WSPs, supports the implementation of WHO's Guidelines on Sanitation and Health (WHO 2018) and Guidelines for the safe use of wastewater, excreta and greywater (WHO 2006). It provides a risk assessment and risk management framework to protect human health from sanitation-related risks, including from reuse of wastewater in agriculture and aquaculture (WHO 2015). Limited case studies for SSP are available (Domini, 2017; Winkler *et al.*, 2017; Halalsheh *et al.*, 2018; Frattarola *et al.*, 2019).

Benefits of the implementation of risk management approaches, such as WSP and HACCP, are improved water quality and operational efficiency. Implementation of these approaches also showed reduced consumers' complaints, production costs, and potential hazardous incidents (WHO 2017a; Tsitsifli and Tsoukalas, 2021). WSP implementation in Iceland resulted in a significant decrease in diarrhea incidence (14%) (Gunnarsdottir *et al.*, 2012). In France and Spain, at only one of the three locations evidence was provided for reduced acute gastroenteritis incidence after a WSP was implemented (Setty *et al.*, 2017). For successful implementation of a WSP relevant factors are critical, such as financial and human resources, staff training, effective hazard and hazardous event identification, correct assessment of the risk based on the occurrence and the severity of the hazards, and efficient monitoring (WHO 2017a; Tsitsifli and Tsoukalas, 2021).

Drinking water supplies and sanitation systems are closely related. Important interactions between the two systems may lead to risks, for example:

- cross-contamination of the drinking water supply across the sanitation service chain and consumption of contaminated drinking water (Narayan *et al.*, 2021);
- on-site sanitation may cause penetration of contamination into groundwater sources used for drinking water, despite soil media filtration (Pitkänen *et al.*, 2011); and
- interrupted water supply causes toilet flushing to not work.

To date, risk management frameworks have addressed drinking water supply and sanitation systems separately. In smaller and more local contexts, drinking water and sanitation are more naturally interlinked, partly due to their close proximity. The same people might even take care of both systems. For rural communities with limited human, financial and administrative resources, the implementation of risk management approaches is not straightforward and support is required (Herschan *et al.*, 2020). Information on an integrated approach and details of its implementation are scarce (Clavijo *et al.*, 2020; Murei *et al.*, 2022).

Combining risk assessment and water quality monitoring

Risk assessment and water quality monitoring are both parts of risk management, and can support each other. Some risk assessment methodologies include water quality data and advocate for water quality monitoring. For example, QMRA strongly relies on water quality data. Source water monitoring should be applied to achieve a representative quantification of the numbers of pathogenic microorganisms in the source water (Schijven *et al.*, 2011). The monitoring should include seasonal variability as well as short term fluctuations of pathogen concentrations (Westrell *et al.*, 2006). Furthermore, data should be collected for each step in the drinking water supply QMRA to quantify the efficiency of the treatment (Schijven *et al.*, 2011), so-called validation monitoring.

A methodology in which both water quality monitoring and risk assessment are combined is the rapid assessment of drinking water quality (RADWQ) (WHO 2012). For this method, drinking water sources are monitored once and at the same time sanitary inspections are carried out as qualitative risk assessment (WHO 2012). The approach provides a nationally representative dataset to create a snapshot of the level of correlation between the designation “improved” of a source and the quality of the drinking water it provides in reality (WHO 2012). Sanitary inspection and water quality analysis are distinct and complementary tools, and both serve important purposes in the on-going process of ensuring water safety (Kelly *et al.*, 2020). Analysis of a single water quality sample provides a snapshot of the source water quality without context of the microbial safety of a single source. Microbes are not evenly distributed throughout a water source; thus, repeated 100 ml samples tested from the same source at the same time yield different results (Kelly *et al.*, 2020). In the literature, authors report mixed results with regard to correlation between risk assessment score and microbial water quality data (Kelly *et al.*, 2020). For individual water supplies, no clear correlation was observed between water quality data and the sanitary condition of sources in Ethiopia (Alemayehu *et al.*, 2020).

Risk-based drinking water monitoring directs towards the most important, relevant parameters for system performance and public health protection (WHO 2019). Risk assessment drives the purpose and focus of water quality monitoring and control measures to mitigate contamination risks effectively at all times (WHO 2019). By identifying the main hazards and hazardous events (and their health risks) in the catchment and throughout the supply chain up to the point of consumption the outcomes of risk assessment could improve water quality monitoring (WHO 2019).

Water quality monitoring can be improved through:

- surveillance by identifying the most significant water quality parameters to protect health;
- compliance monitoring by identifying sample locations, including in high-risk areas of the distribution system and in buildings that host vulnerable members of the population;
- compliance and operational monitoring by informing sampling frequencies for high-risk parameters, taking into account seasonal or climatic variations that may lead to changes in the water quality; and
- operational monitoring by pointing out the most appropriate strategy to ensure that the barriers (treatments) to the high-risk parameters are working properly at all times.

Additionally, water quality monitoring can provide information for risk assessment. The different types of water quality monitoring are able to effectively support risk assessment methods to sustainably supply safe (drinking) water and safe sanitation. Data from source water quality monitoring can identify existing hazards or provide an early warning of source water contamination (Schilderman *et al.*, 1999; Lodder *et al.*, 2010; WHO 2016). The WSP manual provides examples of how (online) measurements support risk management and describes that operational monitoring results should be used on an ongoing basis to validate control measures (WHO 2023). These data can also be used for trend analyses or support setting appropriate and effective critical limits (WHO 2023). Validating control measures under different scenarios is particularly relevant when considering climate risks, especially if an existing control measure has been historically validated for less challenging conditions (WHO 2023).

Aim of the thesis

This thesis will focus on risk management for drinking water supply and sanitation systems to prevent waterborne infectious diseases in different countries and resource settings. Risk management here includes risk assessment and water quality monitoring. The main objective of this thesis is to better integrate water quality monitoring and risk assessment into risk management approaches. Another objective is to integrate risk management approaches for drinking water supply and/or sanitation services in order to reduce waterborne infectious diseases. The final objective is to investigate to which extent risk management methods create resilience to future changes, such as climate change and urbanization.

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Chapter 2

Effect of operational strategies on microbial water quality in small scale intermittent water supply systems: The case of Moamba, Mozambique

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Abstract

Intermittent drinking water supply affects the health of over 300 million people globally. In Mozambique, it is largely practiced in cities and small towns. This results in frequent microbial contamination of the supplied drinking water posing a health risk to consumers. In Moamba, a small town in Southern Mozambique with 2,500 water connections, the impact of changes in operational strategies, namely increased chlorine dosage, increased supply duration and first-flush, on the microbial water quality was studied to determine best practices. To that aim, water quality monitoring was enhanced to provide sufficient data on the microbial contamination from 452 samples under the different strategies. The water at the outlet of the water treatment plant during all strategies was free of *E. coli* complying to the national standards. However, *E. coli* could be detected at household level. By increasing the chlorine dosage, the number of samples that showed *E. coli* absence increased at the two sampling locations in the distribution network: in Cimento from 72% to 83% and in Matadouro from 52% to 86%. Modifying the number and duration of supply cycles showed a different impact on the water quality at both locations in the distribution network. A positive effect was shown in Cimento, where the mean concentrations decreased slightly from 0.54 to 0.23 colony forming units (CFU)/100 ml and 16.7 to 7.3 CFU/100 ml for *E. coli* and total coliforms respectively. The percentage of samples positive for bacteria was, however, similar. In contrast, a negative effect was shown in Matadouro where the percentage of positive samples increased and the mean bacterial concentrations increased slightly: *E. coli* from 0.9 to 1.5 CFU/100 ml and total coliforms 17.6 to 23.0 CFU/100 ml. Enhanced water quality monitoring improved operational strategies safeguarding the microbial water quality. The *E. coli* contamination of the drinking water at household level could point at recontamination in the distribution or unsafe hygienic practices at household level. Presence of fecal contamination at household level indicates potential presence of pathogens posing a health risk to consumers. Increasing chlorine dosage ensured good microbiological drinking water quality but changing the number of supply cycles had no such effect.

Introduction

Safe drinking water is acknowledged as a basic human right (UN 2010) and the Sustainable Development Goal (SDG) 6, target 6.1, aims to achieve "a universal and equitable access to safe and affordable drinking water for all by 2030" (UN 2016). It is widely known that drinking unsafe water may cause exposure to pathogens, which can result in waterborne diseases, such as cholera, gastroenteritis or hepatitis E (Howard and Bartram, 2003). However, inadequate water, sanitation and hygiene still caused 829,000 diarrhoeal deaths worldwide in 2016, which corresponds to about 60% of total diarrhoeal-related mortality rates (Prüss-Ustün *et al.*, 2019). Progress on SDG 6 is monitored using indicator 6.1.1, which is the percentage of population using "safely managed" water supplies, i.e. whether water sources are improved, accessible on premises, available when needed (for more than 12 h per day), and free from microbial contamination. According to the WHO/UNICEF Joint Monitoring Programme (JMP), 29% of the world population does not have access to safely managed drinking water (WHO and UNICEF, 2017). In Mozambique, diarrheal diseases play an important role in deaths and disability, and are strongly associated with precipitation (Horn *et al.*, 2018). Several studies describe the prevalence of infections with waterborne pathogens, such as *Vibrio cholerae, Cryptosporidium* and rotavirus in Mozambique (Sem´a Baltazar *et al.*, 2017; Casmo *et al.*, 2018; Deus *et al.*, 2018).

Over 300 million people globally rely on intermittent water supply (IWS), piped water delivered for less than 24 h per day (Kumpel and Nelson, 2016). Numerous countries in Africa, Asia and Latin America practice IWS as a normal operational strategy because water supply companies are not able to supply water continuously and sustain a positive operating pressure within the distribution network. This is also due to high levels of leakage in distribution networks (Klingel, 2012; Agathokleous and Christodoulou, 2016; Galaitsi *et al.*, 2016). Various studies noted that IWS is multi-faceted and co-produced by lack of water resources, infrastructure deficits and the ever increasing non-revenue water (Galaitsi *et al.*, 2016; Kumpel and Nelson, 2016). IWS can lead to the risk of waterborne diseases due to microbial contamination through ingress of pathogens in non- or low pressurized pipes through cracks or fittings, release of microbial biofilms formed under stagnant conditions during re-pressurization, recontamination during household storage, use of unsafe alternative water sources, or limited water availability for hygiene practices (Coelho *et al.*, 2003; Kumpel and Nelson, 2016). Cases of waterborne illnesses due to IWS continue to be documented and Bivins *et al.* (2017) suggested that, globally, IWS may account for 17.2 million infections causing 4.5 million cases of diarrhoea and 1,560 deaths each year. Which feature of IWS increases the growth of opportunistic pathogens still needs to be investigated (Bautista-de los Santos *et al.*, 2019). When the drinking water supply is turned on after a period without supply, drinking water may contain elevated turbidity, and high concentrations of indicator bacteria can be flushed out of the pipes (Kumpel and Nelson, 2014). Pathogens may also enter the drinking water and upon consumption may cause infections (Skraber *et al.*, 2005).

Despite the high prevalence of IWS in the world, the literature published to date on water quality in IWS systems is limited to a few studies in large urban areas (Kumpel and Nelson, 2016), refugee camps (Alazzeh *et al.*, 2019) and one small town in Central America (Erickson *et al.*, 2017). In particular, small towns in sub-Saharan Africa are experiencing an increase in water demand due to population growth, while the development and appropriate management of water infrastructure and services is lagging behind (Matsinhe *et al.*, 2008). This may lead to water shortage resulting in an increase in IWS in these towns. These towns are not only heterogeneous among themselves, but are diverse within the administrative boundaries as they often have both urban and rural areas, has implications for infrastructure planning and resource allocation (Marks *et al.*, 2020). In Mozambique, water supply is intermittent due to old transport and distribution networks, high levels of leakage, limited hydraulic capacity and increased city demand and population growth. As of 2015, small towns represent 15% of the total Mozambican population, and this share is projected to increase to 18% (about 6.5 million people) by 2030 (World Bank 2018). The majority of the cities in Mozambique experience intermittent supply with variable water supply duration (Gumbo *et al.*, 2003). Therefore, the aim of this study was to evaluate how different operational strategies at full scale can improve drinking water quality in an IWS system in a small town of Mozambique. We studied the impact of increased disinfectant dosage, increased supply duration and first-flush. To the authors' knowledge this is the first study to investigate the effect of operational strategies on drinking water quality in small scale IWS systems in sub-Saharan Africa. The results will be of interest for practitioners and researchers that focus on small water systems, particularly in low-resources settings.

Material and methods

Study area

Moamba district is located in Mozambique, in the southern part of the Maputo province and has an area of 4,628 km^2. The district consists of four towns and has a population of 83,876 inhabitants (Instituto Nacional de Estatistica 2018). Vila de Moamba, one of the four towns of the province, has a population of 24,650 inhabitants and 83% of the population is supplied with piped drinking water.

The water treatment plant (WTP) of Moamba has a capacity of 3,000 m^3/day. The source for the production of drinking water is the Incom´ati river; water is abstracted 3.5 km from the WTP. After infiltration the water is pumped into a buffer tank (80 m^3), which is connected to the WTP with a pipeline. At the WTP, the river water is subjected to:

- coagulation-flocculation based on dosing of aluminium sulphate;
- rapid sand filtration by six pressure filters with a capacity of 40 m^3/h each; and
- disinfection by dosing chlorine solution with a calculated dose of 1.8 mg Cl_2/L.

The WTP is operational in two shifts: from 6:00–12:00 (morning cycle) and 15:00–19:00 (afternoon cycle). The disinfected water is stored in a 500 m^3 reservoir and 150 m^3 water tower before distribution into the network. The water supply system of the WTP covers the areas of the District of Moamba and the Administrative Post of Pessene (14 km from Moamba). The distribution network has a total length of 45 km with approximately 3,336 connections. The distribution network is made of class 9 PVC with diameters ranging from 50 mm to 250 mm. The treated water is intermittently supplied to Moamba from approximately 6:00–10:00 (morning cycle) and 15:00–18:00 (afternoon cycle), whereas Pessene receives drinking water from 10:00–15:00 and 18:00–19:00.

Experimental design

Chlorine dosing

To assess the effect of chlorine dosing on drinking water quality, different dosages of granular high test hypochlorite ($Ca(OCl)_2$) with 65% of active chlorine were applied. A chlorine solution was prepared by diluting $Ca(OCl)_2$ in a 200 L tank and then dosed via an injector chlorinator for 48 h. The tank was fitted with a stirrer and a positive displacement diaphragm dosing pump (Grundfos DMX 14-10, Denmark). The chlorine solution was added to the filtered water to achieve a calculated dosage of 1.8 and 2.2 mg Cl_2/L, respectively. The dosing rate of the injector chlorinator was kept constant throughout the experiments. Samples were taken every hour during supply. All experiments were performed in duplicate. During the different dosing experiments, the concentration of the chlorine dosing suspension was adjusted to achieve the desired chlorine dosage in the different experiments.

Daily supply cycles

During standard operations of the WTP, water is supplied to Moamba for approximately 7 to 9 h in two daily cycles. In between those two cycles the WTP continues operating and water is supplied to the village of Pessene located about 14 km from Moamba. To investigate an effect of supply duration, water was supplied continuously for 10 h and 12 h (one cycle) to Moamba only and compared with normal operation (two cycles).

First flush

To assess the water quality during restart of the drinking water supply after an idle time of not supplying (first flush), samples were taken every 10 minutes during at least 50 minutes at two locations in the distribution network. The first flush was studied during standard operations with two supply cycles per day, resulting in a first flush in the morning and one in the afternoon. The effect of the first flush was examined by pairwise comparison of the results of t = 0 and t = 10 min.

Selection of sampling points

Three sampling points were selected: one at the outlet of the WTP and two household yard taps in different neighborhoods, namely Bairro Cimento and Bairro Matadouro. The two neighborhoods were selected based on the distance from the WTP (800 and 2,200 m, respectively) and spatial patterns of the neighborhoods (Bairro Matadouro is a densely populated neighborhood with lack of formal spatial planning whereas Bairro Cimento is a less dense and planned neighborhood). The sampling points are shown in Figure 2.1.

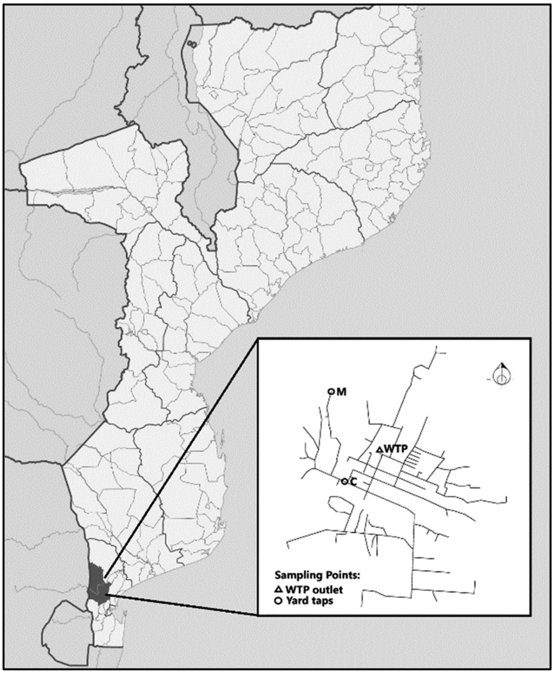

Figure 2.1. *Map of Mozambique with the distribution network of Moamba and location of the WTP and sampling points in Cimento (C) and Matadouro (M).*

Sampling

The experiments were conducted between November 2017 and October 2018. During these experiments, samples were taken every 10 minutes for the first hour after starting the supply cycle and then hourly from the following locations: outlet WTP, Cimento household yard tap (C) and Matadouro household yard tap (M). The tap at the sampling points was cleaned with a clean tissue soaked with ethanol 70% and flamed before samples were taken. Samples for microbiological analyses were collected in 100 ml sterile whirl-pak thiobags® containing sodium thiosulfate for neutralizing the residual chlorine and directly put in a cooling box for transport. The samples were stored at most 24 h prior to microbiological analyses. Samples for physicochemical analyses were collected in 75 ml plastic cups and directly analyzed in the field. In total, 717 samples were collected in this study (Table 2.1).

Table 2.1. *Number of samples taken per experiment.*

Supply duration	**Calculated chlorine dosing concentration ($mg\ Cl_2/L$)**		**Total number of samples**
	1.8	**2.2**	
Standard operations	275	230	505
10 hours	56	54	110
12 hours	64	38	102
Total number of samples	*395*	*322*	*717*

Methods

Physicochemical analyses

Water temperature and pH (PT115 pH meter, Palintest, United Kingdom), free and total chlorine (PTH7100, Palintest, United Kingdom), conductivity (PT157, Palintest, United Kingdom), and turbidity (PTH092, Palintest, United Kingdom) were measured on site for 655 samples.

Microbiological analyses

In 452 samples enumeration of total coliforms and *Escherichia coli* was carried out. This method was based on ISO 9308-1 (ISO 2014), using the membrane filtration method and incubation on chromocult agar nutrient pad sets (Sartorius Stedim Biotech, Germany) for 24 h at 37 °C in a portable incubator (Aquagenx, United States), according to the manufacturer's instructions. For each sample, 100 ml was tested in duplicate. All dark blue to violet colonies were counted as colony forming units (CFU) and provided the presumptive amount of *E. coli* in the filtered water volume. Salmon red colonies were coliform bacteria colonies other than *E. coli* as indicated in the suppliers' documentation. All dark blue to violet and salmon red colonies were counted as total coliforms.

Statistical analyses

Concentrations of *E. coli* and total coliforms, free chlorine concentrations and turbidity were logarithmically (base 10) transformed. Time of sampling was registered in minutes from starting drinking water supply. Water supply was from 7:00 and lasted 9, 10 or 11 h. Or, water supply was stopped after 5 h, and started again 4 h later for a duration of 4 h. Multivariate linear regression analyses of the relationship of the concentrations of *E. coli* and total coliforms respectively with the total distance from the WTP (m), time of sampling, free chlorine concentration, water temperature (°C), pH, turbidity (NTU) and conductivity (µSiemens/cm) were conducted using R (version 3.5.2 (2018-12-20) - "Eggshell Igloo") and lm (Chambers, 1992; Wilkinson and Rogers, 1973). The model with the lowest Akaike information criterion was selected using the step-function (parameter k = 3.84). For graphical presentation of the data package ggplot2 was used (Wickham, 2016). Relations for *E. coli* and total coliforms were analyzed separately, but also for the joint bacteria concentration, whereby the factor bacteria with values "*E. coli*" and "Total coliforms" was included. Similarly, a relation between both bacteria groups was analyzed, as well as effects of the environmental factors, such as water temperature, pH and conductivity, on free chlorine concentration and turbidity.

Results

Effect of increased chlorine dosing

Under standard operations (two supply cycles per day), increasing the calculated chlorine dosage from 1.8 to 2.2 mg Cl_2/L resulted in an increase of the mean concentration of chlorine at the outlet of the WTP by 30% from 0.79 mg Cl_2/L to 1.03 mg Cl_2/L, in Cimento by 77% from 0.52 mg Cl_2/L to 0.92 mg Cl_2/L, and in Matadouro by 75% from 0.36 mg Cl_2/L to 0.63 mg Cl_2/L (Figure 2.2). The concentration of free chlorine observed at the same sampling points using a higher chlorine dosing concentration complied with the national Mozambican standard of 0.2–0.5 mg Cl_2/L (MISAU 2004) and the number of compliant samples at the WTP outlet increased from 90% to 100%, in Cimento from 81% to 100% and in Matadouro from 70% to 82%. The bacterial load at the outlet of the WTP showed absence of *E. coli* in all samples, and the mean concentration of coliforms was 6.5 CFU/100 ml. The concentrations of *E. coli* and total coliforms increased from the outlet of the WTP, through Cimento to Matadouro, but the difference in mean concentration with the two chlorine dosages was minimal (Figure 2.2). The main difference is shown by the number of samples that showed *E. coli* absence: in Cimento it increased from 72% to 83% and in Matadouro it increased from 52% to 86% (see Supplementary Table 1). The increased chlorine dosing had an effect on the compliance with the national Mozambican standard of 0.2–0.5 mg Cl_2/L (MISAU 2004). The number of non-compliant samples containing <0.2 mg/L free chlorine at the WTP outlet decreased from 10% to 0%, in Cimento from 19% to 0% and in Matadouro from 30% to 18%. However, by increasing the chlorine dosing the number of non-compliant samples containing >0.5 mg/L free chlorine increased at the WTP from 68% to 100%, in Cimento from 45% to 75% and in Matadouro from 22% to 49%.

The free chlorine concentration under standard operations is highly significantly dependent on chlorine dose, distance from WTP and temperature (Supplementary Table 2a). The residual concentration of free chlorine increased with dose (1.8–2.2 mg Cl_2/L) and pH (7.2–9.2), and decreased with distance (0–2,200 m) and water temperature (4.2–37 °C).

Increased *E. coli* and total coliforms concentrations at higher distances from the WTP were observed (Figure 2.2), this effect was statistically insignificant, probably because *E. coli* concentrations were low and the majority of data (77%, n = 232) consisted of non-detects. For more statistical power, *E. coli* and total coliform concentrations were jointly statistically analyzed with bacteria as a factor. In this combined analysis, none of the conditions were found to have a significant effect (see Supplementary Table 2b). The turbidity, conductivity, pH and water temperature under standard operations are shown in Supplementary Table 3.

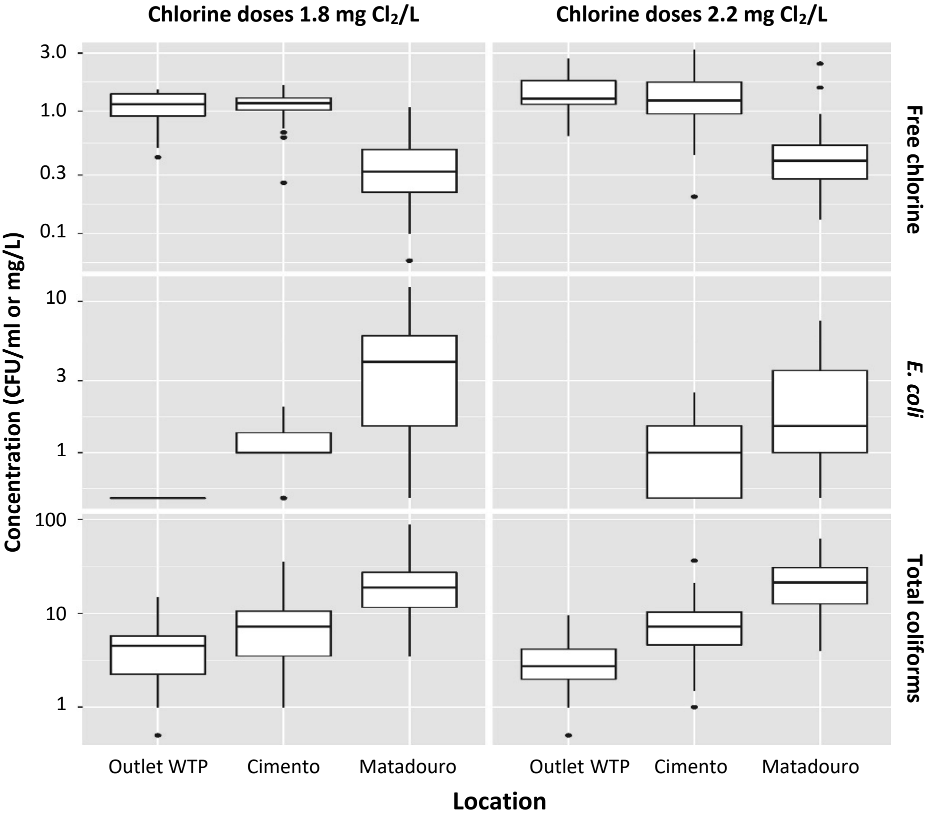

Figure 2.2. *Box-Whiskerplots of free chlorine, E. coli and total coliforms concentrations according to location. Each grid represents the concentration of the free residual chlorine, E. coli or total coliforms achieved with chlorine doses of 1.8 and 2.2 mg Cl_2/L. The box represents the median and quartiles, the whiskers show the 95%-interval and dots are outliers.*

Effect of varying daily supply cycles

Similar results were obtained for the different levels of chlorine when varying the number of daily supply cycles and the overall supply duration with a decrease in the concentration of residual chlorine over the distance from the WTP and higher concentrations of residual chlorine by using higher dosing concentrations. For supply during one or two daily cycles, the percentage of samples positive for microbial contamination with a higher mean concentration of total coliforms and *E. coli* in Matadouro (most distant point from the WTP) than in Cimento (Table 2.2). Specifically, the number of samples positive for *E. coli* increased with distance from 22% in Cimento to 30% in Matadouro for two daily supply cycles and from 20% in Cimento to 42% in Matadouro for one cycle. Comparing standard operation and modified operation, no clear differences could be identified for bacterial or physico-chemical contamination. In Cimento, the percentage of samples positive for *E. coli* and total coliforms was similar while supplying one or two cycles, whereas the mean concentrations decreased with one cycle: *E. coli* decreased from 0.54 to 0.23 CFU/100 ml and total coliforms from 16.7 to 7.3 CFU/100 ml. At Matadouro the percentage of positive samples and mean concentrations of *E. coli* and total coliforms slightly increased when one supply cycle was applied. The percentage of positive samples increased from 29% to 42% for *E. coli* and from 92% to 100% for total coliforms, respectively. The mean concentration for *E. coli* and total coliforms increased from 0.9 to 1.5 CFU/100 ml and from 17.6 to 23.0 CFU/100 ml, respectively. These results show that the effect of modifying the operations can differ by location in the same distribution network.

Table 2.2. *E. coli and total coliforms mean concentrations for different water supply durations.*

Parameter		**Standard operation Water supply during 11 hours (2 cycles)**		**Modified operation Water supply during 10 and 12 hours (1 cycle)**	
		Cimento	**Matadouro**	**Cimento**	**Matadouro**
E. coli	Number of samples	105	103	76	78
	Mean concentration CFU/100 ml (min – max)	0.54 (0 – 11)	0.9 (0 – 15.5)	0.23 (0 – 2.5)	1.5 (0 – 12.5)
	Number of samples with > 1 CFU/100 ml (%)	23 (22%)	31 (30%)	15 (20%)	32 (42%)
Total coliforms	Number of samples	105	103	76	78
	Mean concentration CFU/100 ml (min – max)	16.7 (0 – 100)	17.6 (0 – 75)	7.3 (0 – 36.5)	23.0 (0 – 89)
	Number of samples with > 1 CFU/100 ml (%)	84 (79%)	95 (92%)	62 (80%)	78 (100%)

An increase in the median and average residual concentration of free chlorine were observed at Cimento, but not at Matadouro, when changing the supply from two cycles to one cycle. Dosing experiments with 1.8 mg Cl_2/L showed a median concentration of 0.44 mg Cl_2/L and an average concentration of 0.52 mg Cl_2/L using 2 cycles, while supplying with one cycle median and average concentrations were 1.16 and 1.13 mg Cl_2/L, respectively. In Matadouro the mean and average concentration were similar, 0.32 and 0.39 mg Cl_2/L for one supply cycle versus 0.24 and 0.36 mg Cl_2/L for two supply cycles. Similar results were obtained with dosing experiments of 2.2 mg Cl_2/L. No significant change was observed for different supply durations in *E. coli* and total coliform concentrations. The bacterial concentration on $\log_{10}$ scale was highly significantly dependent on the distance, time and conductivity, see Supplementary Table 2c. Bacterial concentrations increased with distance, but decreased with increasing time and pH. Free chlorine, temperature and conductivity did not play a role according to this model. The turbidity, conductivity, pH and water temperature under modified operations are shown in Supplementary Table 3.

Effect of first flush

In order to ascertain the effect of first flush, samples were collected every 10 minutes after re-starting the water supply to Moamba for the first 50 minutes, both in the morning and afternoon cycles. Figure 2.3 shows the results of the concentration *E. coli* and total coliforms measured in the neighborhood of Cimento (closer to the WTP) and Matadouro (further away from the WTP). The concentration of *E. coli* and total coliforms did not show a considerable increase at the beginning of the supply cycle, during the first 50 minutes. The mean concentration of total coliforms fluctuates at both locations in the morning and afternoon cycle. The mean concentration for *E. coli* in Matadouro slightly increased: in the morning cycle from 0.9 CFU/100 ml at $t = 0$ to 1.8 CFU/100 ml at $t > 50$ minutes and in the afternoon cycle 0.1 CFU/100 ml at $t = 0$ to 1.8 CFU/100 ml at $t > 50$ minutes. No clear correlation was found comparing every pair of measurements at 10 min intervals up to 50 minutes. The clearest first flush effect was expected directly after re-starting the water supply, especially by comparing $t = 0$ and $t = 10$ min.

Comparing the bacterial results at $t = 0$ and $t = 10$ min pairwise, showed that the concentration of coliform bacteria in Matadouro varied between a decrease of 0.47 CFU/100 ml to an increase of 0.26 CFU/100 ml, and in Cimento between a decrease of 0.20 CFU/100 ml and an increase of 0.30 CFU/100 ml. The bacterial concentration was dependent on the free chlorine concentration, but a statistical effect of time ($t = 0$ versus $t = 10$ min) was not found. Turbidity and residual concentration of free chlorine did not show a clear increase or decrease at either of the locations. The deviation of the residual concentration of free chlorine between $t = 0$ and $t = 10$ min varied between -0.04 and 0.47

mg Cl_2/L for Cimento and -0.17 and 0.43 mg Cl_2/L for Matadouro. In 61% and 52% of the samples taken from Cimento and Matadouro respectively, the deviation was less than 0.1 mg Cl_2/L. The deviation in turbidity between t = 0 and t = 10 min varied between -9.4 and 1.8 NTU for Cimento and -7.0 and 2.6 NTU for Matadouro. Turbidity fluctuated between 2.2 and 27.1 NTU in Cimento and 1.2–23.5 NTU in Matadouro during all first flush experiments from t = 0 to t = 50 min, but also in this case no clear increasing or decreasing trend was identified.

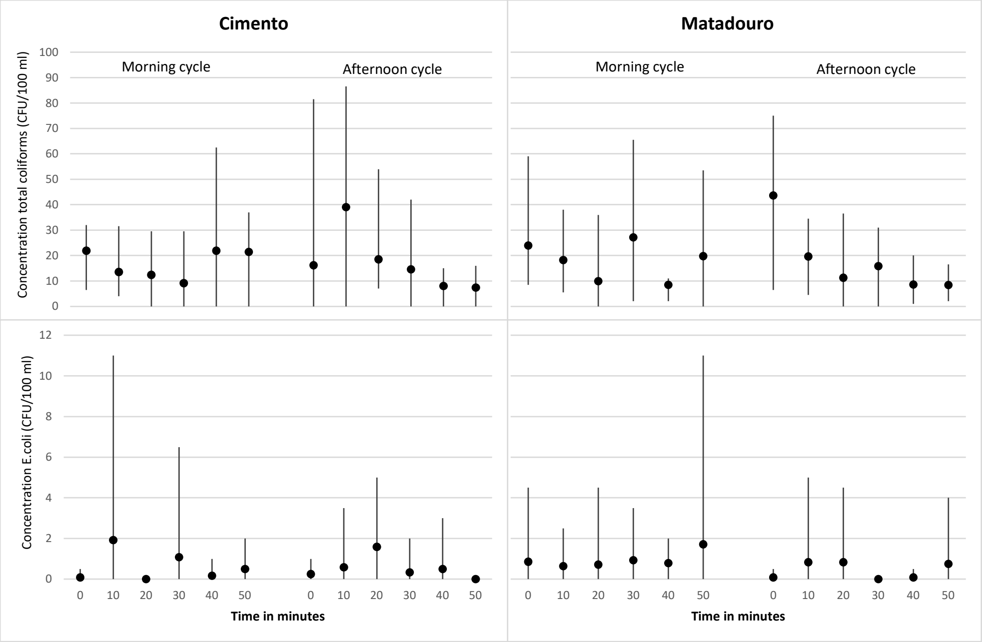

Figure 2.3. *Bacteriological results of the first flush after starting the distribution of drinking water during the morning and afternoon cycle. Total coliform and E. coli concentrations for Cimento and Matadouro are presented as a function of the time since the beginning of the supply cycle.*

Discussion

The aim of this study was to understand the effect of increased disinfectant dosage, number and duration of supply cycles, and first-flush on drinking water quality in an IWS system in a small town in Mozambique. When considering the indicator of fecal contamination, *E. coli*, no contamination was detected in treated water leaving the WTP, nevertheless *E. coli* was detected at the point of delivery at household level. Recontamination of the treated drinking water in the distribution could have occurred from ingress in the pipes. If fecal contamination entered the distribution system, some pathogens could have persisted even though fecal indicators were inactivated, which poses a health risk (LeChevallier *et al.*, 2004). Similar results were obtained in large urban centres in, among others, Pakistan, India and Uganda where the water distribution systems were not capable of maintaining high water quality from the water treatment facilities to the end-user (Hashmi *et al.*, 2008; Matsinhe *et al.*, 2014). Based on literature, Bivins *et al.* (2017) showed that the available evidence suggests large variability in the prevalence of fecal contamination in IWS networks with the proportion of samples positive for *E. coli* ranging from 2% to 32%. In our study the prevalence of *E. coli* was 28% at the sampling point closer to the WTP, but as high as 48% at the furthest sampling point. For research purposes, we recommend detection of waterborne pathogens in the distribution network, such as adenovirus, rotavirus, *Cryptosporidium* and *Vibrio cholerae*, which cause infections in Mozambique (Liu *et al.*, 2016; Sem´a Baltazar *et al.*, 2017; Casmo *et al.*, 2018; Deus *et al.*, 2018). This information supports the need or improvement of control measures, such as chlorination, and the health risk to consumers. An increased chlorine dose of 2.2 mg Cl_2/L improved the residual chlorine level in the distribution network by a minimum of 0.2 mg Cl_2/L, thereby complying with international guidelines (WHO 2017b) and national standards (MISAU 2004). The residual concentration of free chlorine decreased with the distance, which is similar to other studies (Egbe and Bassey, 2016; Karikari and Ampofo, 2013; Sakomoto *et al.*, 2020). The percentage of samples with levels of residual chlorine lower than 0.2 mg Cl_2/L, 19% in Cimento and 30% in Matadouro, was much lower compared to the percentage of samples with *E. coli*, 28% in Cimento and 48% in Matadouro, and total coliforms, 81% in Cimento and 89% in Matadouro. Similar results were obtained in other studies where drinking water samples contained coliforms or *E. coli* even though the concentration of residual free chlorine was above 0.2 mg Cl_2/L (Erickson *et al.*, 2017; Sakomoto *et al.*, 2020). Although the number of samples complying with the national Mozambican standard of >0.2 mg Cl_2/L for residual chlorine increased by increasing the chlorine dose, the number of samples with concentration >0.5 mg Cl_2/L and therefore not complying with the national standards (MISAU 2004) also increased. In this study, the percentage of samples at the WTP outlet with a residual concentration of free chlorine higher than 0.5 mg Cl_2/L increased from 68% to 100%, by increasing the chlorine dose from 1.8 mg Cl_2/L to 2.2 mg Cl_2/L. Of all samples from yard taps containing bacteria, 56% contained coliform bacteria even though the residual concentration of free chlorine was higher than 0.5 mg Cl_2/L. Analogously, in water samples from a WTP outlet to the tap in Ethiopia, coliforms could be detected even though containing 0.5 mg Cl_2/L free chlorine (Duressa *et al.*, 2019). By increasing chlorine dosage, the number of samples positive for *E. coli* in the distribution network decreased (see Supplementary Table 1), in line with other studies in which a weak inverse correlation was observed between free chlorine levels and fecal coliforms (Karikari and Ampofo, 2013). If the range of residual chlorine at the WTP outlet is between 0.2 and 0.5 mg Cl_2/L the concentrations at the tap very distant from the WTP may be less <0.2 mg Cl_2/L. To ensure higher levels of free chlorine further in the distribution network, booster chlorination might be an option as suggested in a study in Uganda (Sakomoto *et al.*, 2020). However, when fecal contamination of a drinking water supply is detected, the World Health Organization recommends that the concentration of free chlorine should be increased to greater than 0.5 mg Cl_2/L throughout the system as a minimum immediate response (WHO 2017b). As *E. coli* concentrations were low and the majority of data (77%, n = 232) consisted of non-detects. For more statistical power, *E. coli* and total coliform concentrations were jointly statistically analyzed with bacteria as a factor. However, no clear inverse relation was shown between increasing chlorine dosing and levels of bacteria.

In the case of Moamba, water is supplied in multiple daily cycles (Silva-Novoa Sanchez *et al.*, 2019). In another study on IWS with multiple daily cycles in rural Nepal, consumers' perception of the level of service in terms of water quality worsens as the duration of supply decreases (Guragai *et al.*, 2017). However, there is no evidence that the duration and number of supply cycles correlate to the water quality. In this study we increased the supply duration to up to 12 h per day, the minimum threshold used by the WHO/UNICEF JMP to track the 'available when needed' factor of target 6.1 of SDG 6. However, no association between increased availability and lower number of daily cycles (one as opposed to two) and microbial water quality was observed. In fact, the effect of modifying the operations in Moamba differed per location within the same distribution network: the bacterial concentration decreased close to the WTP outlet, and increased further in the distribution network. The residual concentrations of free chlorine at the tap closer to the WTP outlet were higher supplying one cycle compared to two cycles, but further in the distribution network the concentrations were similar. In general, microbial growth and public health implications depend on the duration of the stagnation periods, the composition of the microbial community, and disinfectants in IWS (Bautista-de los Santos *et al.*, 2019). Microbial growth due to overnight stagnation has also been reported in continuous water supply (Lautenschlager *et al.*, 2010; Lipphaus *et al.*, 2014). However, our findings have not yet been followed up by further studies to investigate the causes of differentiated water quality outcomes at these specific locations. Research on the composition of the microbial community as described by Bautista-de los Santos *et al.* (2019) or microbial source tracking can clarify these differences or to identify possible contamination sources (Liu *et al.*, 2018). In this study, the effect of first flush on the microbiological water quality is not significant, although the bacterial concentrations are slightly higher at t = 0 min and t = 10 min, after starting the operation, compared with other time points. This is similar to the findings of Alabdula’aly and Khan (2017), who showed that stagnation in the distribution network affects the water quality, but not to a degree that would warrant collective actions. In contrast to our findings, other studies showed an effect of first-flush on the drinking water quality. Kumpel and Nelson (2013) showed more contamination during the first flush after the supply re-started and during periods of low pressure. In another study, the water quality was degraded during some first-flush events and after pipe breaks and repairs (Erickson *et al.*, 2017). In the same study, higher concentrations of heterotrophic plate count and spore-forming bacteria were found during many first flush events, even when total coliform and *E. coli* were not detected (Erickson *et al.*, 2017). Stagnation of water in the piping system caused by pressure deficits and intermittent feeding of the system entails that pathogens may enter and grow in the water distribution network (Jensen *et al.*, 2002; Lee and Schwab, 2005; Andey and Kelkar, 2007). This hazard increases at high temperatures by running pipes close to the surface (Klingel, 2012). In this study, the water stagnated at most 14 h, and no significant difference was found between first flush events that occurred after different stagnation times. Future research is needed to better understand the importance of the effect of first-flush on pathogens.

In addition to these risks inherent to IWS, distribution systems in low and middle-income countries often have additional vulnerabilities which may degrade the water quality. Some examples are frequent pipe breaks (Lee and Schwab, 2005), poor quality control of treated water entering the distribution network (Besner *et al.*, 2002; Lee and Schwab, 2005), and unhygienic repair practices (Besner *et al.*, 2002). In general, for coping with the contamination ingress due to backflow through leaky joints, air valves, perforations in IWS, the WHO (2017b) recommends implementing the following control measures, where feasible: maintain positive pressure, provide continuous supply; maintain minimum chlorine residuals in the distribution network and, if necessary, install secondary/booster chlorination; implement a leak detection and repair programme; implement a pipe and fittings replacement programme; and develop design and construction specifications and standards. Climate change affects safe drinking water supply as it is expected to alter the frequency and severity of extreme weather events (WHO 2017a). As large areas of the country are exposed to cyclones, droughts and flooding, Mozambique is vulnerable for climate change (Arndt *et al.*, 2011). Assuming climate change alters precipitation patterns and subsequently the number of wet days diarrheal cases might increase in Mozambique (Horn *et al.*, 2018). Therefore adaptation of the drinking water supply to climate change is required (WHO 2017a). Implementation of a systematic risk assessment and risk management approach, such as climate-resilient water safety plans, might support better understanding of possible health risks and how these can be managed, including climate change aspects (WHO 2017a).

Study limitations

The results of this study are subject to a few limitations. First, experiments on the effect of supply duration at the full scale were impossible in Pessene in order not to alter the supply pattern, and water supply with even longer duration was not possible due to the existing work shifts of the utility operators. Second, only two chlorine dosages were included in this publication due to limited skills of the operator working in one of the shifts that arbitrarily decided to bypass the chlorine dosing tank and to add chlorine directly in the reservoir, making it impossible to control chlorine concentration. This episode highlighted once again the issue of limited technical capacities locally available in small towns (Tutusaus *et al.*, 2018). Finally, in this study, only negative controls were used for microbial analyses to exclude false positive results. No positive controls were used to exclude false negatives.

Conclusion

The main conclusions of this study are:

- Residual concentration of free chlorine increased with dose and pH, and decreased with distance and temperature.
- No fecal contamination was detected in treated water leaving the WTP, but was assumed to enter in the distribution system. The presence of fecal contamination is indicative of the potential presence of pathogens posing a health risk for consumers.
- Increased chlorine dosage can improve compliance with microbiological water quality standards.
- The presence of chlorine resistant pathogens can still pose a risk for human health.
- The mean concentration of *E. coli* in the two sampling points in the distribution network was nearly unchanged.
- Changing the number and duration of water supply cycles showed a positive impact on microbial water quality in the sampling point closest to the WTP and negative impact in the sampling point furthest from the WTP. Thus, modifying the operations can have different impacts on the different locations in the same distribution network.
- Contrary to published literature, the effect of first flush on the microbiological water quality was not statistically significant in this study.

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Supplementary data

Supplementary data can be found online at https://doi.org/10.1016/j.ijheh.2021.113794.

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Chapter 3

Occurrence of waterborne pathogens and antibiotic resistance in water supply systems in a small town in Mozambique

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Abstract

Microbiological quality of drinking water supplied in Moamba, a small town in southern Mozambique, was assessed by collecting and analyzing 91 water sample from five sampling sites: raw or inlet water, treated water and three household taps along the water distribution system. The presence of *Escherichia coli* as indicator fecal contamination, three bacterial pathogens, *Vibrio cholerae*, *Salmonella* spp. and *Campylobacter* spp., and cefotaxime resistant *E. coli* as antibiotic resistance determinant, was assessed.

The results showed fecal contamination in all types of water samples: *E. coli* was found in 100% of inlet water samples, in 21% of treated water samples, and in 22% of tap water samples. No *Salmonella* spp. was detected during the study. The presence of *V. cholerae* was detected in 42% of all water samples tested: 100% of inlet water samples, in 16% of treated water samples, and in 23% household tap water samples. All *V. cholerae* confirmed isolates where genotyped by PCR as non-O1/non-O139; however, nine isolates showed the presence of the genes encoding for cholera toxin. The presence of *Campylobacter* spp. was detected in 36% of the water samples tested: in 95% of inlet water samples, in 10% of treated water samples and in 23% household tap water samples. Cefotaxime resistant *E. coli* was detected in 63% of inlet water, 16% of treated water, and in 9% of tap water samples, these isolates were also resistant to multiple other antibiotics: ampicillin, streptomycin, tetracycline chloramphenicol. All 70 *V. cholerae* non-O1/non-O139 confirmed isolated were resistant to ampicillin, 51% to streptomycin, 13% to gentamycin, and one isolate was resistant to tetracycline; 13% showed a multidrug resistant profile, being resistant to at least three antibiotics.

The presence of fecal contamination and pathogens in the water treatment system and household taps in Moamba indicates a health risk for the population. This burden increases by the presence of bacterial pathogens showing multidrug resistance.

Introduction

Universal access to water, the source of life for every human being and for the survival of the planet, has been recognized as a universal human right (UN 2010). Several pathogens such as enteric bacteria, viruses and parasites, are transmitted through consumption or exposure to contaminated water and cause major diseases that represent a global public health problem, particularly for children under the age of five (WHO and UNICEF, 2019; Prüss-Ustün *et al.*, 2019). Despite growing efforts to ensure access to safe water, an estimated 20 million citizens will be exposed to contaminated water by 2030, and waterborne infections and epidemics continue to be a major global public health concern (Holcomb and Stewart, 2020). Contaminated water represents also a possible route of human exposure to antibiotic resistant pathogens of environmental origin (Iwu *et al.*, 2021). Antibiotic resistance is increasingly a global public health concern leading to millions of deaths due to drug resistant infection every year, with 700,000 deaths related to antimicrobial resistance are recorded annually (UN 2022). It is estimated that antimicrobial resistant infections may become the leading cause of death globally by 2050 with more than 10 million deaths per year (UN 2022).

Sub-Saharan Africa is the area most at risk among the Sustainable Development Goals (SDG) regions (WHO and UNICEF, 2019). Several countries in this region rely on intermittent water supply (IWS), which provide piped water to consumers for less than 24 h per day. The risk of waterborne diseases due to microbial contamination of water in IWS is often high due to the ingress of pathogens in non or low-pressurized pipes through intrusion, back flow, release of particulates, or sloughing of biofilms (Kumpel and Nelson, 2014; Bivins *et al.*, 2017). Furthermore, the reduced availability of piped water associated with IWS forces households to store water and/or the use alternative unsafe water sources, practices that increase the exposure to contaminated water (Klingel, 2012; Agathokleous and Christodoulou, 2016; Galaitsj *et al.*, 2016).

In order to reduce waterborne diseases, an adequate assessment of the presence of pathogens is essential to implement appropriate water treatment practices (Kumar *et al.*, 2003; Peletz *et al.*, 2016; Holcomb and Stewart, 2020). However, there are no universal methods of detection and identification of waterborne pathogens that are applicable to different socio-economic contexts, which makes it difficult to obtain comparable measures and formulate appropriate policies (Cotruvo, 2017; Bridle, 2020). Current standard methods for monitoring microbial water quality are based on the detection of fecal indicator bacteria (FIBs), such as *Escherichia coli* or *Enterococcus faecalis*, the presence of which indicates fecal contamination of water (LeChevallier and Au, 2004). However, an inconsistent relationships between FIB and enteric pathogens occurrence in drinking water have been reported in different settings worldwide (Figeras and Borrego, 2015). Consumption of water free from FIBs have been associated with diarrheal disease outbreaks, likely due to treatment processes that are unable to completely eliminate the pathogens (Saxena *et al*., 2015; Nhampossa *et al*., 2015). Moreover, detection of pathogens in water is not part of routine water quality monitoring, and is restricted to research studies or in case of suspected outbreaks (Nhampossa *et al*., 2015).

In Mozambique, information on waterborne diseases infections is relatively scarce but confirms that diarrheal diseases are a significant contributor to morbidity and mortality, especially among young children (8–10%) (Chissague *et al*., 2018; Deus *et al*., 2018; Raza *et al*., 2020). Enteric infections are predominatly caused by rotavirus (Sumbana *et al*., 2015), pathogenic *E. coli* (Mandomando *et al*., 2015; Mandomando *et al*., 2020; Sumbana *et al*., 2021; Manhique-Coutinho *et al*., 2022), *Salmonella* (García *et al*., 2018; Knee *et al*., 2018), *Campylobacter* (Ansaruzzaman *et al*., 2004) and *Vibrio cholerae* (Mandomando *et al*., 2007; Langa *et al*., 2015; Dengo-Baloi *et al*., 2017). The latter continue to represent a major public health burden as Mozambique continues on experiencing recurrent annual outbreaks of cholera in different parts of the country, caused by multidrug resistant (MDR) *V. cholerae*, with incidences ranging from 0 to 211 per 100,000 population and periodically high case-fatality ratios (Taviani *et al*., 2008; Semá Baltazar *et al*., 2017; Cambaza *et al*., 2019). However, little information is available on the contamination of raw and stored water, mostly limited to the detection of FIB, with few studies detected the presence of waterborne pathogens by molecular methods (Salamandane *et al*., 2021; Macario, 2022).

In Mozambique, water supply in small towns, where 15% of the country population resides (Gumbo *et al*., 2003), relies on IWS and it is characterized by high levels of leakage, limited hydraulic capacity and short water supply duration (< 12 h). Van den Berg *et al*. (2021) investigated the effects of operational strategies, such as increased disinfectant dosage, increased supply duration and first-flush, on drinking water quality in an IWS system in a small town of Mozambique. It demonstrated that water in distribution chain is fecally contaminated based on *E. coli* as indicator. The aim of this study was to deepen the work conducted by Van den Berg *et al*. (2021) by investigating the presence of waterborne pathogens and antibiotic resistant bacteria in an IWS system in a small town of Mozambique. Standard cultivation methods were coupled with molecular techniques for the detection of *E. coli* as FIB, Extended Spectrum Beta Lactamase *E. coli* (cefotaxime resistant *E. coli*) as indicator of antimicrobial resistant bacteria, and waterborne pathogens: *Vibrio cholerae*, *Salmonella* spp. and *Campylobacter* spp., and their antibiotic resistance profile. The results of this study are relevant for water operators, policy makers and researchers.

Methods

Study area

This study was conducted in Moamba, a town located in the southern Maputo province of Mozambique. Moamba District has an area of 4,628 km^2, and the town has a population of 24,650 inhabitants (Anuário Estatístico, 2018). Since 2013 the town is supplied by an IWS system with the capacity of 3,000 m^3/day. The source for the production of drinking water is the Incomáti river. Water is abstracted 3.5 km from the water treatment plant (WTP) and subjected to coagulation-flocculation based on dosing of aluminium sulphate, rapid sand filtration by six pressure filters with a capacity of 40 m^3/hour each, and disinfection by dosing chlorine solution with a calculated dose of 1.8 mg Cl_2/L (Van den Berg *et al.*, 2021).

The system supplies water to three neighborhoods of Moamba, reaching 83% of the population through a distribution network with a total length of 45 km with approximately 3,336 connections. The WTP is operates in two cycles (morning and afternoon) and disinfected water is stored in a 500 m^3 reservoir and 150 m^3 water tower before being distributed via the network (Van den Berg *et al.*, 2021).

Sampling locations

Samples were collected from the intake source water for drinking water production (inlet, I), treated water at WTP (outlet, T0), drinking water from three taps in different neighborhoods of Moamba: Cimento (T1), Matadouro (T2) and Barrio Sul (T3). Sampling locations had a piping distance from the WTP of 800 m, 2,200 m and 1,863 m for T1, T2 and T3, respectively (Figure 3.1). Nineteen sampling rounds were carried out on a monthly basis between March 2018 and October 2019 during both the dry (April 2018 - October 2018 and April 2019 - September 2019) and wet seasons (March 2018 and November 2018 - March 2019). A total of 91 water samples were collected at the 5 sampling sites. For sampling trips in March and April 2019 it was not possible to collect water from household T3, and in April 2019 it was not possible to collect water from households T2 and T3, because residents reported lack of water at these sites.

Water samples collection

Water samples were collected in the morning supply cycle, between 10 and 11 AM. For microbiological and physicochemical analyses, 500 ml water samples were aseptically collected in sterile collection bottles previously cleaned with detergent and sterilized by autoclaving for 20 min at 121 °C prior to use. Collection bottles were supplemented with thiosulfate tablets (Starplex Scientific Inc, USA) to inactivate available chlorine.

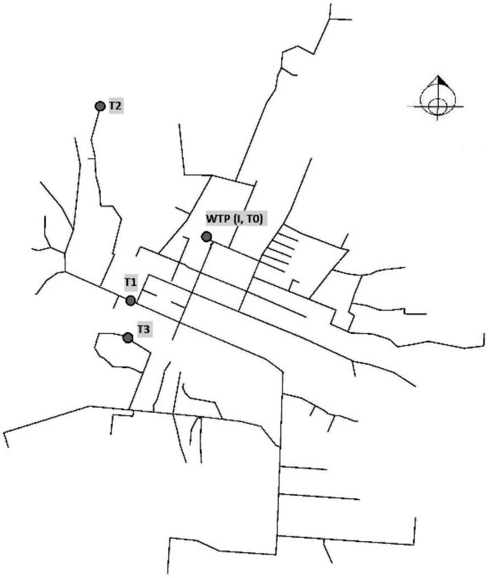

Figure 3.1. *Map of the distribution network of Moamba and location of the WTP (I, T0) and sampling points in Cimento (T1), Matadouro (T2) and Bairro Sul (T3).*

Water samples of inlet and outlet of the WTP and tap water were collected after flushing the water for two minutes and kept in a cooler during transport to the Center for Biotechnology laboratory for analysis. Samples were processed on the same day, typically within 2–4 h of collection.

Microbiological parameters

Enumeration of *E. coli* and cefotaxime resistant *E. coli* as well as detection of the pathogens *Vibrio cholerae*, *Salmonella* spp. and *Campylobacter* spp. was carried out in all water samples (I, T0, T1, T2, T3) by testing different volumes per parameters and per sample location (Supplementary Table 1).

Volumes ranging between 10 ml to 100 ml were filtered through a 0.47 µm polycarbonate membrane and incubated in plates or 50 ml of enrichment selective media. Volumes of 0.1 ml and 1 ml were either added to sterile PBS and filtered or added directly to 10 and 9 mL of specific enrichment selective media, respectively (Supplementary Table 1).

E. coli

Enumeration of total *E. coli* was done according to ISO 9308-1 standard method (ISO 2014). Different volumes (Supplementary Table 1) were filtered and membranes were placed on Tryptone Bile X-glucuronide (TBX) (Merck KGaA, USA) plates and incubated overnight at 35–37 °C. After incubation, plates were checked for growth and presumptive *E. coli* β-glucuronidase-positive (blue/green) colonies in each plate containing less than 300 were counted as colony forming units (CFU).

Cefotaxime resistant E. coli

Different volumes (Supplementary Table 1) were filtered and membranes were placed onto TBX plates supplemented with cefotaxime (CTX) (4 µg/ml) and incubated overnight at 35–37 °C. After incubation, plates were checked for growth and presumptive *E. coli* β-glucuronidase-positive (blue/green) colonies in each plate containing less than 300 were counted as CFU. At least five colonies were picked and stored in glycerol at -80 °C for species confirmation by PCR and antibiotic resistance profile.

Vibrio cholerae

V. cholerae was detected as described by Huq *et al*. (2012). After filtration, membranes were incubated in Alkaline Peptone Water (APW). The enrichment broth was incubated overnight at 35–37 °C, followed by plating onto Thiosulfate Citrate Bile salt Sucrose (TCBS) agar (BD, USA) and overnight incubation at 35–37 °C. Plates were checked for growth of presumptive *V. cholerae* (yellow, with a diameter of 2–3 mm) and at least five presumptive *V. cholerae* colonies were picked, plated onto Luria Bertani agar, incubated over night at 35–37 °C and then stored in glycerol at -80 °C for molecular analysis and antibiotic susceptibility.

Salmonella

Salmonella spp. detection was carried out according to the ISO 19250 standard method (ISO 2010). Membranes were transferred to Buffered Peptone Water for non-selective enrichment during overnight incubation at 35–37 °C. The following day 0.1 ml of enrichment was added to 10 ml of Rappaport-Vassiliadis soya peptone broth for selective enrichment for 24 h at 41.5 °C. Samples exhibiting growth were plated onto Xylose Lysine Deoxycholate (XLD) agar and incubated 24 h at 35–37 °C. Tubes not exhibiting growth were re-incubated for 24 h at 35–37 °C and then checked for exhibiting growth. XLD plates were checked for growth and presumptive *Salmonella* colonies were subcultured onto Brilliant Green (BG) agar plates and incubated for 24 h at 35–37 °C for further confirmation. At least five presumptive *Salmonella* (pink-red on BG) colonies were stored in glycerol at -80 °C for molecular analysis for confirmation and antibiotic resistance profile.

Campylobacter

For *Campylobacter* spp. detection Preston Broth was used for enrichment after filtration and incubated 48 h at 35–37 °C in microaerophilic conditions. Tubes exhibiting growth were plated onto Karmali agar and incubated 48 h at 35–37 °C in microaerophilic conditions. Karmali plates were checked for growth and five presumptive *Campylobacter* spp. colonies were stored in glycerol at -80 °C or molecular analysis.

Molecular analysis: species confirmation and typing

DNA was isolated from the cells by the boiling method. Isolates were retrieved from glycerol stocks by plating onto specific media (see above) and incubated over night at 37 °C. From each isolate, one colony was picked and added to 500 µL of sterile PCR grade water and incubated 10 min at 95 °C and the boiled cells immediately transferred onto ice for 15 min. Tubes were then centrifuged at 13,000 rpm for 10 min and 400 µl of supernatant was transferred in a clean sterile tube and 3 µL was used as template for PCR.

Species confirmation and typing of the isolates was done by PCR, by using selected primers as shown in Supplementary Table 2. *Vibrio cholerae* was confirmed by PCR based on the *ompW* gene encoding for the outer membrane protein (Nani *et al*., 2000). A multiplex PCR was performed to check if the isolates were *V. cholerae* O1 or O139 and if the isolates possessed the genetic potential of producing cholera toxin (Hoshino *et al*., 1998). *Campylobacter jejuni* in the isolated *Campylobacter* colonies was confirmed by a PCR assay based on the presence of the gene *hsp60* encoding the heat stable protein as described by Park *et al*. (2011). *Salmonella* suspected colonies were tested by a PCR as described by Martinez-Ballesteros *et al*. (2012). A PCR assay was performed as to confirm *E. coli* based on 16 S and *uidA* gene (Bei *et al*., 1991).

Antibiotic resistance profile

Selected *V. cholerae* and cefotaxime resistant *E. coli* isolates were tested for their antimicrobial susceptibilities by replica plating onto Mueller Hinton agar supplemented with antibiotic at breakpoint concentrations (Supplementary Table 3) (CLSI 2021). The minimal inhibitory concentrations (MIC) considered to represent resistance to a given antibiotic were those determined by Clinical and Laboratory Standards Institute (CLSI) (CLSI 2021). Isolates with intermediate susceptibility were categorized as being susceptible.

Physicochemical parameters

Physicochemical water quality parameters were measured on site. Conductivity and pH were measured using a PT157 (Palintest, United Kingdom) probe, and water temperature was recorded using a PT155 (Palintest, United Kingdom) probe. The Palintest Turbimeter Plus PTH092 was used to analyze the turbidity of the samples. The Palintest Photometer 7100 PTH7100 was used to analyze free and total chlorine. Meters were calibrated on a monthly basis.

Statistical analysis

Concentrations of *E. coli* (CFU/100 ml) and physicochemical parameters were logarithmically (base 10) transformed. For *E. coli*, removal was calculated as the difference between the concentrations obtained in the raw water (I) and the concentrations obtained in the treated water (T0). The normality distribution of *E. coli* concentrations and physicochemical parameters data was checked by Shapiro-Wilk's test and data analysis and plots were performed using R Studio software *V.* 1.4.1103. The correlations among different parameters using Spearman's correlation test. Spearman's coefficient (r) with P values < 0.05 were considered statistically significant.

Results

Microbiological parameters

E. coli

E. coli was detected in all sample types: in 100% of inlet water (I), in 21% of treated water samples (T0), and in 22% of tap water samples (T1, T2 and T3). *E. coli* counts for I ranged between 26 and 500 CFU/100 ml, with a mean of 185 CFU/100 ml. T0 showed fecal contamination only in May (133 CFU/100 ml), August (37 CFU/100 ml) of 2018 and May 2019 (20 CFU/ml) (Table 3.1). The T1 tap sampling point showed *E. coli* in 3 out of 19 samples with counts less than 10 per 100 ml. At tap T2 *E. coli* was detected in 5 out of 18 samples with *E. coli* concentrations ranging between 11 and 100 CFU/100 ml in August 2018 and June 2019, respectively. In water collected at T3 *E. coli* was detected in 4 out of 16 samples, with concentrations ranging between 6 and 69 CFU/100 ml. In May 2018, all five locations were positive for *E. coli* and/or cefotaxime resistant *E. coli* (Table 3.1).

Cefotaxime resistant E. coli

Cefotaxime resistant *E. coli* counts were reported in all sample's types: in 63% (I), 16% (T0), and 9% (T1, T2 and T3). Counts for I ranged between 1 and 216 CFU/100 ml, with a mean of 14 CFU/100 ml (Table 3.1). Presence of cefotaxime resistant *E. coli* was confirmed in treated water T0 in May (76 CFU/100 ml), August (1 CFU/100 ml) and November (11 CFU/100 ml) of 2018. As for the taps, counts were positive for T1 only in May 2018 (29 CFU/100 ml), for T2 in 3 out of 18 samples with concentrations ranging between 3 and 31 CFU/100 ml in May, August 2018 and June 2019, and for T3 in May 2018 (5 CFU/100 ml) (Table 3.1). Overall a significant correlation with the indicator in water samples was observed ($r = 0.6$, p-value = 3.945e-10). In May 2018, counts were high for all sample's types except for T1 water where cefotaxime resistant *E. coli* was detected but not the indicator (Table 3.1).

Salmonella

Salmonella spp. was not detected in any of the samples.

Table 3.1. *Results of the microbiological analyses: E. coli and cefotaxime resistant E. coli (CTX EC) concentrations (CFU/100 ml) and presence/absence of Vibrio cholerae (VC) and Campylobacter (Camp). Mean, median, minimum and maximum concentrations for each water samples are shown.*

		WTP inlet (I)				**WTP outlet T0**				**T1**				**T2**				**T3**			
		E.coli	CTX EC	*VC*	*Camp*	*E.coli*	CTX EC	VC	*Camp*	*E.coli*	CTX EC	VC	*Camp*	*E.coli*	CTX EC	VC	*Camp*	*E.coli*	CTX EC	VC	*Camp*
2018	March	505	5.4	+	-	0	0	-	-	4	0	+	-	7	0	-	-	n/a	n/a	n/a	n/a
	April	414	3.6	+	+	1	0	-	-	0	0	-	-	0	0	-	-	n/a	n/a	n/a	+
	May	165	3.6	+	+	133	75.7	-	-	0	28.8	-	-	91	30.6	-	-	69	5.4	-	-
	June	107	0.9	+	+	0	0	-	-	0	0	+	-	0	0	-	-	0	0	-	-
	July	212	0	+	+	0	0	-	-	3	0	+	-	0	0	-	-	0	0	-	-
	August	70.2	2.7	+	+	0	0	-	-	0	0	-	-	32	19.8	-	-	0	0	-	-
	August	53.1	11.7	+	+	37	0.9	+	-	0	0	+	-	11	0	-	+	0	0	-	-
	September	50.4	0	+	+	0	0	-	-	0	0	-	+	0	0	-	-	13	0	+	-
	November	28.8	5.4	+	+	0	10.8	-	-	9	0	+	-	0	0	-	+	7	0	+	+
	December	27.9	0	+	+	0	0	+	-	0	0	-	+	0	0	-	-	0	0	-	-
2019	February	61.2	13.5	+	+	0	0	-	+	0	0	-	-	0	0	-	+	6	0	-	+
	March	67.5	7.2	+	+	0	0	+	-	0	0	-	-	0	0	-	-	0	0	+	+
	April	42.3	0	+	+	0	0	-	-	0	0	+	+	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
	May	149	216	+	+	20	0	-	-	0	0	+	+	0	0	-	-	0	0	+	-
	June	136	1.8	+	+	0	0	-	-	0	0	-	-	100	2.7	-	-	0	0	+	-
	July	67.5	2.7	+	+	0	0	-	-	0	0	+	-	0	0	+	-	0	0	-	-
	August	255	0	+	+	0	0	-	-	0	0	-	-	0	0	-	-	0	0	-	-
	September	266	0	+	+	0	0	-	+	0	0	+	-	0	0	-	-	0	0	-	-
	October	262	0	+	+	0	0	-	-	0	0	+	-	0	0	-	+	0	0	-	-
	N	19	19	19	19	19	19	19	19	19	19	19	19	18	18	18	18	16	16	16	17
	N positive samples	19	12	19	18	4	3	3	2	3	1	10	4	5	3	1	4	4	1	5	4
	% positive samples	100	63	100	95	21	16	16	11	16	5	58	21	28	17	5	22	25	6	31	24
	Mean	185	14			9	1			0	1			12	3			4	0		
	Std Dev	162	49			31	7			0	7			31	8			17	1		
	Min	26	0			0	0			0	0			0	0			0	0		
	Max	500	216			133	29			0	29			100	30			69	5		

Vibrio cholerae

V. cholerae was detected in 42% of all water samples tested: 100% (I), 11% (T0), and in 23% (T1, T2 and T3) (Table 3.1). All *V. cholerae* confirmed isolates where genotyped as non-O1/non-O139 by PCR. In four inlet water samples (September - December 2018 and August 2019) *V. cholerae* non-O1/non-O139 was isolated with the genetic potential of producing cholera toxin (ctx^{+}). All other *V. cholerae* non-O1/non-O139 isolates were CTX negative.

Campylobacter

Campylobacter spp. was detected in 36% of the water samples tested (n = 29): 95% (I), 10% (T0) on February and April 2019, and 23% (T1, T2 and T3) on April 2018, from August 2018 through May 2019 and in August 2019 (Table 3.1).

Antibiotic resistance

Selected cefotaxime resistant *E. coli* confirmed isolates were tested for susceptibility to 12 antibiotics. All 15 strains tested showed resistance to multiple antibiotics. As expected all isolates were resistant to cefotaxime, all were also resistant to ampicillin. Additionally, 10 isolates were resistant to streptomycin, 7 to tetracycline and 1 isolate was also resistant to chloramphenicol (Table 3.2). Isolates resistant to at least one other antibiotic were detected in all types of water samples tested. Ten isolates (62%) showed a MDR profile, being resistant to at least three antibiotics, five of these were isolated from I, two from T0, one from T1 and two from T2. All 70 *V. cholerae* non-O1/non-O139 confirmed isolated were tested for susceptibility to 11 antibiotics. Of these, 69 isolates were resistant to ampicillin, 35 (51%) were resistant to streptomycin, 9 (13%) were resistant to gentamycin, and 1 isolate was resistant to tetracycline (Table 3.2). Nine isolates (13%) showed a MDR profile, being resistant to at least three antibiotics, and of these 5 isolates originated from I, 2 from T1 and one from each T1 and T3.

Table 3.2. *Number (No.) of strains, isolation source and antibiotic resistance pattern of cefotaxime resistant E. coli and Vibrio cholerae. Cefotaxime (CTX), ampicillin (AMP), chloramphenicol (CHL), tetracycline (TET), gentamycin (GEN) and streptomycin (STR).*

Source	**Cefotaxime resistant *E. coli***		***Vibrio cholerae****	
	No. of isolates	**Antibiotic resistance**	**No. of isolates**	**Antibiotic resistance**
Inlet WTP	4	CTX, AMP	16	AMP
	4	CTX, AMP, TET, STR	21	AMP, STR
	1	CTX, AMP, TET, STR, CHL	5	AMP, STR, GEN
			1	AMP, STR, TET
Outlet WTP	1	CTX, AMP	3	AMP
	1	CTX, AMP, TET, STR		
	1	CTX, AMP, STR		
T1	1	CTX, AMP, TET, STR	9	AMP
			4	AMP, STR
			1	AMP, STR, GEN
T2	2	CTX, AMP, STR	2	AMP, STR, GEN
T3			6	AMP
			1	AMP, STR, GEN
Total	15		69	

* Information was not shown in the original paper.

Physicochemical parameters

Free chlorine levels varied greatly among sample type. The highest mean value of chlorine was 1.2 mg Cl_2/L detected at the outlet of the WTP (Figure 3.2, Supplementary Table 4). Free chlorine levels at T0 ranged between 0.3 and 2.6 mg Cl_2/L. In household tap water the level of free chlorine ranged between 0.1 mg Cl_2/L at T1 in April 2019 to 2.3 mg Cl_2/L at T2 in April 2018. T1 household water had the lowest mean values of free chlorine (Figure 3.2). In 7% of all the samples taken at household taps (n = 44) free chlorine was less than 0.2 mg Cl_2/L, in 57% had values comprised between 0.2 and 1 mg Cl_2/L, and in 36% of the samples the free chlorine exceeded 1 mg Cl_2/L (Figure 3.2, Supplementary Table 4).

Differences observed between free chlorine values detected at T1 and T0, T2 and T3 were significant at the 95% interval ($p < 0.05$). Turbidity of the inlet water ranged between 0.5 and 12.3 NTU (Figure 3.2, Supplementary Table 4). For T0 water turbidity ranged between 0.1 and 10.3 NTU. Turbidity values recorded at household taps ranged from a minimum of 0.8 NTU to a maximum of 22.7 NTU. 93% of the total number of tap water samples (n = 44) analyzed were greater than 1.0 NTU of which 45% did not comply with the national standards of 5.0 NTU. The highest turbidity level at tap was recorded in March 2018. Statistically, the differences observed were not significant at 95% interval ($p < 0.05$). The average temperature of the water remained stable at around 23°C at all sampling locations throughout the study period. The average pH of the water ranged between 8.1 and 8.3, values fell within the range of legal requirement for drinking water (pH 6.5–8.5).

Conductivity of the treated and household tap water ranged between 298 µS/cm and 721 µS/cm, within the range of 50−2,000 µS/cm as legal requirements, with average values above 500 µS/cm (Figure 3.2, Supplementary Table 4).

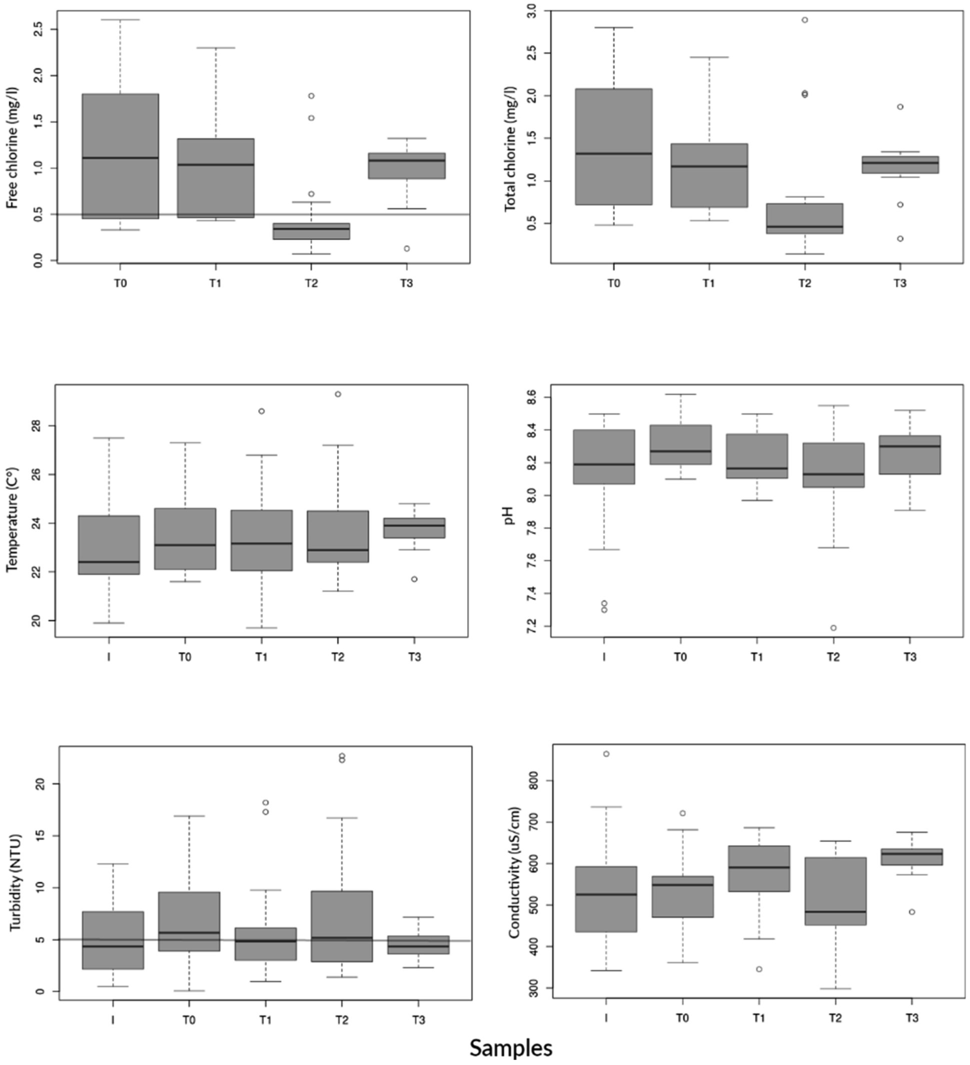

Figure 3.2. *Box-Whiskerplots of physicochemical parameters of the water collected at sampling points. Grey lines show legal requirements for free chlorine (0.5 mg/l) and turbidity (5 NTU). The box represents the median and quartiles, the whiskers show the 95%-interval and dots are outliers.*

Discussion

During this study, the level of fecal contamination of the Inkomati river water (mean value of 185 CFU/100 ml) was lower than levels reported for surface water in the neighboring Limpopo Province in South Africa (mean values of 0.3 to 1.4 × 10^4 CFU/100 ml) (Mwabi *et al.*, 2012), and lower than three rivers in Ecuador (128 to 1,248 MPN/100 ml) (Rao *et al.*, 2015). *E. coli* counts were in average higher in the dry season (April-October) than in the wet season (November-March) indicating that lower level of river water affects the concentration of microbes.

When considering the removal of fecal contamination at the Moamba WTP, we could not detect *E. coli* in 79% of the treated samples. Samples showing *E. coli* contamination were collected in April, May and August of 2018 and June of 2019, and showed 2.62, 0.09, 0.16 and 0.33 $\log_{10}$ removal, respectively. In 95% of tap water samples the turbidity was greater than WHO level (1.0 NTU) to guarantee an effective disinfection process (LeChevallier and Au, 2004; WHO 2017). High turbidity in filtered water is associated with poor removal of pathogens, sloughing of biofilms and ingress of contaminants through broken pipes (LeChevallier and Au, 2004).

Removal efficiency of fecal contamination did not correlate significantly with any of the physicochemical parameters assessed. Our findings reported re-contamination during distribution at all three household taps, where 23% of water samples exceeded national standards for potable water parameters for *E. coli*. Fecal contamination monitoring of a IWS system in India reported 32% of samples exceeding WHO drinking water quality guidelines (Kumpel *et al.*, 2014). Water collected from household taps was generally free of *E. coli* contamination as 77% of the samples consisted of non-detects. Prevalence of household tap samples contaminated with *E. coli* were 16%, 28% and 25% for Cimento, Matadouro and Bairro Sul, respectively. These values are in line with those reported in another study conducted on the same WTP (Van den Berg *et al.*, 2021). Similar values of *E. coli* contamination were reported in Maputo tap water (23% of samples) (Marcario, 2022). Also, the prevalence of fecal contamination observed in our study reflects the variability observed in other IWS distribution networks (Kumpel *et al.*, 2014; Shaheed *et al.*, 2014; Bivins *et al.*, 2017).

Limitations of the fecal indicator paradigm have long been reported with the inconsistent relationships between FIB occurrence, enteric pathogens, and associated health risks (Korajkic *et al.*, 2018; Kelly *et al.*, 2020; Charles *et al.*, 2020; Nowicki *et al.*, 2021). The absence of *E. coli* does not eliminate the risk of the water being contaminated by enteric pathogens that may show a higher resistance to disinfection and the ability to persist in the distribution network in biofilms (Holcomb and Stewart, 2020; Kumpel *et al.*, 2014). In our study we have detected the presence of pathogens such as *Vibrio cholerae* and *Campylobacter* throughout the water supply in Moamba even when *E. coli* was not detected. Eleven percent of the treated water samples were positive for *V. cholerae* and *Campylobacter*, while the two pathogens were detected in 18% and 15% of tap water samples, respectively, in absence of *E. coli*. *V. cholerae* survives better in estuarine waters than *E. coli* resulting in poor correlation of *V. cholerae* levels with fecal coliform concentrations in estuarine waters (Colwell and Huq, 1994). Also, *V. cholerae* non-O1 is a natural inhabitant of waters and therefore it may have a greater fitness in water than the enteric commensal *E. coli*. In a recent study monitoring treated water quality, *V. cholerae* abundance was higher compared to *E. coli* after three days (Djaouda *et al*., 2020). The extended survival of *V. cholerae* in treated water may have serious public health implications. *V. cholerae* non-O1/non-O139 was present in 74% of samples from Inkomati river. This pathogen has been widely reported in surface water in Mozambique and worldwide (Taviani *et al*., 2008; Hasan *et al*., 2013; Trindade *et al*., 2021). Nine *V. cholerae* non-O1/non-O139 isolates were positive for the presence of the *ctxAB* genes encoding the cholera toxin, the virulence factor causing the severe diarrhea associated with cholera disease. These isolates originated from inlet (5), treated water (1) and tap water (3) samples. Although rare in the environment, the detection of *ctx* genetic determinant in *V. cholerae* non-O1/non-O139 has been reported in several countries (Bhattacharya *et al*., 2006; Biswas *et al*., 2022), including Mozambique (Colombo *et al*., 1994). The presence of this microorganism has been linked to cases of diarrheal diseases, representing a risk for the population consuming the water (Igere *et al*., 2022).

Detection of *Campylobacter* in 10% of treated water samples and in 19% household tap water samples further suggested environmental contamination and persistence of pathogens along the Moamba drinking water distribution network. The contamination of water by *Campylobacter* can be linked to the presence of a major hatchery and the widespread smallholder family poultry producers in Moamba district (FAO 2013). *Campylobacter* detection did not correlate with the occurrence of *E. coli* as indicator of fecal contamination in treated and household tap water samples. In other studies the presence of *Campylobacter* spp. showed a lower correlation with fecal indicators with respect to other pathogens such as *Cryptosporidium*, *Giardia*, pathogenic *E. coli*, and *Salmonella* spp. (Korajkic *et al*., 2018).

Antibiotic resistant bacteria have been increasingly reported globally, not only restricted to clinical settings but also recovered from environmental samples, especially water. The pandemic diffusion of Extended Spectrum Beta Lactamase (ESBL)-producing Gram-negative bacteria in drinking water distribution systems is a major health concern, affecting mostly low-income countries in Asia and Africa (Tacão *et al*., 2014; Mahmud *et al*., 2020; Johnston *et al*., 2020; Macario 2022). In our study, cefotaxime resistant *E. coli*, and *V. cholerae* strains resistant to several class of antibiotics were detected in inlet, treated and household water samples. The high prevalence (62%) of MDR *E. coli*, indicated that different classes of antibiotics are being co-selected with β-lactam resistance in the aquatic environment. A much lower incidence (10.7%) of ESBL *E. coli* was reported in tap water of Maputo (Macario 2022). Also, 13% of *V. cholerae* non-O1/non-O139 isolates showed a MDR profile. Our results confirmed the rapid dissemination of antimicrobial resistance in environments that are not directly affected from major clinical inference, implying an overuse and misuse of antibiotics in local communities (Iskander *et al*., 2020).

Conclusion

From our finding we can conclude that the detection and monitoring of major microbial pathogens at different points of the drinking water treatment process and distribution network is crucial for water quality management, especially in IWS where non- or low pressurized pipes permit re-contamination of treated water. A contaminated water distribution system may act as source of waterborne pathogens and a mean for spreading them between communities. On the other hand, in settings like the ones surveyed in our study where low level of sanitation and hygiene allow for circulation of FIB and pathogens between humans, animals and household environment, measurement of standard tap drinking water quality alone may not be sufficient to accurately predict the safety and health implications associate with its consumption. New practices are needed to support monitoring approaches that go beyond the routine measurement of *E. coli* or FIB as mean to assess drinking water quality.

Supplementary information

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Chapter 4

How current risk assessment and risk management methods for drinking water in the Netherlands cover the WHO water safety plan approach

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Abstract

In the Netherlands, ten drinking water companies provide safe and sufficient drinking water to the general population. To guarantee safe drinking water the World Health Organization (WHO) developed a water safety plan (WSP), a risk assessment and a risk management (RA/RM) framework. The objective of the study was to identify legally required RA/RM approaches, to document application of RA/RM activities at Dutch drinking water companies and to determine to what extent these RA/RM activities as a whole cover all the elements of the WHO WSP approach. This study could be of interest to both managers of large water utilities and decision makers.

The assessment was performed by means of a policy review and interviews with two to four staff members involved in RA/RM from all ten Dutch drinking water companies combined with a joint workshop. The drinking water companies are well aware of the potential hazards and risks that can influence the drinking water quality. To guarantee the supply of safe and sufficient drinking water, the Dutch drinking water sector uses six different legally required RA/RM approaches. This study shows that by using the six legally required RA/RM approaches, all WSP steps are covered. WSP entails a generic risk assessment for identifying all hazards and hazardous events from source to tap, whereas the six legally required RA/RM each focus on specific risks at an advanced level. Each risk assessment provides information on specific hazards and hazardous events covering a part of the water supply chain. These legal requirements are complemented with additional RA/RM activities at sector and water company level such as codes of practices and standard operating procedures. The outcomes of all RA/RM approaches combined provide information from source to tap. When using multiple RA/RM approaches, it is crucial to share and combine information derived from the different activities.

Introduction

Drinking water is the final product of the production chain – from source to tap – which is monitored to ensure drinking water of sufficient quality and thus protect public health (WHO 2015). Although monitoring of drinking water as a final product has been the norm to determine if drinking water is safe and clean, over the past decades it has become clear that this monitoring can often be too little and too late (WHO 2015). Consequently, the detection of risks might be too late or might not happen at all, which may lead to infectious diseases or other negative health effects (WHO 2015). Therefore, a preventative risk based approach for the whole drinking water supply as a system has been introduced, including risk assessment (RA) for identification of the risks and risk management (RM) for managing the risk, generally referred to as an RA/RM approach. RA/RM approaches have been introduced worldwide, not only for drinking water but also for other waters, such as bathing waters (bathing water profiles) and shellfish production areas (sanitary surveys) (EU 2006; WHO 2010).

In 1994, Havelaar explored the application of hazard analysis and critical control point (HACCP), a food safety management system, to drinking water supply systems (Havelaar, 1994). In some countries, for example Switzerland, the drinking water supply was also regulated through the law for food protection and therefore already required HACCP. Between 1999 and 2001, an international group of experts discussed the potential to increase consistency in approaches of assessment and management of water-related microbial hazards, which led to the 'Stockholm Framework' (Fewtrell and Bartram, 2001). This further explored the application of HACCP to the drinking water supply. The third edition of the WHO Guidelines for Drinking-Water Quality (WHO 2004) included the 'Framework for Safe Drinking-water', which encompasses setting health-based targets, an RA/RM approach and independent surveillance. The risk management approach was referred to as a water safety plan (WSP). At the same time, the International Water Association published the Bonn Charter for Safe Drinking Water, which provides a high-level framework describing the operational and institutional arrangements that are basic requirements for managing water supplies from catchment to consumer (IWA 2004). Various publications provided further support for the implementation of a WSP, such as the WSP manual (Bartram *et al.*, 2009) and WSP for small community water supplies (WHO 2012). WSPs require a RA including all steps in the water supply from catchment to consumer, followed by implementation of control measures and by improvement with a focus on high priority risks (WHO 2011, 2017a).

Over the last decade, WSPs have been successfully implemented in both high- and low-income countries. To date, WSPs are being implemented to varying degrees in 93 countries globally, with 30% of countries at an early adoption stage; 46 countries report having policy or regulatory instruments that promote or require WSPs and in another 23 countries such instruments are under development (WHO 2017b). There are reports of many benefits from WSP application, such as improved system management of water supplies; increased awareness, knowledge and understanding among staff; improved communication and collaboration with other stakeholders and also within water supply companies; and improved water quality (Gunnarsdottir *et al.*, 2012). The way WSP is applied varies with the development level of the water supply and the resources available.

In the Netherlands, drinking water is produced from surface water (38%) and groundwater (62%) to provide the Dutch population with safe, clean and sufficient drinking water (VEWIN 2017). In the Netherlands, only a few hundred small private supplies, mainly campsites and recreational parks, produce drinking water to supply staff or guests (ILT 2018). Ten very large public drinking water companies serve the general population, serving between 435,000 – 5.7 million people each (VEWIN 2017). These drinking water companies provide drinking water by collecting and treating groundwater or surface water and providing it to the customer’s tap via a pipeline network. The drinking water production and supply are prone to contamination with microbial and chemical hazards from humans and their activities in the environment or from naturally occurring contamination (WHO 2004). Various hazardous events can impact the chemical, microbial or physical quality of the drinking water somewhere between the source and the tap, such as sewage discharge, chemical waste disposal and damaged pipes in the distribution network due to external construction works (WHO 2004). The production and distribution of safe and sufficient drinking water by drinking water companies in the Netherlands is regulated under the Dutch Drinking Water Act (I&W 2009). The human Environmental and Transport Inspectorate (ILT) of the Ministry of Infrastructure and Water Management (I&W) is the governmental body that supervises the water supply companies, with the Minister of I&W having the ultimate responsibility for ensuring safe drinking water (I&W 2009).

Although there is no specific policy or legislation mentioning the specific wording WSP in the Netherlands, the policy and legislation is based on the same principles of RA/RM. Guaranteed continuous drinking water supply and quality of drinking water have always been the focus of the national policy and the Dutch drinking water companies (De Moel *et al.*, 2006). In 2001, already before WHO and IWA launched the WSP, the first de facto WSPs were initiated by the drinking water utilities in the Netherlands (Smeets and Puijker, 2013). The software program MarRiskA (Van Lieverloo *et al.*, 2003) was developed as a tool to facilitate this RA implementation in a uniform format. A number of companies collaborated on the development of tools at the international level such as the TECHNEAU Hazard Database (Beuken *et al.*, 2008). Until 2010, different drinking water companies completed approximately seventeen WSPs applying the principles of the WHO approach (Smeets and Puijker, 2013). In 2013, the Dutch water companies discussed the need to uniformly implement WSP as a framework for RA/RM (Smeets and Puijker, 2013). At that time, it was considered an extra burden on top of existing risk management requirements and practices with no added benefit, as most of the steps in WSP were presumed to already be in place.

This study was performed by the National Institute for Public Health and the Environment (RIVM) and KWR Watercycle Research Institute (KWR) on behalf of the Ministry of I&W. The goal of this study is to identify applied RA/RM components in policy and legislation and activities at all ten Dutch drinking water companies and to determine to what extent these RA/RM activities as a whole cover the elements of the WHO WSP approach.

Methods

To construct an overview of all RA/RM approaches and activities, information was gathered by

1. Conducting a policy review to identify all relevant legislations and policies on RA/RM for drinking water.
2. Identifying all RA/RM activities conducted per drinking water company
   - Staff members from drinking water companies were selected based on their experience in RA/RM. They were interviewed using a questionnaire to identify all RA/RM activities applied within their drinking water company. The questionnaire was designed according to the steps of a WSP.
   - Based on the policy review and the interviews, a draft overview of the data per company was made and sent back to the interviewees for feedback. After the collection of all feedback, the data from all drinking water companies was collated to provide an overview of all RA/RM activities with similarities and differences between drinking water companies.
   - A workshop was organized to discuss the results of the interviews and to examine the current use of risk-based management in the production of drinking water.

The information gathered from the policy review and interviews were examined to determine to what extent the RA/RM approaches and applied activities cover the WHO WSP approach according to the WSP Manual (Bartram *et al.*, 2009) and to identify any gaps, possible improvements and best practices.

Results

Policy review: legal requirements for risk assessment and risk management

In the Netherlands, legislation does not mention WSP specifically, but prescribes RA/RM in legislation and policy. Based on the policy review, the following RA/RM approaches were identified:

- Quantitative microbial risk analysis (QMRA)
- Drinking water protection files
- Disturbance risk analysis (DRA) as part of the drinking water supply plans
- *Legionella* prevention control in drinking water installations
- Code of hygienic practice for drinking water supply
- Monitoring program drinking water quality – risk based

All RA/RM approaches corresponded to certified quality management systems and standards, which are legally required. All water companies were certified according to the new ISO 9001: 2015 standard (ISO 2015a). RA/RM activities are an explicit part of this new version of the ISO 9001 standard for quality management, and ISO 14000 offers an environmental framework for RA/RM (ISO 2015b). Other legal requirements support RA/RM and contribute to the protection of drinking water quality from source to tap. Examples are requirements for intake of raw water, a specific requirement with regard to identifying compounds of concern and hygienic requirements for materials and chemicals used in the drinking water system.

QMRA

The Dutch Drinking Water Act (I&W 2009) prescribes that the index pathogens (Entero)viruses, *Cryptosporidium*, *Giardia* and *Campylobacter* should not exceed an infection risk of one infection per 10,000 individuals per year. To demonstrate the microbial safety of drinking water, Dutch drinking water companies must conduct a QMRA at least every four years for these so-called index pathogens. Since 2005, Dutch drinking water companies have conducted QMRAs as described in the Dutch Inspectorate Guideline 5318 (Anonymous, 2005) for all surface water production plants. The QMRA includes a system description, as well as the identification of possible microbial hazards and hazardous events and a monitoring requirement from source to treatment. Drinking water companies using surface water for the production of drinking water estimate the infection risk using the computational tool QMRAspot (Schijven *et al.*, 2011). The estimated risks are evaluated and discussed in close collaboration between drinking water companies, RIVM and ILT (Bichai and Smeets, 2013). QMRAspot facilitates the evaluation of effective preventative measures and supports policy makers and other involved parties in risk prioritization and the formulation of mitigation strategies. ILT advises the drinking water companies when to take action or develop an improvement plan.

Drinking water protection files

As described in the policy brief on Drinking Water (I&W 2014), competent authorities and drinking water companies agreed to jointly set up drinking water protection files for intake zones. The drinking water protection files contain information about the quality of the resources, sources of pollution and the vulnerability of the water system. Within the drinking water protection files, risks regarding all possible contaminants of the drinking water are assessed. Based on the assessment, different stakeholders are involved to identify measures, aimed at prevention and risk management. Monitoring the control measures and a regular update of the drinking water protection file are also part of this approach. The drinking water protection files have to be updated every six years (Wuijts *et al*., 2017). In the Netherlands, the drinking water protection files are part of the implementation of articles 7, 8 and 11 provisions of the Water Framework Directive (2000/60/EC). The protection files are also an instrument to work together and exchange information between the drinking water companies, competent authorities responsible for Water Framework Directive implementation and other stakeholders. The drinking water companies use the outcomes of the analysis for their risk based monitoring programs.

Disturbance risk analysis

As a requirement in the Drinking Water Decree (I&W 2011a), drinking water companies draw up a DRA as part of the drinking water supply plan. They assess the risks of a long list of threats and hazards which potentially affect the quantity or quality of the water supply. Based on the outcomes of the DRA, additional control measures are included in the drinking water supply plan, to minimize risks for the public drinking water supply. These drinking water supply plans (including the assessment) have to be revised every four years and are approved by the ILT.

Legionella prevention control in drinking water installations

Legionella prevention control is mandatory for drinking water installations in buildings used by people with higher risk for *Legionella* infection, such as hospitals, retirement homes, hotels and swimming pools (I&W 2011a). The owner of the building is responsible for assessing the risks according to Dutch regulations (microbial hazards), which should be done by a certified person or organization. Furthermore, a control plan is required that describes control measures such as flushing, temperature control and a monitoring program.

The drinking water company supplying the water inspects whether the owner of the building has fulfilled their responsibility according to *Legionella* prevention. Non-compliances within the monitoring of *Legionella* need to be reported to the ILT. If improvement is necessary after the inspection by the drinking water company, an improvement plan has to be made by the owner.

Code of hygienic practice for drinking water supply

The Dutch Drinking Water Decree (I&W 2011a) refers to European standards and Dutch codes for working hygienically (Meerkerk 2016). The code of hygienic practice "Drinking water", was made by the drinking water companies as an integral system for quality management and risk management to ensure the microbiological safety of drinking water during storage and distribution. The main topics of this code of hygienic practice are:

- proper infrastructure and hygienic requirements for materials and chemicals (I&W 2011b);
- preventive management for working hygienically;
- sensitive detection systems for contamination and deviations;
- effective corrective actions for contamination and deviations;
- periodic inventory and evaluation of risks; and
- instructions and training for employees to do construction work according to hygienic rules.

Monitoring program drinking water quality

The Drinking Water Decree prescribes that drinking water should meet the regulation, and by complying to the regulation the drinking water companies ensure the supply of safe drinking water. Drinking water companies are required to set up an annual monitoring program as prescribed by the regulations. The monitoring program, based on the assessment of the microbial, chemical and physical risks as described in the EU Directive 2015/1787/EC (EU 2015), entails monitoring from source to tap. The monitoring programs are updated annually, and have to be approved by the ILT. In addition to these monitoring programs, the drinking water companies perform screenings. The legislation prescribes alert values for known substances and for unforeseen substances to trigger further research to identify the risk.

Identify RA/RM activities per drinking water company

Two of the authors interviewed two to four staff members who were involved in RA/RM within their company. Dutch drinking water companies were obliged to carry out the legally required RA/RM approaches, which was underlined during the interviews with all drinking water companies. Furthermore, the representative staff members provided us with information on sector- or company specific RA/RM activities per WSP step they practiced which was complementary to the legal requirements.

All information was documented by the interviewers, and sent to the interviewees for feedback. After collecting all feedback, the data from all drinking water companies was collated to provide an overview of all RA/RM activities. Similarities and differences between drinking water companies were thus identified. The results from the interviews and policy review were presented at a workshop organized at RIVM, which was attended by 52 people from the Ministry of I&W, drinking water companies, ILT, RIVM and KWR. During the workshop, integrated RM, risk analyses and data and policy on RA/RM were discussed in breakout sessions. The moderators of the breakout sessions collected the information and presented the outcomes at the end of the workshop. The next paragraph contains an overview on which RA/RM activities were undertaken per WSP step, based on the policy review, interviews and workshop, and we outline experiences from the drinking water companies.

Coverage of each step of the WHO WSP

All drinking water companies reported teams that focus on RA/RM (**WSP step 1**), but sometimes different people or teams were involved in the different RA/RM activities as well as all legally required RA/RM approaches. The teams were mostly internal teams of a broad and multidisciplinary composition, and occasionally the teams were assisted by external experts. Two drinking water companies had an overarching team responsible for RA/RM, whereas the other eight water companies had several teams involved in RA/RM. For communication between different teams all companies nominated a linking pin: a person or a department.

Five of the legally required RA/RM approaches prescribed a system description and all ten drinking water companies had a complete and up-to-date description of the drinking water system (**WSP step 2**). The system description from source to tap also included working practices and/or procedures and was digitally available at all drinking water companies. Over 20 different software systems for the system description were present for the different components (source-treatment-tap) and sometimes even within one component various systems were available to record the data. Examples of these software systems were geographical information systems, design software and network information systems. Because of the use of different software systems for the water supply system descriptions from source to tap, these systems were not automatically linked to each other. Only three companies had linked all system descriptions from source to tap (including processes and procedures).

All drinking water companies have always been aware of the potential risks to the drinking water supply and have put a lot of effort towards reducing these risks. Besides all six legal requirements, the Dutch drinking water companies had sector or company specific activities for identifying hazards and hazardous events and for performing a risk assessment (**WSP**

step 3). Examples applied in some drinking water companies are:

- internal audits focus on irregularities, incidents or possible risk and follow up;
- inspections and technical screening;
- trend analysis for identifying future risks;
- risk analyses for asset management, such as failure mode effect and criticality analysis (FMECA) and hazard and operability study (HAZOP);
- a WSP approach for identifying hazards and hazardous events from source to tap;
- risk analyses for their monitoring, and screenings for non-regulated substances.

All drinking water companies prioritized the risks. However, there were many different ways of weighing the risks, varying from quantitative risk assessment to expert opinion. For some assessments, the method for weighing the risks varied between drinking water companies, but also between the prioritization methods used within one drinking water company. Documentation of the identified risks varied per drinking water company, depending on the available systems: one central database or different files or systems per RA/RM approach.

The drinking water companies had many different control measures in place to reduce potential risks (**WSP step 4**). Control measures were prescribed by the legal requirements for RA/RM, but also by other legal requirements, advisory guidelines or company specific management procedures. During the interviews, examples were given of control measures in place to prevent contamination of drinking water by the drinking water companies. Textbox 4.1 contains some examples.

To ensure that control measures work effectively, the Dutch drinking water companies assess the effectiveness of control measures. For this assessment, field data from the specific drinking water company is most valued to assess the efficiency of control measures, followed by pilot data generated by the specific drinking water company. If location specific data are unavailable literature, study outcomes or trend analyses are also considered. Within the sector, the drinking water companies work together in research to validate control measures (Brouwer *et al*., 2018). Examples of such joint research are reduction of pathogens by slow sand filtration (Schijven *et al*., 2013), soil infiltration (Hornstra *et al*., 2018) or UV disinfection (Hijnen *et al*., 2006) and breakdown of micropollutants by UV-peroxide advanced oxidation (IJpelaar *et al*., 2010).

As for the legal requirements for risk assessment, all drinking water companies also prepared improvement plans for potential risks identified (**WSP step 5**) based on these company specific risk assessments.

Textbox 4.1. *Examples of control measures in place to prevent contamination of drinking water.*

Existing control measures in the catchment and at the abstraction:

- groundwater protection area;
- instruction from Technology platform for transport, infrastructure and public space on careful digging process;
- management agreements regarding existing and known contaminants in groundwater protection areas;
- manure regulation policy;
- requirements for intake of raw water;
- specific requirement with regard to identifying compounds of concern;
- policy on discharge permits (reducing the amount of pesticides);
- agreements with the Safety Regions for timely alerts;
- policy for soil protection;
- protection against (deliberate) pollution and calamities;
- association of River Waterworks (RIWA) and/or Maas alarm model for source water monitoring;
- drinking water protection files; and
- policy for water protection.

Existing control measures in the treatment and the distribution:

- treatment process (e.g. UV disinfection, slow sand filtration, soil infiltration, ozonation, activated carbon filtration and advanced oxidation process);
- products and chemicals in contact with drinking water used by the water supplier need to be certified according Regulation (I&W 2011b);
- preventative maintenance;
- process automation system;
- work permit for external employees (e.g. construction);
- limited access for employees and additional rules for visitors; and
- hygienic areas: colour code (e.g. blue – raw water; red – disinfected water).

Existing control measure at household level (consumer):

- preventive notice to boil drinking water.

Apart from the DRA, all legal requirements prescribe the microbial and physicochemical parameters to be monitored for different purposes, such as source water quality monitoring, operational and verification monitoring. For operational monitoring (**WSP step 6**) the Dutch legislation prescribes parameters to be tested for monitoring of control measures. Examples are measuring pH, turbidity, flow rate, dosing of chemicals and pressure, which are measured online at most water companies. Besides measurements, also visual inspections are periodically done for both infrastructure and procedures.

Furthermore, all drinking water companies had additional water quality monitoring at the source, treatment and distribution (extralegal measurements), such as additional samples, biomonitoring and screening for unknown and non-standardized emerging substances. All ten drinking water companies reported procedures for abnormalities in the control process and water quality measurements. In some drinking water companies, the completion of the procedures for abnormalities differed, but all operated 24/7. In all drinking water companies the corrective action procedures were known to act on anomalies.

The data, derived from monitoring programs, were stored in various data collection systems and databases. Six drinking water companies indicated that the (monitoring) data were already linked, but that there was room for improvement as well. Those drinking water companies that had not linked (monitoring) data had the intention to link information from the databases.

Effectiveness of the WSP was verified using three different methods: compliance monitoring, auditing RA/RM and customer satisfaction surveys (**WSP step 7**).

1. Compliance monitoring was used to determine the effectiveness of RA/RM-activities. Therefore, all drinking water companies showed the use of legal requirements, such as water quality monitoring, including QMRA, and the performance comparison (benchmark), including substandard delivery minutes to verify the effectiveness of the RA/RM activities. *Legionella* prevention prescribes monitoring at household level for the detection of *Legionella*. In addition, specific water quality monitoring, registration of failures and technological audits were shown to be used to determine the effectiveness of the RA/RM-components. The Drinking Water Decree describes the framework for reporting defects to ILT (I&W 2011a).
2. With the legal requirement of ISO 9001 (for both versions 2008 and 2015) the processes around drinking water supply were subjected to internal and external audits (ISO 2008; ISO 2015a). Internal and external audits were also obligatory for ISO 14000 (ISO 2015b).
3. Customer satisfaction surveys were done by all drinking water companies as part of the performance benchmark (VEWIN 2016). All drinking water companies also have 24/7 customer complaints services. Evaluation of the customer satisfaction surveys provided information on customer perception of water quality.

The Drinking Water Act requires drinking water companies to have certified quality management systems (I&W 2009). Management involvement is important for creating a framework for the implementation of RA/RM by addressing financial and other resources. Furthermore, management plays an important role in the development of procedures and communication in identifying potential risks, and improvement in the organization (**WSP step 8**). As part of the quality management system, all drinking water companies have standard operating procedures for their daily work. Furthermore, the legislation prescribes to only test water at accredited laboratories that automatically should have standard operating procedures. The code of hygienic practice for drinking water supply prescribes procedures for quality and risk management. The Dutch Drinking Water Act holds legal requirements with respect to an uninterrupted supply of drinking water during ‘normal or undisturbed’ as well as ‘disturbed’ circumstances, in the present as well as in the future.

These requirements mostly focus on water quantity and include elements like emergency response, security of the water supply system, and the supply of drinking water during failure of the water supply system. All water supply companies have implemented security and contingency plans.

All drinking water companies carried out many supporting activities to raise awareness of the risks of unsafe drinking water and the risks of contamination (**WSP step 9**). In two legally required approaches, the code of hygienic practice for drinking water supply and drinking water protection files, supporting activities were explicitly mentioned. Furthermore, the interviewees provided several RA/RM supporting activities, see Textbox 4.2.

Periodic reviews were carried out to keep the system description up-to-date (**WSP step 10**). The validity of the system description for the distribution network was checked and corrected as part of the daily activities where necessary, using tablets so that changes or observations in the field could be included directly in the description. The system descriptions for abstraction and treatment were stable and therefore up to date. Periodic reviews also took place for the legally required RA/RM approaches, including RA and improvement plans. The specific cycle of review per legally required RA/RM is described above (see legal requirements for RA/RM).

Textbox 4.2. *Examples of supporting activities, given during the interviews with the drinking water companies.*

Communication with consumers

- public participation for sampling;
- newsletter;
- website that contains information from source to tap. These websites also contain public water quality data and information about possible faults, disconnection and activities;
- social media (Twitter, Facebook) and email to inform and engage consumers;
- open days, information sessions, campaigns, meetings or visitor centers.

Training

- training, training modules and courses for employees and subcontractors;
- certification of subcontractors to demonstrate awareness of the risks to drinking water before starting restoration and maintenance work; and
- training and exercises on what to do in case of an emergency or disruption.

Communication with different stakeholders

- creating awareness and sharing information with municipalities, provinces, water authorities, health safety regions and related sectors such as other utilities or railroad companies; and
- regular contact with fire brigades, police and nature conservation organizations.

Since the 1990s, the drinking water companies had developed guidelines for the continuity of the drinking water supply and for the water supply during emergencies and disasters (**WSP Step 11**). As mentioned in WSP step 8, all drinking water companies had implemented security and contingency plans. An incident should be reported to ILT, and afterwards the RA should be revised and information given on how to prevent re-occurrence of this incident.

Coverage of WSP steps by the six legally required RA/RM approaches

The six RA/RM approaches described in this study cover different steps of the WSP approach which is shown in Figure 4.1. For all approaches (1 – 6) a team was assembled and hazards and hazardous events were identified to assess the risk. The system description was carried out in 5 RA/RM approaches. QMRA and Drinking water protection files covered most WSP steps, as they cover 10 and 11 WSP steps respectively.

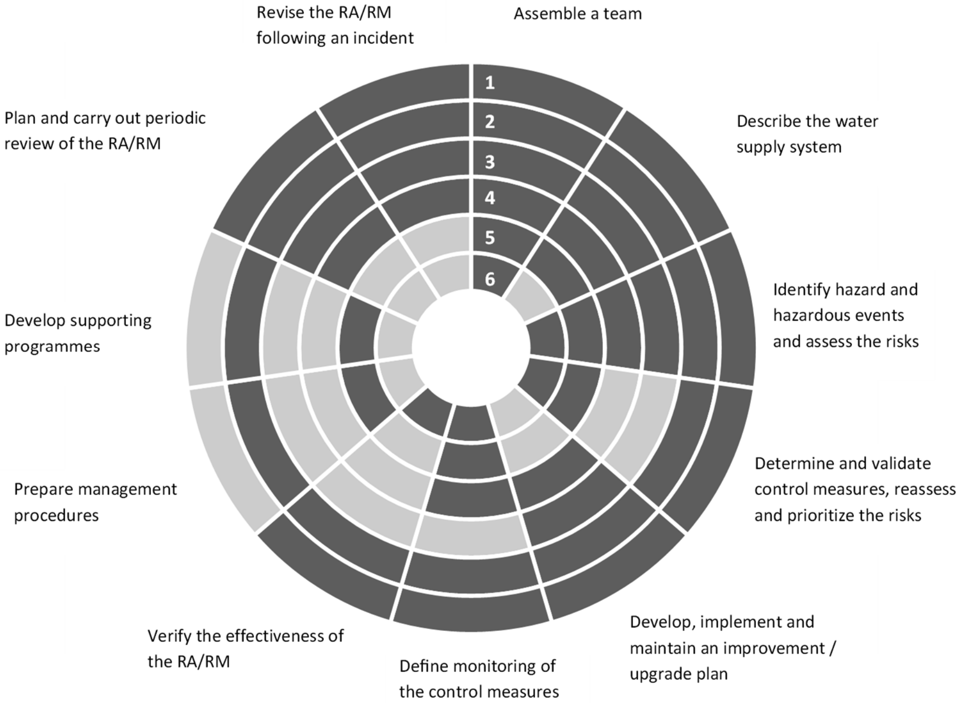

Figure 4.1. *Schematic overview of the WSP steps covered (dark gray) and not covered (light gray) by the six legally required RA/RM approaches represented per ring: 1. QMRA; 2. Drinking water protection files; 3. DRA; 4. Legionella prevention; 5. Code of hygienic practice for drinking water supply; 6. Monitoring program drinking water quality.*

Figure 4.1 shows that only one RA/RM approach covered all of the 11 WSP steps, and a combination of RA/RM approaches was needed to fully cover all WSP steps. Sector- and company specific RA/RM activities complemented the six legally required RA/RM approaches and covered the different steps as well. The six legally required RA/RM approaches contained advanced and detailed risk assessment methods, especially developed to generate more information on specific hazards or specific parts of the water supply system. Although all 11 WSP steps were covered with these six legally required RA/RM approaches, none of these approaches individually:

- identify microbiological, chemical as well as physical hazards;
- cover the complete drinking water supply chain; and
- are applicable to all drinking water supply systems.

This showed that multiple RA/RM approaches were needed to provide full information on all hazards and hazardous events from source to tap by combining the specific and detailed information gathered by the individual RA/RM approaches. By using multiple RA/RM approaches as described above the following challenges were observed during the inventory:

- sharing knowledge between the different RA/RM approaches due to the involvement of different teams;
- combining information due to multiple systems used for data collection in the different RA/RM approaches; and
- prioritizing risks based on different methodologies for assessing and rating the risks.

Discussion

Baum and Bartram stated that guidelines, regulations, tools and resources are elements of the enabling environment that encourage adaptation and implementation of WSPs in high-income countries (Baum and Bartram, 2018). In the Netherlands, legislation is available for multiple RA/RM approaches, but not specific for WSP as described by WHO (WHO 2004; Bartram *et al.*, 2009). While guidelines and regulations promote the uptake of risk management such as WSP, other conditions such as cultures and norms also influence risk management practices (Amjad *et al.*, 2016). The focus of the policy and the legislation in the Netherlands is on continuous drinking water supply of good quality. The focus of Dutch drinking water companies has always been on the quality of drinking water and continuous improvement of the water supplied. This has resulted in a variety of RA/RM methodologies used in the Netherlands, those legally required, sector specific and company specific ones. The drinking water companies continuously improve these different specific RA/RM methods by collaboration between drinking water companies, external experts and government. The monitoring program within QMRA is adapted to include worst-case scenario, so-called peak events. This is in line with recent development for risk-based monitoring (WHO 2015; EU 2015). Another example is including climate change scenarios in the DRA to identify all hazards and hazardous events that affect the quantity or quality of the water supply. This corresponds to climate resilience recently incorporated into the WSP (WHO 2017c).

The WSP approach is a very useful overarching approach for a systematic RA/RM from source to tap, to identify all hazards and hazardous events. For drinking water systems, risk assessment is an integral part of WSPs, and many different risk assessment methodologies from more simple to complex are available such as sanitary inspections, WSP risk matrix and QMRA. In the Netherlands, the six legally required RA/RM approaches contain advanced and detailed risk assessment methodologies. Combining all outcomes from these RA/RM approaches provides information on all hazards and hazardous events from source to tap and can be used as input for an overarching framework such as WSP. Comparable results were reported by Setty *et al*. (2019) showing that individual utility approaches need not be limited to one risk management programme as alternatives can be complementary. However, some challenges of using multiple RA/RM approaches were identified, compared to using a single approach, such as WSP. The first challenge of using six different RA/RM is combining and centralizing all identified risks and improvement plans from source to tap derived from the different RA/RM, e.g. using one centralized system or document. Using one RA/RM approach from source to tap, all information from source to tap is collected together. Another challenge is that the different risk assessments, used within one drinking water company, have many ways of weighing the risks, varying from quantitative risk analysis to expert opinion. It is important that drinking water companies can compare risk scores generated by different assessments to prioritize the most important hazardous events (based on severity and likelihood), instead of having separate risk outcomes. Within the WSP framework, not much guidance is given on how to include different assessment methods and how to prioritize. The European Standard EN 15975-2 is an appropriate option to provide such guidance, and incorporates fundamental elements of RA/RM (EN 2013). Compared to the WSP approach, different teams were involved in different RA/RM activities at Dutch drinking water companies, and therefore it is crucial to have an appointed responsible linking pin, as person or department, to share this information and harmonize how to use and interpret results from different risk assessments within one drinking water company. Another challenge is how to deal with all existing information and how to combine information between different systems, e.g. system description from source to tap. Traditionally, drinking water companies have separate pillars for abstraction, production and distribution, and therefore combining the system descriptions and monitoring data from these pillars would be an improvement for the drinking water companies. The obligation for risk based monitoring from source to tap is an important motivation for combining these pillars in the context of a risk based monitoring program.

Globally, the goal of the drinking water companies is to provide safe water, and therefore many steps of the WSP might be applied already even though the drinking water companies are not aware that they are carrying out parts of a WSP. An inventory, as shown in this study, of which steps of the WSP are already covered by the drinking water company is essential to show what has already been tackled and how, and what has not (completely) been tackled. As shown in this study, all steps of the WSP approach as described by WHO are covered by the legal requirements for RA/RM and even strengthened by sector and company specific RA/RM activities.

Of the countries that provided information on urban versus rural WSPs, 62% reported implementing WSPs in both urban and rural settings, reaffirming that WSP principles apply across all system types and sizes (WHO 2017b). Not all six legally required RA/RM approaches are applicable to all system types and sizes. For example, in the Netherlands QMRA is legally required for drinking water companies using surface water or vulnerable groundwater sources for the production of drinking water. However, the tool QMRAspot only supports drinking water companies using surface water. The Dutch drinking water companies, KWR and RIVM are investigating how QMRA can be achieved for drinking water companies using vulnerable groundwater sources. Furthermore, the six legally required RA/RM are too extensive and require too much expertise and resources to be applied for small water suppliers. For the 250 small supplies in the Netherlands, a more basic WSP can play an important role in improving water safety for small systems. WHO identified an important role for WSP also for improving water safety for small systems. Valuable resources have been developed to support WSP implementation for small systems (WHO 2017b). Nevertheless there remains a need for additional guidance materials and tools (WHO 2017b).

In the highly professionalized and knowledge intensive context of the Dutch drinking water sector there is a clear notion that continuous improvements can always be made. With the current developments within the WHO and the European Union (WHO 2015; EU 2017), water quality monitoring is moving towards risk based surveillance. This development holds the promise of increasing cost-effectiveness of monitoring and surveillance efforts without jeopardizing public health. Therefore, it is crucial that the ten Dutch drinking water companies perform risk analysis to provide evidence for adapting testing parameters. As shown in this study, several risk assessments are indeed performed to identify possible (future) hazards. Based on these assessments the water quality monitoring plan can be adapted to become even more risk based.

Conclusions

Providing safe drinking water requires a proactive and preventative RA/RM approach. Whereas the WHO recommends WSP as a RA/RM approach, Dutch drinking water companies use multiple RA/RM approaches, including different legally required RA/RM approaches. The six different RA/RM approaches are very specific and detailed, and focus on parts of the water supply. This study showed that these legal requirements, complemented by sector specific and company specific activities, cover all steps of the WSP. A long tradition of preventive risk management in the Netherlands, based on technical and theoretical insight, research and experience, has led to this combination of RA/RM approaches even before the WSP framework was developed. The six legally required RA/RM approaches provide advanced and detailed information on specific hazards and hazardous events in each part of the water supply chain. Therefore the outcomes need to be combined to provide information on all hazards and hazardous events from source to tap. Although the RA/RM in the Dutch drinking water sector is uniform, there are slight differences between individual companies. Using the various RA/RM approaches and subsequently combining and sharing all information (data) and systems is a challenge and a more harmonised approach could lead to improvements with respect to data sharing. The obligation of one WSP format seems to be too prescriptive for the current situation. However, generic arrangements for an integral RA/RM system would help to develop a more uniform and transparent approach to further improve current practices.

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Supplementary data

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Chapter 5

Legionella detection in wastewater treatment plants with increased risk for *Legionella* growth and emission

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Abstract

Legionnaires' disease (LD) is a severe pneumonia mainly caused by the bacterium *Legionella pneumophila*. Although many environmental sources of LD have been described, the sources of the majority of non-outbreak LD cases have not been identified. In several outbreaks in the Netherlands, wastewater treatment plants (WWTPs) were identified as the most likely source of infection. In this study, four criteria for *Legionella* growth and emission to air and surface waters were selected based on the literature and a risk matrix was drafted. An inventory was made of all WWTPs and their characteristics in the Netherlands. The risk matrix was applied to identify WWTPs at risk for *Legionella* growth and emission. Wastewater was collected at WWTPs with moderate to high risk for *Legionella* growth and emission. In 18% of the sampled WWTPs, *Legionella* spp. was detected using culture methods. The presented risk matrix can be used to assess the risks of *Legionella* growth and emission for WWTPs and support surveillance by prioritizing WWTPs. When *Legionella* is detected in the wastewater, it is recommended to take action to prevent emission to air or discharge on surface waters and, if possible, reduce the *Legionella* concentration.

Introduction

Legionnaires' disease (LD) is a severe pneumonia mainly caused by *Legionella pneumophila*. In the Netherlands, the LD incidence increased in recent years (Reukers *et al.*, 2019). *Legionella* bacteria can occur naturally in water and soil, often in low concentrations (Steinert *et al.*, 2002). Higher concentrations are sometimes observed in water installations, such as building water systems, wet cooling towers and whirlpools, because the temperature is more favorable for growth and competition from other bacteria is lower (Steinert *et al.*, 2002). Although many possible and confirmed environmental sources of LD have been described (Van Heijnsbergen *et al.*, 2015), a source of infections cannot be identified for the majority of sporadic (non-outbreak) cases. In the Netherlands, potential sources were systematically sampled as part of source-finding investigations, of which the majority of sampled sources were drinking water systems. Between 2002 and 2012, all potential sources of exposures were sampled for 392 non-outbreak patients from whom a clinical isolate was available, and the clinical and environmental isolates were compared using sequence-based typing. Only for 11% of these patients a genotypic match between the clinical isolate and the environmental isolate was found, showing that the source of infection remained unknown for the majority of patients, despite systematic source investigations. Moreover, there appears to be a mismatch in the Netherlands between the sequence types (STs) found in isolates from patients and environmental isolates from drinking water systems (Euser *et al.*, 2013; Den Boer *et al.*, 2015). The *L. pneumophila* serogroup 1 strain detected most frequently in sampled water systems was the strain ST1, but this strain was found in only a small portion (4–5%) of all clinical isolates. On the other hand, the ST-type of the *L. pneumophila* isolates found most frequently in patients (ST47) was not found in the sampled tap water systems at all (Euser *et al.*, 2013; Den Boer *et al.*, 2015). These data indicate that important sources of infections were not yet included in the source investigations and suggest that other sources of *Legionella* are present in the environment, that are causing LD.

In 2017, a wastewater treatment plant (WWTP) at a food processing company was identified as the likely source of contamination for a local increase of pneumonia caused by *L. pneumophila* observed (Loenenbach *et al.*, 2018). In this installation, high *Legionella* concentrations were detected in the wastewater of an identical ST as found in five patients: *L. pneumophila* serogroup 1 ST1646. Another LD cluster with 54 patients could be linked to an industrial WWTP located at a rendering company (Loenenbach *et al.*, 2018; Reukers *et al.*, 2018). Both WWTPs had a biological process which treated nutrient-rich process water at temperatures between 30 and 35 °C (Loenenbach *et al.*, 2018). Other studies also reported LD in employees or local residents in which WWTPs may have played a role

(Nguyen *et al.*, 2006; Nygård *et al.*, 2008; Kusnetsov *et al.*, 2010; Olsen *et al.*, 2010; Maisa *et al.*, 2015; Nogueira *et al.*, 2016).

In several studies, *Legionella* was detected in wastewater samples using culture and/or molecular methods (Allestam *et al.*, 2006; Blatny *et al.*, 2008; Schalk *et al.*, 2012; Lund *et al.*, 2014; Loenenbach *et al.*, 2018). High concentrations (up to 10^9 colony forming units per liter (CFU/L)) of *L. pneumophila* have been detected in the aeration tanks of industrial WWTPs (iWWTPs; Olsen *et al.*, 2010; Loenenbach *et al.*, 2018). During the wastewater treatment process, aerosols are formed, which may disperse *Legionella* from WWTPs to the environment. In air samples at WWTPs *Legionella* bacteria were found, with concentrations of up to 3,300 CFU/m^3 directly above aeration tanks (Medema *et al.*, 2004; Blatny *et al.*, 2008; Loenenbach *et al.*, 2018). Air measurements showed that aerosols contaminated with *Legionella* emitted from the aeration tanks can spread through the air over a distance of at least 3 km (Reukers *et al.*, 2018). Vermeulen *et al.* (2021) showed that exposure to aerosols from WWTPs likely caused LD in residents living near WWTPs. *Legionella* may also be discharged to surface waters through contaminated WWTP effluent (Olsen *et al.*, 2010; Nogueira *et al.*, 2016; Loenenbach *et al.*, 2018). In the Netherlands, an overview of all WWTPs is missing and no legal requirements are in place for monitoring *Legionella* at WWTPs. To better understand the contribution of WWTPs to LD more information on the amount, location and characteristics of WWTPs as well as the presence of *Legionella* in the installations is needed.

In this study, an inventory was made of WWTPs. Furthermore, we developed a risk matrix to identify and prioritize WWTPs in the Netherlands at risk for *Legionella* growth and emission. *Legionella* spp. and *L. pneumophila* were quantified in process water of moderate to high risk WWTPs employing culture methods.

Material and methods

Risk Matrix

In 2019, a literature study was performed to identify risk criteria associated with *Legionella* growth and emission from WWTPs (Bartels *et al.*, 2019). Peer-reviewed English-language articles describing WWTPs that were recognized as the direct or indirect source of *Legionella* infections were selected. In eight case studies, four similar characteristics were described that may have led to *Legionella* growth and emission to air and surface waters from WWTPs (Gregersen *et al.*, 1999; Isozumi *et al.*, 2005; Allestam *et al.*, 2006; Nguyen *et al.*, 2006; Blatny *et al.*, 2008; Kusnetsov *et al.*, 2010; Olsen *et al.*, 2010; Maisa *et al.*, 2015; Nogueira *et al.*, 2016; Loenenbach *et al.*, 2018). These characteristics were considered criteria for increased risk, as listed below.

Type of WWTP (biological or non-biological)

All WWTPs described in the case studies were biological WWTPs using aerobic bacteria for wastewater treatment. These bacteria have similar requirements for optimal growth as *Legionella* bacteria, including oxygen demand (via aeration) and temperature. *Legionella* growth is not inhibited or outcompeted by these bacteria (Caicedo *et al.*, 2019). Non-biological WWTPs were not described in the literature as a source of LD.

Type of industry

Relevant industries for *Legionella* growth are industries where many organic compounds such as proteins, and nutrients such as phosphorus and ammonia are discharged into the wastewater. This organic/nutrient-rich wastewater is beneficial for *Legionella* growth, and high *Legionella* concentrations have been detected in this water (Caicedo *et al.*, 2019). The following industries with organic/nutrient-rich wastewater were linked to patients with legionellosis:

- food industry, including meat processing and a brewery (Nogueira *et al.,* 2016; Loenenbach *et al.,* 2018);
- paper and wood industry (Allestam *et al.,* 2006; Olsen *et al.,* 2010);
- rendering companies (processing cadavers) (Loenenbach *et al.,* 2018; Reukers *et al.,* 2018); and
- petrochemical companies (Nguyen *et al.*, 2006).

These types of industries were included in the risk matrix (Table 5.1). The occurrence or growth of *Legionella* in treatment plants from other industrial sectors cannot be excluded, including WWTPs receiving wastewater from other industries. However, no case studies for these industries were described at the time of this study, and therefore, the risks for *Legionella* growth and emission in these other industries are classified as ‘unknown’.

Temperature of the process water

L. pneumophila grows within a temperature range from approximately 25 to 45 °C with an optimum between 35 and 37 °C (Wadowsky *et al.*, 1985; Falkinham *et al.*, 2015). WWTPs that were directly or indirectly identified as the source of LD all had a wastewater temperature between 30 and 37 °C, mainly between 35 and 37 °C (Allestam *et al.*, 2006; Kusnetsov *et al.*, 2010; Olsen *et al.*, 2010; Loenenbach *et al.*, 2018). Based on these case studies, water temperatures between 30 and 38 °C were considered as high risk for *Legionella* growth. For water temperatures between 25 and 29 °C and between 39 and 45 °C, the risk was categorized as moderate. When the water temperature at a WWTP is always below 25 °C, *L. pneumophila* growth to high concentrations is unlikely (low risk). However, WWTPs with water temperatures below 25 °C may receive wastewater with high *Legionella* concentrations (influent). Furthermore, some other *Legionella* spp. besides *L. pneumophila* may be able to multiply at these low temperatures. For water temperatures higher than 45 °C, *Legionella* growth was categorized as possible. These temperature ranges were included in Table 5.1 as well as their indicative risk level.

Table 5.1. *Risk matrix for Legionella growth and emission from WWTPs in the Netherlands.*

Type of WWTP	**Type of industry from which wastewater is treated**	**Temperature process water**	**Aeration**	**Risk category for emission to air**	**Risk category for emission to surface waters**
Biological	• Food industry • Paper and wood • Rendering companies • Petrochemical • Communal WWTP[a]	30–38 °C	Yes	High	High
			No	Moderate	
		25–29 °C or 39–45 °C	Yes	Moderate	Moderate
			No	Low	
		<25 °C or >45°C	Yes	Low	Low
			No	Very low	
	Other industries[b]	25–45 °C	Yes	Unknown	Unknown
			No	Unknown	
		<25 °C or >45°C	Yes	Unknown	Unknown
			No	Unknown	
Non-biological	Not relevant	Not relevant	Not relevant	Very low	Very low

[a] In WWTPs without an elevated water temperature (>25 °C), very high concentrations of *Legionella* may present when wastewater is received from industries with high concentration of *Legionella* (influent).

[b] No case studies were available for other industries and therefore the risk is unknown. However, it cannot be ruled out that other industries with nutrient-rich influent might have an increased risk.

Aeration

Aeration of wastewater in aeration basins plays a role in spreading *Legionella* as described in multiple articles (Olsen *et al.*, 2010; Loenenbach *et al.*, 2018; Caicedo *et al.*, 2019) and therefore included as a risk criterium. Aerosols formed through aeration allow *Legionella* bacteria to spread over a distance of more than 3 km (Reukers *et al.*, 2018). Unfortunately, available literature did not yield sufficient information to differentiate the degree of risk of *Legionella* emission through the use of different types of aeration such as surface aeration or bubble diffusers.

Based on the four identified risk criteria, a risk matrix was developed for assessing the risk of *Legionella* growth and emission from WWTPs (Table 5.1). In this matrix, four risk categories were distinguished:

- *High risk*: High to very high *Legionella* concentrations in aeration tanks (≥10^6 CFU/L) and effluent (≥10^4 CFU/L) can be expected, as shown in the case studies. This may result in a high risk of exposure to *Legionella* bacteria if aerosols are emitted from the wastewater or (discharged) effluent.
- *Moderate risk*: (Temporary) *Legionella* growth to high concentrations is possible

depending on the conditions. There is a risk of exposure to *Legionella* bacteria if aerosols are formed and emitted from wastewater or effluent.

- *Low risk*: *Legionella* may be present, but a high concentration ($\geq 10^6$ CFU/L) is not expected under these conditions. Incidentally, the concentration of *Legionella* in the process water might be increased, due to influent water with high concentrations of *Legionella*. Possible risk of exposure if aerosols are emitted from wastewater or effluent.
- *Very low risk: Legionella* is not likely to be present or is present at a very low concentration. Very low risk of exposure if aerosols are emitted from the wastewater or effluent.

Risk assessment for *Legionella* growth and emission to air and surface waters from WWTPs

To assess the number of WWTPs that potentially pose a risk for *Legionella* growth and emission to air and surface waters, an overview of existing WWTPs and their characteristics was needed. In November 2018, the Association of environmental agencies in the Netherlands (Omgevingsdienst NL (ODNL)) asked all 29 regional environmental agencies (ODs) to provide an overview of all iWWTPs in their region. Based on a questionnaire, additional information was gathered about the WWTPs. The Foundation for Applied Water Research (STOWA) made an inventory of all communal WWTP (cWWTPs) managed by the Dutch Water boards.

Based on the information received, the risk matrix was used to classify the WWTPs into four risk categories for *Legionella* growth and emission to air and discharge on surface waters from WWTPs: high, moderate, low, and very low risk. These risk levels do not provide information on the risk of infection or illness caused by *Legionella*.

Detection of *Legionella*

Sampling locations and sampling procedure

To verify the presence of *Legionella* in WWTPs with moderate to high risk of *Legionella* growth and emission, these WWTPs were sampled. Water samples were taken from the selected WWTPs in June and July 2019. Samples were taken at one time, so-called grab samples, according to the procedures described in ISO 19458:2006, if possible directly from the aeration tank or using a tap (ISO 2006). Water temperature (PT100 thermometer, Hanna Instruments) was measured on-site, in all samples. After sampling, samples were directly placed on melting ice or cooling-elements, and transported to the laboratory for microbiological analysis within 24 h after sampling.

Microbiological analysis

The water samples (500 ml) were concentrated using membrane filtration. Filtration was done by vacuum filtration (550 bar) with the aid of a vacuum controller (Innotech Europe BV; Moergestel; NL). The scraping technique, as described in ISO 11731:2017 annex E, was used for the removal of the organisms from the membrane (ISO 2017). Residues were resuspended in 1 ml of sterile water. Due to the composition, not all samples could be concentrated. When this was the case, direct material was used. Of the suspension or direct material, 100 µl was inoculated without dilution and after a 10-fold and 100-fold dilution on three different agar plates at 35 °C, with increased humidity. The three agar plates used were (i) buffered charcoal yeast extract (BCYE) supplemented with α-ketoglutarate and L-cysteine (BCYE-α L-cysteine), (ii) the antibiotics polymyxin B, cefazolin, and pimaricin (BCYE AB); and (iii) the antibiotics polymyxin B, anisomysin, and vanomycin (MWY) (Thermo Fisher Scientific, Cheshire, UK). To reduce the growth of other microorganisms than *Legionella*, which can interfere with the recovery, portions of the water samples were also subjected to heat treatment and acid treatment. Heat treatment was done by adding the sample (concentrated or not concentrated) to a sterile container and placing it in a water bath at (50 ± 1) °C for (30 ± 2) min. Acid treatment was done by diluting one volume of the sample (concentrated or not concentrated) with nine volumes of the acid solution as described in ISO 11731:2017 annex D, mixing well and leaving it for (5.0 ± 0.5) min (ISO 2017). The first examination was performed on day three of the total incubation period of 7 days using a dissection microscope. Suspected colonies were isolated and identified using MALDI-TOF. *L. pneumophila* strains were serotyped using commercially available kits containing antisera against *L. pneumophila* serogroups 1–14 according to manufacturer's protocol (*Legionella* latex test, Oxoid Limited, Hampshire, UK; *Legionella* antisera 'Seiken', Denka Seiken Co. Ltd, Tokyo, Japan).

Results and discussion

Risk assessment of WWTPs

In total, 451 iWWTPs were identified by the ODs as competent authority. The number of iWWTPs listed per environmental agency varied between 0 and 74 implying that not all iWWTPs were identified in some regions. Some environmental agencies only reported iWWTPs with biological treatment, which indicates there might be underreporting of the number of non-biological treatment iWWTPs. In addition to the iWWTPs, a total of 327 cWWTPs owned and managed by the Dutch Water Authorities were reported by STOWA. In contrast to iWWTPs, for cWWTPs, an up-to-date database was available. In total, 778 WWTPs were identified as shown in Figure 5.1.

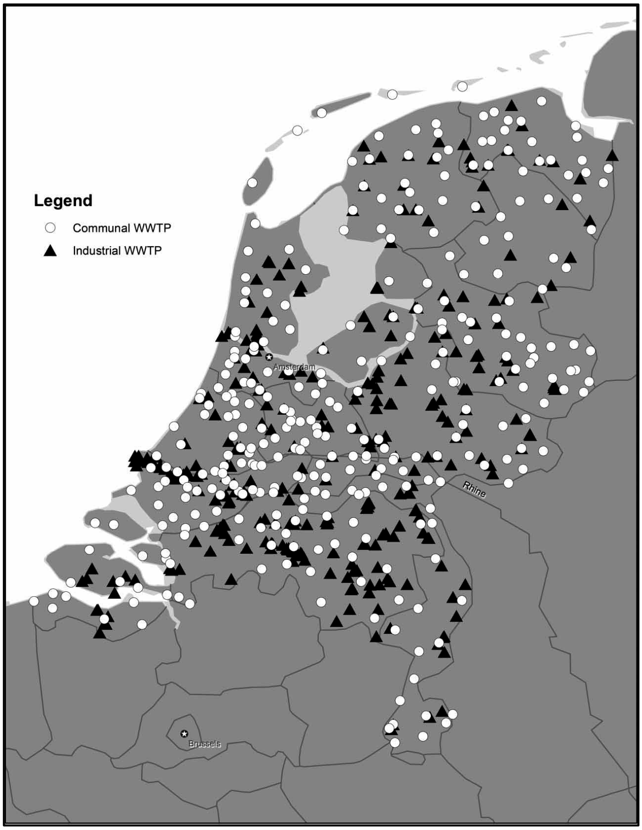

Figure 5.1. *Overview of communal (white dots) and industrial (black triangles) WWTPs in the Netherlands, based on the inventory.*

A risk assessment was done for all 778 WWTPs using our newly developed risk matrix (Table 5.1), based on the four risk criteria for *Legionella* growth and emission to air or effluent. A biological treatment process was part of all 327 cWWTPs and they belonged to the type of industries with organic/nutrient-rich wastewater. Aeration was used in all 327 cWWTPs during biological (aerobic) treatment. The temperature of the process water of 315 cWWTPs (96%) depended on the ambient temperature and varied between 8 and 20 °C and these were identified as 'low' risk. Twelve cWWTPs (4%) (partly) operated between 30 and 38 °C for optimal operation in the conversion of ammonium and nitrite into nitrogen gas using Anammox bacteria. These twelve cWWTPs were classified as 'high' risk (Table 5.2).

For iWWTPs, 219 out of 451 (49%) had a biological treatment process. The risk of the 232 non-biological iWWTPs was categorized as 'very low'. There were 90 (41%) biological iWWTPs that received wastewater from other industries. Insufficient information about the composition of the influent was available and no case study for these industries was described. Therefore, no risk could be estimated and these 90 iWWTPs were classified as ‘unknown’ risk. More than half of the biological iWWTPs (59%, n = 129) belonged to the selected industries with organic/nutrient-rich wastewater: food industry (n = 101), petrochemical companies (n = 10), wood and paper industry (n = 9), rendering companies (n = 1), and (partly) communal wastewater (n = 8). For three out of these 129 iWWTPs, the risk for *Legionella* growth and emission could not be assessed (unknown) as the temperature of the process water was not reported. The risk for *Legionella* growth and emission was categorized as ‘low’ for 45 out of these 129 iWWTPs with a temperature of the process water not in the range of 25–45 °C. For 81 out of these 129 iWWTPs, the temperature of the process water was listed between 25 and 45 °C. For 18 iWWTPs, the temperature of the process water range was reported to be completely within the range of 30–38 °C, and therefore, the risk for *Legionella* growth and emission was categorized as ‘high’. For the remaining 63 iWWTPs with a biological treatment process, the water temperature was reported as between 25 and 45 °C or only indicated as elevated temperature. Therefore, the risk was estimated as ‘moderate to high’ (Table 5.2).

For some locations, the requested information was not specified, incomplete or not correct. At some locations, for example, the reported temperature of the process from the inventory differed from the measured water temperature during sampling. If data is not correctly reported, this might have a negative effect on the risk assessment and WWTPs with a ‘moderate’ to ‘high’ risk might be assessed lower. Compared to cWWTPs for iWWTPs, it was more complicated to collect this information as a central registration of iWWTPs does not exist. To provide more information on the role of *Legionella* growth and emission from WWTPs and to assess the risk, an accurate overview of all WWTPs and their characteristics should be made available and kept up-to-date.

Table 5.2. *Overview of the number of WWTPs in the Netherlands with the risk of Legionella growth and emission to air or surface waters based on the reported characteristics in the inventory. Preventive measures to prevent the emission of Legionella were not taken into account.*

Risk of *Legionella* growth and emission to air and/or surface waters	**Number of WWTPs**	
	Communal	Industrial
High	12	18
Moderate to high	0	63
Low	315	45
Very low	0	232
Unknown: missing information or no case studies available	0	93

Detection of *Legionella*

Together with STOWA, the Dutch Water Authorities responsible for all cWWTPs developed voluntarily their own sampling strategy for high risk cWWTPs and were not included in this study. The *Legionella* detection focused on iWWTP classified as 'moderate to high' or 'high' risk (Table 5.2). In total, 81 iWWTPs were selected for sampling and analysis of which 63 iWWTPs with 'moderate to high' risk and 18 iWWTPs with 'high' risk. At the request of the environmental agencies, eleven iWWTPs were added to the final selection ending up with 92 iWWTPs. Finally, 85 of the 92 iWWTPs were sampled and analyzed for the detection of *Legionella* (Table 5.3). At seven iWWTPs, no samples could be taken due to maintenance, miscommunication, or problems during sampling.

During sampling, the water temperature sometimes differed from the reported information at the inventory. The water temperature of eight iWWTPs during sampling was below 25 °C compared with the reported temperature range of 25–40 °C, resulting in a lower risk than expected. One iWWTP that was added at the request of the environmental agency, had a temperature of 33 °C, but had been reported as below 25 °C in the inventory and was therefore originally not included in the selection of high risk iWWTPs. iWWTPs were categorized based on the registered temperature of the water during sampling and was used for assessing the risk level (Table 5.3).

In this study, we found *Legionella* spp*.* 15 iWWTPs (18%). At 13 locations (15%), *L. pneumophila* was detected and at two locations *Legionella* non-*pneumophila* was found of which at one location it was determined as *L. bozemanii* (Table 5.3). Lund *et al.* (2014) found comparable results: 21 out of 130 analyses (16%) from iWWTPs samples were positive for *Legionella* spp*.* and 12 (9%) were positive for *L. pneumophila* (Lund *et al.,* 2014). The concentrations for *Legionella* varied between 1 x 10^5 and 3 x 10^8 CFU/L which was similar to concentrations found in other studies (Olsen *et al.*, 2010; Loenenbach *et al.*, 2018).

Serotyping of *L. pneumophila* showed a diversity of serogroups and in five iWWTPs serogroup 1 was found (Table 5.3). *L. pneumophila* serogroup 1 is the causative agent for most of the patients with diagnosed LD in the Netherlands (Reukers *et al.*, 2019). Sequence typing of *L. pneumophila* serogroup 1 gave the following STs: ST47, ST474, ST1095, and ST1646. One iWWTP sample contained both ST1095 and ST1646. Notable, one *L. pneumophila* serogroup 1 isolate was ST-type 47. This virulent strain is found in 41% of the clinical isolates of Dutch non-travel-associated patients (Den Boer *et al.*, 2015). Although this ST-type has also been detected in spa pools and soil (Schalk *et al.*, 2014), it was not found in other water samples during ten years of systematic sampling of potential sources (Den Boer *et al.*, 2015).

Table 5.3. *Overview of the results of Legionella detection in iWWTPs categorized on the risk level. The number of sampled iWWTPs is given per industry and the number of Legionella positive iWWTPs is shown between brackets. For the Legionella positive iWWTPs, the species including serogroup is given per positive iWWTP. L.p = Legionella pneumophila and sg = serogroup.*

Risk level	**Measured temperature of process water (°C)**	**Number iWWTPs sampled (*Legionella* positive iWWTP) per type of industry**						
		Food industry	Petro-chemical company	Wood- and paper industry	Rendering company	Non-specific WWTP	Sludge and manure	Total
Low	<25	9 (0)	1 (0)					10 (0)
Moderate	25–29	13 (1) 1: L.p sg5	3 (1) 1: L. spp	2 (2) 1: L.p 7–14 2: L.p 7–14		3 (1) 1: L.p sg1		21 (5)
High	30–38	32 (3) 1: L.p sg1 and L.p sg2 2: L.p sg3 and L.p sg5 3: L.p sg2	5 (2) 1: L.p sg1 and L.p sg5 2: L.p sg5	6 (2) 1: L.p sg1 2: L.p sg6	1 (1) 1: L.p sg1	3 (0)		47 (8)
Moderate	39–45	3 (0)						3 (0)
Unknown	30–38					1 (0)	2 (2) 1:.*L. bozemanii* 2: L.p sg3 and L.p sg6	3 (2)
Total		57 (4)	9 (3)	8 (4)	1 (1)	8 (1)	2 (2)	85 (15)

The positive *Legionella* results were directly communicated to the responsible OD for follow-up with the owner of the iWWTP and investigated if additional control measures are required to protect public health. When *Legionella* is detected in the wastewater, it is recommended to take action to prevent emission to air or discharge on surface waters and, if possible, reduce the *Legionella* concentration. The first step is treating biological systems for selectively reducing *Legionella* without interfering with the biological process in the system, such as adding an ‘Expanded Granular Sludge Blanket’ reactor (Nogueira *et al.*, 2016). If reduction of *Legionella* is not possible, then minimize the emission of *Legionella* from the WWTP. To reduce emission from WWTPs to air, the aeration basins could be covered with a tarpaulin or floating balls although the effect depends on how well the surface is covered (Lodder *et al.*, 2019). When the air was extracted from closed aeration basins and subsequently filtered and disinfected with UV radiation the number of *Legionella* bacteria in the air decreased dramatically (Lodder *et al.*, 2019). An additional treatment of effluent water, such as (membrane) filtration or UV treatment, could be applied to improve the water quality of the receiving surface waters (Collivignarelli *et al.*, 2018). ISO 11731: 2017 is often used for determining the number of *Legionella* bacteria in water, but when applied to wastewater, this method can give a wide variation of results (ISO 2017). Due to high concentration of other (interfering) microorganisms and the high detection limit for *Legionella* (approximately 10,000 CFU/L), it is rather difficult to detect *Legionella* bacteria in highly polluted wastewater. The sample pre-treatment conditions for wastewater are harsh and can affect *Legionella*'s cultivability (Whiley and Taylor, 2016). Furthermore, culture-based methods such as ISO 11731 are not able to detect *Legionella* in the viable but non-culturable (VBNC) state (Whiley and Taylor, 2016; Caicedo *et al.*, 2019). This leads to an underestimation of the real *Legionella* concentration in the sample (Whiley and Taylor, 2016). *Legionella* bacteria in the VBNC state can grow under the correct conditions. If *Legionella* cannot be detected in a wastewater sample with the used method, this means that no viable *Legionella* or less than 10,000 CFU/L *Legionella* bacteria are present in the sample. The iWWTPs in our study were sampled only once by taking a grab sample in the months of June and July. It is unknown how many WWTPs false negative results were obtained because of the high detection limit or because of other factors like the time period when samples were taken. The consequence of false negative results can be that no control measures are taken or control measures might appear more effective than they really are (Caicedo *et al.*, 2019). Using molecular techniques, *L. pneumophila* and *Legionella* spp. are more frequently detected as compared with culture methods (Medema *et al.*, 2004; Lund *et al.*, 2014). Lund *et al.* (2014) investigated various industrial and communal WWTPs for the presence of *Legionella* and found that most samples (99%) were *Legionella* spp. positive by PCR, whereas *Legionella* could be detected less frequently (16%) using culture methods (Lund *et al.*, 2014). We found similar results in our study (data not shown). The benefits of PCR are rapidly available results and the detection of *Legionella* bacteria in the VBNC state. The PCR method does not differentiate between alive or dead *Legionella* bacteria, and therefore, no information is gathered on the viable *Legionella* bacteria present (Caicedo *et al.*, 2019). However, based on the limitations of the culture method for wastewater and the high detection rates with molecular techniques, the number of *Legionella* positive iWWTPs in this study is expected to be underestimated.

The majority of sampled iWWTPs (67%) processed wastewater from food industries whereas only one iWWTP from a rendering company was sampled. In all different types of industry, at least one iWWTP tested positive for *Legionella* varying between 7 and 100% *Legionella* positive iWWTPs per industry, which is in line with other published studies (Blatny *et al.*, 2008; Kusnetsov *et al.*, 2010; Loenenbach *et al.*, 2018; Caicedo *et al.*, 2019).

In addition to industries with organic/nutrient- rich process water as described in the literature, other industries may also produce nutrient-rich wastewater, which promotes *Legionella* growth (Caicedo *et al.*, 2019). In this study, two industries treating manure and sludge were *Legionella* positive but were not included as high risk industries. Furthermore, industries without nutrient-rich wastewater may sometimes receive nutrient-rich wastewater from other companies for processing in their WWTP. This may be the case for both cWWTPs and iWWTPs. Therefore, it is recommended to assess the risk for other industries, especially if nutrient-rich water is expected. If other industries should be included the risk matrix needs to be adapted.

The process water of the 15 *Legionella* positive samples had a temperature between 25 and 29 °C (n = 5) or 30 and 38 °C (n = 10). Similar to other studies, all *Legionella* positive samples were derived from iWWTP with process water temperatures between 25 and 38 °C (Blatny *et al.*, 2008; Kusnetsov *et al.*, 2010; Maisa *et al.*, 2015; Loenenbach *et al.*, 2018). This indicates that the selected range is an important criterion with increased risk. The percentages of *Legionella* positive iWWTPs with water between 25–29 °C and 30–38 °C were respectively 24 and 17%. Ten iWWTPs with temperatures of the process water below 25 °C and three iWWTPs with a temperature of the process water above 39 °C tested negative for the detection of *Legionella* spp. (Table 5.3). Based on the findings, we would suggest to divide the process water temperature below and above 25 °C. Furthermore, if WWTPs with a water temperature below 25 °C receive wastewater with high *Legionella* concentrations this may also result in high concentrations in the aeration tanks, although growth will be limited. *Legionella* growth does not only depend on the water temperature. There may also be circumstances where the temperature is stratified in the basin or changes temporarily, so the concentration could increase or decrease over time (Caicedo *et al.*, 2016). The risk matrix for assessing the risk of *Legionella* growth and emission from WWTPs was based on available information in the literature. New data might lead to an addition to or change in risk criteria.

For WWTPs with 'moderate' to 'high' risk of *Legionella* growth and emission, it is needed to check the existing measures in place. Existing control measures might contribute to a lower risk of *Legionella* growth or reduce the concentration of *Legionella* are a prolonged anaerobic step or treating effluent with an extra purification step, e.g. UV or additive biocides (Nogueira *et al.*, 2016; Collivignarelli *et al.*, 2018). Control measures to reduce the emission of *Legionella* include covering the aeration tank with concrete, tarpaulin or balls, and use of pure oxygen instead of normal aeration (Caicedo *et al.*, 2019). These control measures aim to limit or prevent the emission of *Legionella*. However, when the high concentration of *Legionella* in the WWTP remains and in case of a hazardous event, it may still spread to the environment. When treated effluent or surface waters receiving treated effluent containing *Legionella* is used in cooling towers or for other activities, aerosols with *Legionella* can be spread (Nogueira *et al.*, 2016). Due to water scarcity re-use of treated wastewater becomes an important alternative water source. For aerosol-forming applications such as irrigation and cooling towers, the presence of *Legionella* in treated wastewater might pose a health risk (Caicedo *et al.*, 2019). Regulation of *Legionella* in effluent water used for application with a risk of aerosolization is needed to protect human health, such as the requirement of a maximum of 1,000 *Legionella* CFU/L in the EU regulation (EU 2022).

For high risk WWTPs, it is recommended that both operational monitoring of the WWTP is initiated and a management plan is developed. More research on control measures is needed to identify how *Legionella* growth and emission could be controlled. For existing WWTPs, it is recommended to adjust the treatment process so that *Legionella* growth is prevented or at least controlled as much as possible similar to *Legionella* prevention in wet cooling towers. If *Legionella* is present in high numbers it is recommended to reduce the concentration and avoid emission of *Legionella*, especially via air. A sustainable solution for new WWTPs is to design a process that limits the growth and/or emission of *Legionella*. To support owners of WWTPs to identify, interpret, and control *Legionella* risks, a guidance document was drafted using the findings of this study (Oesterholt and Hollebekkers, 2022). To improve the risk assessment for the emission of *Legionella* to air or surface water from WWTPs, existing measures and their local application to prevent the emission of aerosols should be taken into account.

Conclusion

Biological WWTPs are possible sources of LD and need to be considered during source investigation. In this paper, a risk assessment methodology was drafted to identify WWTPs at risk of *Legionella* growth and emission. The risk matrix is based on information about the WWTPs that is relatively easy to identify, and therefore, this approach is likely applicable for many countries to identify possible sources for *Legionella* emission from WWTPs. To provide more information on the role of *Legionella* growth and emission from WWTPs and to assess the risk, an accurate overview of all WWTPs and their characteristics should be made available and kept up-to-date.

In this study, 18% of the iWWTPs with moderate to high risk for growth and emission of *Legionella* were *Legionella* spp. positive, and two iWWTPs with unknown risks were positive. The true proportion of *Legionella* positive WWTPs may be even higher, as the culture method does not detect low concentrations of *Legionella* in wastewater. Based on the risk assessment of WWTPs and *Legionella* spp. detected with culture methods revising the risk matrix based on new scientific evidence and information on *Legionella* outbreaks is needed. Additional research is needed to further improve the risk matrix. Nevertheless, the current risk matrix can be used to develop a risk-based monitoring program starting at WWTPs with the highest risk of *Legionella* emission. For WWTPs with a high risk, it is recommended that both operational monitoring of the WWTP is initiated and a sampling plan is drawn up and implemented, a so-called management plan. For assessing the risks for WWTPs, it is crucial that the location of such installations is known, therefore registration of WWTPs is recommended. For WWTPs still to be built or when new treatment technologies are installed, it is recommended to guarantee that *Legionella* growth is prevented or at least controlled as much as possible.

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Chapter 6

Linking water quality monitoring and climate-resilient water safety planning in two urban drinking water utilities in Ethiopia

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Abstract

Unsafe drinking water is a recognised health threat in Ethiopia, and climate change, rapid population growth, urbanization and agricultural practices put intense pressure on availability and quality of water. Climate change-related health problems due to floods and water-borne diseases are increasing. With increasing insight into impacts of climate change and urbanization on water availability and quality and of required adaptations, a shift towards climate resilient water safety planning was introduced into an Ethiopian strategy and guidance document to guarantee safe drinking water. Climate resilient water safety planning was implemented in the urban water supplies of Addis Ababa and Adama providing drinking water to five million and 500,000 people respectively. Based on the risks identified with climate resilient water safety planning, water quality monitoring can be optimized by prioritizing parameters and events which pose a higher risk for contaminating the drinking water. Water quality monitoring was improved at both drinking water utilities and at the Public Health Institute to provide relevant data used as input for climate resilient water safety planning. By continuously linking water quality monitoring and climate resilient water safety planning, utilization of information was optimized, and both approaches benefit from linking these activities.

Introduction

Safe and readily available drinking water is important for public health and was declared a UN human right in 2010 (UN 2010). In 2015, 2.1 billion people still lacked safely managed drinking water services of which 582 million people abstracted water from non-improved and unprotected sources worldwide (WHO and UNICEF, 2017). Drinking unsafe water may cause exposure to pathogens, which can result in waterborne diseases such as cholera, gastroenteritis and hepatitis E (Howard and Bartram, 2003). Although considerable improvements have been achieved, poor drinking water quality still is a recognized health threat in Ethiopia since a considerable burden of disease originates from unsafe water. In Ethiopia, the mortality rate attributed to unsafe water, sanitation and hygiene services is 43.7 per 100,000 persons (WHO 2018), and the average Ethiopian child suffers five to twelve diarrheal episodes yearly as a result of poor drinking water and poor environmental sanitation (MoH 2011). An outbreak of acute watery diarrhoea was reported in nine regions of Ethiopia between January and December 2017 (WHO Afro 2018). Another outbreak with 1,117 suspected cases of Hepatitis E and 21 deaths was reported among refugees residing in the Gambella region from April 2014 to January 2015 (Browne *et al.*, 2015). Adane *et al.* (2017) concluded that acute diarrhea among children under five years of age in slums of Addis Ababa can be reduced by continuously available piped water supplies and education of urban caregivers.

The importance of water, sanitation and hygiene for development, poverty reduction and health had previously been recognized in amongst others the United Nations Millennium Declaration and is reflected in Sustainable Development Goal (SDG) 6.1 of the UN declaration *Transforming our world: the 2030 Agenda for Sustainable Development* (UN 2015) which calls for achieving universal and equitable access to safe and affordable drinking water for all by 2030. The UNICEF/WHO Joint Monitoring Programme update and SDG Baseline report on drinking water, sanitation and hygiene (WHO and UNICEF, 2017) showed that in Ethiopia, the proportion of the population with access to an improved water supply was increased from 58% to 75% between 2000 and 2015, with an increase of the population with access to safely managed supplies from 5% to 11% in the same timeframe (WHO and UNICEF, 2017).

The frequency and intensity of extreme weather events with possible consequences for drinking water safety are increasing due to climate change (Depla *et al.*, 2009; Seneviratne, 2012). In the context of climate change, a degradation trend of drinking water quality leads to an increase of at-risk situations leading to potential health impacts (Depla *et al.*, 2009). Eastern Africa is one of the most vulnerable regions with respect to the impacts of climate change (Samson *et al.*, 2011), and in Ethiopia climate change-related health problems, such as mortality and morbidity due to droughts, floods and waterborne diseases are increasing (Simane *et al.*, 2016). The effects of climate change will continue to magnify without the right adaptation and mitigation measures. Besides climate change, rapid population growth, urbanization and inappropriate farming put intense pressure on available water (Simane *et al.*, 2016).

The WHO established a 'Framework for Safe Drinking-water' which encompasses setting health-based targets, a risk assessment and risk management approach and a system of independent surveillance (WHO 2011). A so-called water safety plan (WSP) is the recommended risk assessment and risk management approach, including all steps in water supply from catchment to consumer. In 2017, the WSP approach was extended by also identifying and managing climate-related impacts from catchment to consumer (WHO 2017). In 2015, the Federal Ministry of Water, Irrigation and Energy published guidelines for climate resilient water safety plan (CR-WSP) implementation for urban utility managed piped drinking water supplies (MoWIE 2015). The steps for implementing CR-WSP are shown in Figure 6.1.

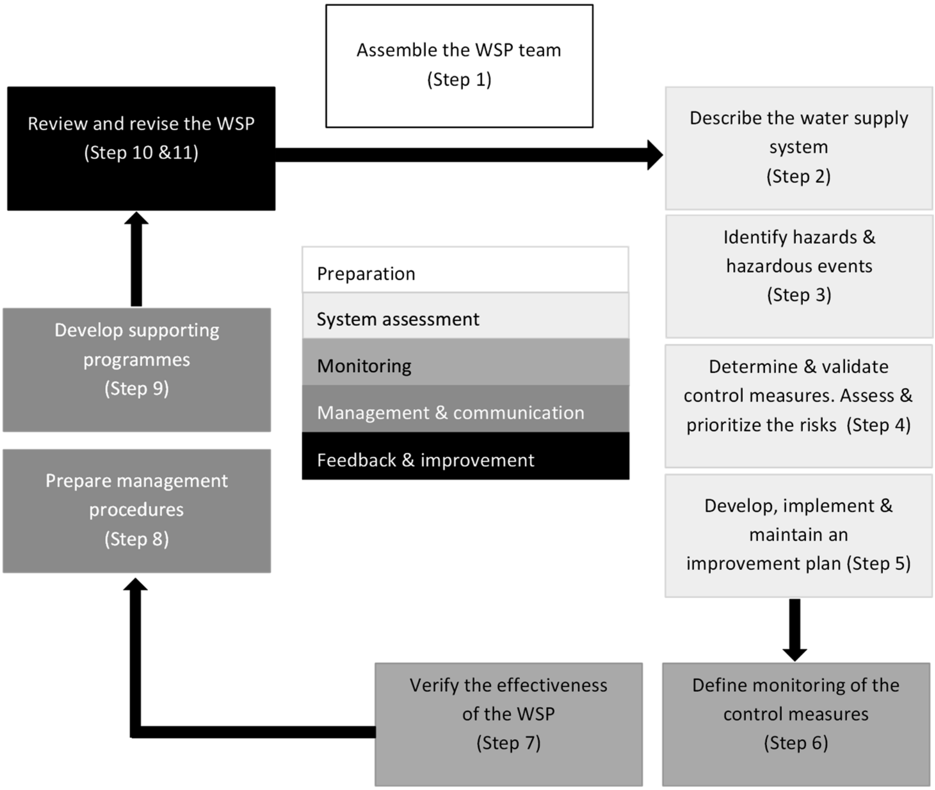

Figure 6.1. *Steps of CR-WSP (adapted from MoWIE, 2015).*

As part of the WSP approach, different monitoring should be applied to provide information on drinking water operations and drinking water quality:

- testing and monitoring of the quality of drinking water (verification monitoring) and source water used for drinking water production;
- monitoring to demonstrate the performance of control measures under normal and exceptional circumstances (validation monitoring); and
- monitoring operational parameters showing whether the processes in drinking water supplies are operated safely (operational monitoring).

These types of monitoring are key elements that contribute to drinking water safety, and thereby protecting public health (WHO and IWA, 2009).

In Ethiopia, the physical, chemical, and bacteriological requirements for drinking water are described in the Compulsory Ethiopian Standard for Drinking Water Specification (Ethiopian Standards Agency 2013). Ethiopian drinking water utilities are mandated to comply with the requirements of these standards for drinking water supply. The ‘Framework for Safe Drinking-water’ prescribes that surveillance should be carried out by an independent agency. The established regulatory arm of the Federal Ministry of Health which is Food, Medicine and Health Care Administration and Control has an Inspection and Surveillance directorate responsible for regulating water quality monitoring (MoH 2014).

The project Source to Tap and Back (S2TAB) started in 2013 in Ethiopia. The goals of the project were to establish:

- a stakeholder dialogue and capacity development approach for improved financial and environmental sustainability of drinking water services;
- improved control of wastewater discharges in the catchments;
- water resource protection;
- measures to reduce sedimentation in the catchment reservoirs;
- improved water supply and service delivery in two drinking water supplies;
- increased access to improved sanitation; and
- reduction of non-revenue water to reduce water loss.

Vitens Evides International (VEI) coordinated this Dutch – Ethiopian project, which involved four Ethiopian and five Dutch partners: Addis Ababa Water and Sewerage Authority (AAWSA); Adama Water Supply and Sewerage Enterprise (AWSSE); Oromia Water Mineral and Energy Bureau (OWMEB); Ethiopian Public Health Institute (EPHI); Vitens Evides International (VEI); Dutch Water Authority Vallei en Veluwe; Dutch Water Authority Zuiderzeeland; MetaMeta and Dutch National Institute for Public Health and the

Environment (RIVM). For the implementation of CR-WSP, the German Environment Agency (UBA) was subcontracted.

In this study, we focussed on the part of the project S2TAB to improve water supply and service delivery. The objectives of this part of the project were to implement CR-WSPs in the urban water supplies of Addis Ababa and Adama and to strengthen drinking water quality monitoring (WQM) at the laboratories of both drinking water utilities and EPHI. The study aims at showing how a strong link was established between CR-WSP and WQM and how these complementary approaches benefit from joined implementation to promote sustainable provision of safe drinking water in urban utilities.

Material and Methods

Drinking water utilities and laboratories

The Adama Water Supply and Sewerage Enterprise (AWSSE) is a company responsible for water and sewerage services in Adama. AWSSE provides water supply service to approximately 500,000 people. The company was established under the municipality of Adama in 1930 and under the Oromia Water Mines and Energy Bureau. AWSSE is governed by a board consisting of representatives from several government offices. AWSSE has one drinking water treatment plant, at the site of which the laboratory for testing drinking water is also located.

The Addis Ababa Water and Sewerage Authority (AAWSA) is responsible for providing water and sewerage services to Addis Ababa, the capital of Ethiopia. The service area (over five million people) is limited to the boundaries of Addis Ababa city administration. AAWSA was established in 1971 and is governed by a board consisting of representatives from several government agencies. AAWSA has two drinking water treatment plants located in Legedadi and Gefersa. The laboratory for testing drinking water is located at the head office of AAWSA.

EPHI, as technical body of the Federal Ministry of Health, is focusing on priority disease research and strengthening the national public health laboratory services in the country. This institute is located in Addis Ababa and conducts research on the causes and spread of diseases on food and water safety issues, and thereby supports activities for the improvement of health in the country. EPHI has knowledge and expertise on water quality testing, such as physicochemical and microbiological analyses. Previously EPHI conducted a rapid assessment of drinking water quality (Tadesse *et al.*, 2010), and carried out several water quality studies in Ethiopia. Some objectives of EPHI regarding drinking water are monitoring quality and safety of water and development of associated guidelines.

Baseline study

At the start of the project S2TAB, a baseline study was conducted to identify the awareness of WSP at both drinking water utilities and the level of WQM at the drinking water utilities and EPHI. During the baseline study, both drinking water utilities and EPHI were visited twice in 2013 by a team from RIVM, UBA, VEI and EPHI. During the baseline study, information was gathered on the roles and responsibilities of the organizations. Furthermore, the need for implementing WSPs and improving WQM was identified by interviewing staff from the water supplies and EPHI.

Implementation of climate-resilient water safety plans

Within the project, implementation of WSPs started in 2014 in both drinking water utilities, following the WSP manual (WHO and IWA, 2009). Within the implementation period, the WSP approach was amended to integrate a focus on climate resilience (Rickert *et al.*, 2019) according to the steps of CR-WSP as described in the Ethiopian Guidelines for Urban Utility Managed Piped Drinking Water Supplies (MoWIE 2015), see Figure 6.1.

External experts of the WHO Collaborating Centres at UBA and RIVM led the implementation of CR-WSP and supported the teams within the water utilities. As a local partner, EPHI joined the trainings for the implementation of CR-WSP to gain experience for future scaling up. For the implementation, a step-by-step approach was followed during the implementation period 2014 – 2018 with regular visits by the external experts. During these visits, nine trainings and consultation meetings were provided to facilitate WSP implementation within the teams of the utilities, and two annual workshops were conducted to exchange experiences, challenges and success factors between the drinking water utilities.

Strengthening water quality monitoring

At the national level, WQM was strengthened by training EPHI laboratory staff in pathogen detection, such as enterovirus, *Campylobacter* and *Cryptosporidium* in water. It is not mandatory to measure these pathogens in drinking water (Ethiopian Standards Agency 2013), but information of the presence of these pathogens in source waters used for the production of drinking water is of added value. The detection methods for these pathogens were based on molecular techniques. Training was conducted in a series of four trainings in the period 2013-2018 to prepare trainees for organising WQM and share knowledge with drinking water laboratories.

WQM was strengthened at the drinking water laboratories at both utilities by improving the laboratory infrastructure and providing necessary equipment. EPHI supported in purchasing materials, media and equipment from the local market. One training on WQM for both water utilities was organized by EPHI in 2015. This training focussed on the detection of core parameters, such as pH, water temperature, turbidity, free chlorine and *Escherichia coli*. Furthermore, trainings were given by RIVM at both laboratories to introduce and implement new methods, both for physicochemical and microbial testing, and to update existing procedures.

Results

Baseline study

The baseline study identified that both drinking water utilities and EPHI had no or only limited knowledge on WSP. WQM was in place at the laboratories of both drinking water utilities, and staff was well trained in the methods used. Nevertheless, there was a need to improve the methods and to test for additional parameters to show compliance with the national standards. The laboratories were differently equipped and both laboratories were in need of upgrading. At both drinking water utilities, monitoring data was collected and stored in logbooks, but data was not available electronically. Independence surveillance is limited to date. Although EPHI performs several analyses for WQM, the baseline study found that WQM at EPHI should be improved by detection of additional parameters, such as heavy metals, pesticides and pathogenic bacteria, viruses and parasites. Furthermore, applying molecular techniques for the detection of pathogens was identified as an advantageous improvement for the water quality testing at EPHI.

Kick-off workshops

Before starting the implementation of CR-WSP and improvement of WQM, a two-day workshop was held at each of the drinking water utilities in December 2014. The goal of these kick-off workshops was to create enthusiasm and support for the implementation. During the workshops, information was provided by RIVM and UBA on waterborne infectious diseases, WQM, CR-WSP and how CR-WSP and WQM complement each other. In total, 14 persons from the Adama water supply and 35 persons from the Addis Ababa water supply participated. Four persons from EPHI participated at both workshops to be able to support the further implementation of CR-WSP and WQM. For each water utility, a team was established for conducting the implementation of the CR-WSP and another team for improving WQM. In both utilities, there was an overlap of staff between the teams to create synergies between WQM and CR-WSP. Furthermore, each team of the water utilities had an EPHI representative assigned to support the activities.

Implementation of climate-resilient water safety plans

Nine trainings and consultation meetings on the steps of CR-WSPs were provided by UBA and RIVM from December 2014 to January 2018 to both water utilities. The training visits were used to facilitate and discuss CR-WSP implementation with the teams.

As the first task for the CR-WSP teams, a comprehensive system description was developed at each of the water utilities, based on all relevant existing documentation, and additional information was collected if needed. Based on this system description, both teams identified all hazardous events which could occur in their water supply system, potentially introducing hazards for public health. For this step, a generic list of hazardous events was used and adapted to reflect the situation in the respective water supplies (Rickert and Van den Berg, 2018). The teams also considered hazardous events related to climate conditions such as storm events, heavy rainfall or extended drought periods. For confirmation of the system description and hazardous events, field visits were conducted by the teams. During these field visits, new hazardous events were identified and documented. One example of such a new hazardous event was the identification of broken vent screens at storage reservoirs which may lead to contamination of the water through ingress of insects. For all identified hazardous events, the teams investigated whether control measures were already in place. For these existing control measures, the teams validated their effectiveness by providing evidence (e.g. monitoring data, literature data or reports of visual inspections). Afterwards, risks were assessed based on the likelihood and severity of each of the hazardous events, also taking into account the presence and effectiveness of the existing control measures. Both teams agreed on definitions for the likelihood categories (‘unlikely’, ‘possible’ and ‘most likely’) and severity categories (‘no or minor impact’, ‘moderate impact’ and ‘major impact’), as well as for the risk categories (‘low’, ‘medium’ and ‘high’ risk) to be applied for the risk assessment, which were based on the definitions included in the Ethiopian guidance for urban utilities (MoWIE 2015). These definitions and the risk matrix were translated into Amharic language. Based on a table, documenting all possible hazardous events and resulting risks for each of the utilities, the teams drafted improvement plans for risks identified as being high or medium, and gave an overview of all proposed actions to reduce the risks. The resulting improvement plans triggered both drinking water utilities to initiate some improvements immediately, such as

- creating awareness of hygienic work and train chemical flow workers and laboratory technicians;
- developing hygiene leaflets for drinking water production areas to create awareness of workers at the utilities and reservoirs, as well as of consumers collecting drinking water at public taps;
- conducting training on maintenance of plumber technicians and supervisors;
- updating the standard operating procedure (SOP) for sand filtration; and
- preparing and updating the operation manuals at the treatment plants.

Within the project, a budget was allocated for improvements achievable in a short period and at low cost (so-called ‘quick wins’), based on the improvement plans. Both teams selected some improvements from their respective improvement plans and presented these to the higher management. The following quick wins were initiated within project:

- relocating effluent discharge of dry-sludge bed further downstream;
- cleaning of dry-sludge bed;
- building fences around four public taps;
- renewing existing vent screens at storage reservoirs in the distribution system;
- installing manhole covers;
- installing chlorine mixers in reservoirs; and
- removing plants around a storage reservoir.

In addition to the quick wins and the achievements made by the drinking water utilities, further improvements were carried out within other parts of the project. Examples include:

- planting 12,000 indigenous trees in the catchment to reduce the amount of run-off and erosion (coordinated by MetaMeta);
- building 24 check dams in the catchments to reduce hazards from run-off entering the source water during rainfall (coordinated by MetaMeta and VEI); and
- improving sanitation in the catchment by construction toilets in schools and at market places (coordinated by MetaMeta and VEI).

Operational monitoring shows whether the control measures in place were working effectively. Based on the previously existing practices at the water utilities, both teams updated operational monitoring plans, including documentation of all activities already in place. Some examples of operational monitoring activities were:

- checking the infrastructure of a storage reservoir on a daily basis;
- controlling water in the clarifier (e.g. for algae) by visual inspection on a daily basis;
- measuring the filter depth of the sand filters every two months; and
- measuring the turbidity at the treatment effluent twice per day.

The teams also documented which actions should be undertaken when the monitored parameters exceed their critical limits as specified in the operational monitoring plan. At both drinking water utilities the effectiveness of WSP activities was verified using auditing and compliance monitoring, as well as information on customer satisfaction. Information gathered by WQM was applied to check whether drinking water complied with drinking water quality standards. Outcomes were fed into the CR-WSP to analyze effectiveness of implemented measures and provide information on relevant hazards. The emergency plans of both utilities were updated within the project. These include information on who should be contacted in case of an emergency, and describe several concrete emergency situations such as flooding, power disruption and bursting of chlorine gas storage cylinders. The Addis Ababa team developed separate emergency response plans for each of the utility's branch offices responsible for a specific distribution area, and documented SOPs in one book with instructions (operational manual for the production location and analysis book for the laboratory), rather than short documents to take into the field, laboratory or to the treatment plant. This team extracted the necessary information for SOPs for plumbers (branch officers) and operators (treatment plant) from the operational manual, to facilitate regular application when needed. In Adama, work instructions were available and the team updated the available work instructions and prepared additional SOPs.

At both water utilities, several supporting programs existed, such as creating awareness on pollution of source water among consumers and the population in the catchment (farmers and industries), providing information on websites, research, training and maintenance. The project increased the awareness raising:

- by sharing the utilities’ concerns about activities in the catchment, threatening the production of drinking water. Concerns were discussed with stakeholders, such as the water resource protection team established within the project.
- by preparing leaflets for public taps to provide information to the consumers how to safely collect and store water in a container and how to avoid contamination of the area around the public taps. The leaflets were provided in Amharic and in Oromian language.
- among stakeholders excluding the water utilities for the possible hazards and hazardous events for the production of safe drinking water, by the utilities joining a stakeholder platform established within the project, coordinated by the Dutch Water Authorities, MetaMeta and VEI.
- among people living in the catchment on improved sanitation coordinated by MetaMeta.
- with industries on emission reduction, based on data gathered within the project by the Dutch Water Authorities, MetaMeta and EPHI.
- by preparing leaflets for drinking water production areas and at storage reservoirs in the distribution network to provide information to the workers to work hygienically. The leaflets were produced in Amharic and in Oromian language.

Within the project S2TAB, additional training was conducted to improve the skills of employees, plumbers, laboratory staff and chemical workers.

The project provided a platform for exchange between stakeholders such as AAWSA, AWSSE, Oromia Water, Mines and Energy Bureau, basin authorities, environmental protection entities including practitioners, water resource managers, EPHI and climate scientists from Addis Ababa University to support decision-making based on knowledge of the complex water security trade-offs needed for resilience through providing a platform for a multi-stakeholder process. The CR-WSP teams developed a schedule for regularly revising and reviewing their CR-WSPs. Furthermore, both utilities planned to conduct a peer review between the two drinking water utilities to learn from each other.

Strengthening water quality monitoring

Within the project, WQM was strengthened at both utilities by harmonizing methods according to the Ethiopian drinking water legislation and by adding parameters based on possible risks identified from CR-WSP. Both utilities used different methods for the detection of the fecal indicator *Escherichia coli*, and rarely used membrane filtration for enumeration. Therefore, membrane filtration was introduced or improved respectively to harmonize the methods at both laboratories. In Addis Ababa, chemical analyses were already conducted regularly on samples, however in Adama, physicochemical analyses such as testing for turbidity, conductivity, fluoride, manganese, free chlorine and iron needed to be improved. At both laboratories, there was an interest in improving good laboratory practices and data analysis. Based on the needs' assessment, equipment, media and custom-made trainings were provided and the laboratories' infrastructure was upgraded.

In 2015, EPHI organized a centralized training that focused on basic WQM, including sampling, physicochemical analyses (temperature, pH, turbidity and free residual chlorine) and microbiological analyses (*E. coli* and coliforms) using membrane filtration techniques. The custom-made trainings followed up the centralized training and facilitated the implementation of the methods for basic WQM at both laboratories. In Adama, follow up trainings were conducted for implementing basic WQM using the supplied materials and trained methods. The basic WQM was extended by chemical analyses for 11 new parameters, such as fluoride, nitrate, manganese and phosphate within an additional training. In Addis Ababa, trainings were conducted that focused on strengthening the current WQM program and included both chemical and microbial WQM techniques, good laboratory practice, documentation and data analysis. At the laboratories of AAWSA and AWSSE, the staff developed 20 and 14 SOPs respectively for routine laboratory work and sampling. In between the trainings, RIVM and EPHI followed up with regular visits to identify challenges and needs regarding WQM.

At both drinking water utilities, the laboratories were upgraded with logistical support of VEI. In Addis Ababa, three different laboratories were present for chemical analysis, microbiological analysis and general laboratory work. These laboratories were outdated and were therefore renovated within the project. AWSSE had one laboratory for WQM, which was upgraded through renovation activities using the utility's own budget. Within the project, expansion of the laboratory with another room to separate chemical and microbiological analyses was initiated.

At EPHI, WQM was strengthened at the national level by introducing pathogen detection methods for enteroviruses, *Cryptosporidium*, *Giardia* and *Campylobacter* spp. and improving the detection of *Escherichia coli* as fecal indicator using membrane filtration in combination with chromogenic media. During annual practical and theoretical trainings by RIVM during the timeframe 2013-2018, participants became familiar with these techniques. At one of these training sessions, the drinking water utilities participated to create more awareness on pathogen (instead of indicator) detection. In the future, EPHI may disseminate these methods to different (regional) laboratories. Together with EPHI's department of food and water microbiology, a monitoring plan was developed and carried out for the detection of fecal contamination and pathogens in source waters in and around Addis Ababa. The information from this study may be used as an input for future CR-WSP revisions. Additional research, supported within S2TAB and conducted by EPHI, will provide information for the drinking water utilities, such as efficacy of water treatment during extreme weather events and the presence of antibiotic resistant bacteria in the source waters used for drinking water production.

Linking water safety planning and water quality monitoring

The project linked CR-WSP and WQM activities from the start. To guarantee that information from the laboratory reached the teams involved in the implementation of CR-WSP and vice versa, one or two persons from the laboratory were a member of the CR-WSP team. In this way, risks identified during the implementation of CR-WSP or requests for additional information on water quality could be addressed directly to the laboratory.

During the regular visits by the external experts, CR-WSP and WQM trainings were held back to back in order to stress the importance of linking these components. With combined sessions during the visits, information was shared between both teams of each water utility to increase awareness for CR-WSP and WQM. Three annual workshops were conducted to facilitate exchange of experiences, challenges and success factors between the drinking water utilities and EPHI.

As part of the CR-WSP, both drinking water laboratories reviewed and revised the existing WQM plan in place, also taking into account hazardous events relevant in their respective utilities. This resulted in adding parameters to future monitoring in raw water, such as fluoride in Adama and *Cryptosporidium* and *Giardia* in Addis Ababa. To harmonize management procedures, the WQM staff developed, in line with CR-WSP, SOPs for sampling, transport, chemical and microbiological analyses.

Discussion

Traditionally, both drinking water utilities addressed problems in the drinking water system, such as pipe breaks or customer complaints, mainly when they occurred. The WSP framework however is designed to proactively prevent hazards or hazardous events (WHO and IWA, 2009).

CR-WSPs were successfully implemented at both drinking water utilities. As part of the CR-WSP approach, WQM is applied to provide information on the operation of drinking water treatment processes and drinking water quality. One of the challenges for the implementation of WSP is a lack of laboratory facilities and methods for WQM (WHO and IWA, 2009; Rahman and Paul, 2011). Therefore, facilities and methods on WQM were also strengthened.

Challenges identified during implementation

One of the challenges during this process was the high turnover of staff, particularly at one of the utilities. This resulted in frequent changes in the WSP teams, which had a negative effect on sustainable implementation. During the project, organizing team meetings was a challenge because of absence of team members due to other responsibilities. The attitude of management has been described to drive or hinder effective WSP implementation in the East-Africa region as they prioritise the work of the staff (Parker and Summerill, 2013), and was also observed in the subject project implementation. This has been recognized for other regions as well, like the Asian Pacific region (Kumpel *et al.*, 2016), which may be caused by insufficiently supporting management. Moreover, the role and motivation of the team leader is crucial, and at both utilities the team leader changed during the project. The replaced team leaders had significantly different ideas on implementing CR-WSPs, such as composition of the teams, regular meetings and priority areas. Long time periods between project visits due to several factors reduced motivation and commitment to continue.

Challenges that affected the WSP and WQM activities were comparable to challenges as described by Peletz *et al.* (2016), such as procurement processes that were delayed due to the requirement of multiple bids; lack of a provider in the country and availability of a vehicle for sampling of the distribution network. At the beginning of the project, laboratories appeared to have had equipment obtained from previous projects. Based on this, the project management decided not to invest in similar products, but to pay more attention to training and application. This advocates for a thorough needs' assessment to identify existing materials and needs of the laboratory before investments, as was done in this project.

Lack of support from other stakeholders, particularly on catchment management is a barrier for CR-WSP implementation (Parker and Summerill, 2013). Management of water resources is essential to achieve sustainability of water services. This is impeded when there is a lack of coordination such as between the urban service delivery operator and stakeholder acting in rural catchment areas. In these situations, instead of establishing dialogue and collaboration, the priority has often been to develop technical capacity for managing the engineered water supply infrastructure. Addressing climate resilience requires new skill sets and collaborations to consider the catchment, the management of the land and the other water users, as well as how all of these aspects are expected to change. Delivering climate resilient water supply services to the growing, developing populations of Addis Ababa and Adama is a major coordination challenge to promote inter-sectoral cooperation.

Benefits identified during implementation

Kumpel *et al.* (2018) describe an assessment scheme for showing impact of implementing WSP based on performance indicators, including infrastructure improvements, increased financial support, changes in operations and management practices, non-revenue water, water quality testing and monitoring consumer satisfaction. According to WHO and IWA (2017), main benefits of WSP implementation are:

- improved system management;
- increased awareness, knowledge and understanding among staff;
- increased promotion and knowledge sharing;
- improved communication and collaboration;
- improved water quality;
- improved monitoring;
- increased capacity building and training;
- improved record keeping; and
- improved managerial and operational procedures.

This shows that most benefits are related to performance indicators which cannot easily be numerically measured, with the exception of improved water quality. One challenge in documenting improvement is that the water supplies for which data is available at the outset tend to be higher-capacity supplies with high indicator levels at baseline already, whereas other supplies with more room for improvement tend to lack baseline data, making it challenging to document improvement (Kumpel *et al.*, 2018). Although at the beginning of the project, no clear performance indicators were set to measure the impact of WSP implementation, most of the above mentioned benefits of WSP implementation could be confirmed, including the linkage to improved monitoring activities. Both water utilities reported that implementation of CR-WSPs resulted in a more proactive attitude and approach of managing the water supply, thereby preventing health hazards. Adama reported an improved reputation with the consumers. Prioritizing risks was assessed by the teams to be a solid basis for action and for attracting funding for improvements. Recently, positive customer feedback was received for action taken in case of leakages or complaints for both water utilities. During the implementation of CR-WSP, most time was spent on the first four steps of CR-WSP. This was also described by Sutherland and Payden (2017) for South-East Asia. A budget was allocated for quick wins, which were executed two to three years after the beginning of project implementation. It might be more motivating to have quick wins at an earlier stage, to motivate the team and management. These quick wins demonstrated the positive effects of the CR-WSP approach using the risk assessment as a basis.

Compared to the situation prior to the project, the laboratory staff was more motivated, because of investments that were done in the first part of the project, such as purchasing of materials and implementation of methods. The laboratory staff did not change frequently and this had a positive influence on the motivation of WQM implementation. The project introduced monitoring of additional parameters, or monitoring existing parameters more frequently, to extend information on compliance with the national standards (Ethiopian Standards Agency 2013).

Although over the past decades, monitoring of drinking water as a final product has been the norm globally to determine whether drinking water is free from contamination, it has become clear that this is often too little and too late. Final product monitoring only provides information on a limited amount of water sampled at a given time, a limited number of parameters, and results are only available after possibly contaminated water has been consumed. Introduction of a risk management approach, such as CR-WSP, provides more focus on safely managing the processes to guarantee safe drinking water. WQM is important to verify whether the CR-WSP is effective. Due to introducing or improving methods for WQM, the laboratories were able to provide information on source water quality, drinking water operations and drinking water quality that could inform CR-WSP.

Both CR-WSP and WQM benefit from strengthening the link between them: better risk management decisions could be made based on WQM data, whereas WQM could be adapted based on the input from CR-WSP. Another benefit was that there was better understanding on the activities and needs from different departments. For example, staff in the distribution network did not understand why results of *Escherichia coli* were given only after two to three days. By sharing the need for rapid results in the distribution network and the supply information on the methodology for detection, mutual understanding was created. Through consultation with the laboratories, an alternative was found to provide reliable results within a shorter time period. This example showed that communication between laboratory staff and CR-WSP is important. During the project the importance of linking CR-WSP and WQM was stressed in annual meetings and during regular visits.

A risk-based WQM approach is important for independent surveillance, particularly when limited funding is available to optimally detect evidence for the existence of risks for public health. Several organizations, such as WHO and United Nations Economic Commission for Europe, support cost-effective and risk-based drinking water quality surveillance approaches (WHO 2015). During CR-WSP implementation, several risks were identified that affected water quality from source to tap. These risks were communicated to the WQM team for adapting existing WQM to a more risk based approach. By initiating trend analyses in the water quality, information will be gathered on the presence of several parameters over the years, and will also inform about seasonal fluctuations. As a result, identified trends or fluctuations can support preventing future exceedances of the threshold values. Climate change can have negative effects on the water quality of source water and drinking water (Depla *et al.*, 2009), and this project initiated research on the effects of extreme weather events on source water quality and drinking water operations to support risk based WQM and CR-WSP. Not only climate change, but also other developments, such as urbanization, population growth, illegal settlements or industrialization, might have a negative impact on the source water quality and introduce or exacerbate risks and should therefore be considered in CR-WSPs, and may require adaptation of WQM.

To increase the sustainability and the visibility of the work of the CR-WSP and WQM teams at the drinking water utilities, participation in platforms is crucial to address water safety in the catchment and to be supported by external stakeholders. Within this project, collaboration between both drinking water utilities and the government (EPHI) was established, in order to share experiences and address challenges on both CR-WSP and WQM. Stakeholders who participated in the trainings and who supported the implementation of CR-WSP and improvement of WQM at both drinking water utilities may serve as (national) focal points for further CR-WSP implementation and improving WQM in Ethiopia in the future. Small and medium water supplies could benefit from the knowledge and expertise on CR-WSPs implementation gained within this project to implement CR-WSP in conjunction with WQM in their supplies. At the end of 2018, four members from the CR-WSP teams were involved to support the implementation of CR-WSP in four small towns in Oromia and may continue with other small towns.

Conclusions

Continuously linking WQM and CR-WSP showed to be beneficial for both project parts, as knowledge was constantly shared between different experts within the water utilities and the use of this obtained information could be optimized. At both drinking water utilities, CR-WSPs, as described in the Ethiopian Guidelines for Urban Utility Managed Piped Drinking Water Supplies (MoWIE 2015), were implemented, and WQM was strengthened, including compliance monitoring and operational monitoring at the utilities and source water quality monitoring at EPHI. This supports the benefit described in literature that one of the main benefits of WSP implementation is improved water quality testing and monitoring consumer satisfaction. Monitoring data provided evidence that was used for risk management decisions as part of CR-WSP and identified risks from CR-WSP were used to adapt WQM in a way that data is more meaningful. Such a risk-based WQM approach is better suited to anticipate environmental variations caused by climate change, urbanization, population development or seasonal fluctuations. The improvement of WQM and introduction of CR-WSP in one project at the same time supported adaptation of WQM approaches through introducing additional parameters and new methods. If the risk assessment of a CR-WSP identifies the need to analyze for additional parameters in order to verify water safety, it is very advantageous if funds have already been allocated for improving and extending WQM, as was the case in this project.

CR-WSP and WQM are iterative processes, indicating that drinking water utilities need to continue with these approaches also after project completion for sustainable application.

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Chapter 7

Experiences from piloting integrated water and sanitation safety planning in small rural systems in Serbia

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Abstract

The WHO recommends risk management approach to ensure safe drinking water and sanitation, so-called water safety planning and sanitation safety planning. However, applying these risk-management approaches separately in small-scale drinking water supply and sanitation systems might be challenging for rural communities with limited human, financial and administrative resources. An integrated approach seems a better option. In this study, an integrated water and sanitation safety planning (iWSSP) approach was developed together with guidance and training material for practical application of this novel approach. The integrated approach was piloted in three small systems in rural Serbia to identify benefits and suggestions for improvement which can be used for potential future scaling up. Implementing iWSSP at the pilot sites contributed to a better understanding of both drinking water supply and sanitation system. It also resulted in increased awareness, knowledge and understanding among staff of drinking water supply and sanitation services. Key experts, including external facilitators, played a crucial role in the implementation of iWSSP. Future scaling-up of the integrated approach could be enabled if more guidance, easy-to-use training materials and templates become available which can be adapted and updated as needed.

Introduction

In Serbia, as in many other countries worldwide, access to safe drinking water in rural areas is a challenge (WHO 2017c). Currently, a third of rural water systems in Serbia do not meet standards for microbiological drinking water quality (WHO 2017c), and more than 60% are exposed to possible contamination from latrines, sewers and other nearby sources of contamination, such as animal farming, agriculture, roads and industry (Jovanović *et al.*, 2022). Citizens living in rural areas have less access to safely managed drinking water and sanitation compared to urban areas (UNICEF and WHO, 2022), and relevant stakeholders in Serbia have taken up activities in small rural systems to improve their situation and gather more information. Examples of activities aiming at improving small systems in Serbia include a rapid assessment of drinking water quality (RADWQ) in rural areas (Jovanović *et al.*, 2017) and a project on ensuring safely managed on-site sanitation systems (SMOSS) (IPH 2021). In rural areas, people mainly rely on small-scale sanitation, such as on-site systems like pit latrines, septic tanks, and small collective sewerage systems with or without wastewater treatment (WHO 2022a). Small (community-managed) systems providing drinking water to rural populations may include simple piped water systems or a range of point sources, such as boreholes with hand pumps, dug wells and protected springs (WHO 2017b).

The WHO recommends risk assessment and -management approaches to ensure safe drinking water (WHO 2017b) and sanitation (WHO 2018) – water safety planning (WSP) and sanitation safety planning (SSP) respectively. The most effective means of consistently ensuring the safety of a drinking water supply is through the use of a comprehensive risk assessment and risk management approach that encompasses all steps in the water supply from catchment to consumer (WHO 2022b). SSPs encompass all steps in the sanitation chain from capture to reuse / disposal and should protect human health from sanitation-related risks, including from reuse of wastewater in agriculture and aquaculture (WHO 2015). WSP takes into account possible contamination affecting the drinking water, including sanitation (WHO 2022b). SSP also takes into account other exposure routes such as direct contact and different exposure groups. With SSP additional and specific information from the sanitation chain might be collected which pose a threat for the drinking water supply. Within SSP control measures can be taken which might have a positive effect on the drinking water quality (WHO 2015). Both WSP and SSP should also be applicable for small systems (WHO 2015, 2022b). Health risks from exposure to contaminated drinking water can be significantly reduced through applying WSP as shown in Iceland (Gunnarsdottir *et al.*, 2012). Winkler *et al.* (2017) described benefits of the SSP approach which may affect public health such as reducing pollution, eliminating dumping and minimizing the release of hazardous chemicals and materials. However, no further information has been published so far about the health benefits when applying SSP. WSP has been implemented in more than 93 countries (WHO 2017a), whereas SSP implementation is lagging behind, but a much stronger focus on sanitation is needed (WHO 2022a). In Serbia, both WSP and SSP are scarcely implemented. For rural communities with limited human, financial and administrative resources, the implementation of such approaches is not straightforward and support is required (Herschan *et al*., 2020). In smaller and more local contexts, drinking water and sanitation management are inevitably interlinked, partly due to their close proximity. The same people might even take care of both systems. An integrated water and sanitation safety planning (iWSSP) approach could be a good context-specific option. Information on an integrated approach and details of its implementation are scarce. Barrington *et al*. (2013) adapted WSP to small systems in rural Nepal by including sanitation and hygiene. Clavijo *et al*. (2020) described a water and sanitation safety planning in a metropolitan area in Latin America and another study in South Africa described the barriers of water and sanitation safety planning implementation in rural areas (Murei *et al*., 2022). Huber *et al*. (submitted) developed an integrated climate-resilient water and sanitation safety planning in South Africa. However, there is only limited experience with piloting these integrated approaches and these studies do not specifically address the implementation in small supplies.

The project “Developing an innovative approach to improving drinking water and sanitation safety in small systems through integrative management in Serbia” was conducted from April 2021 to May 2022. The National Institute for Public Health and the Environment (RIVM) in the Netherlands acted as coordinator with the Institute of Public Health of Serbia (IPH) and the German Environment Agency (UBA) as project partners. The WHO Regional Office for Europe supported the project but was not an official partner. This paper describes the development and piloting of an integrated approach for water and sanitation safety planning for small systems.

Methods

Developing an integrated water and sanitation safety planning approach

The WSP approach for small systems and the SSP approach encompass seven and six systematic steps respectively (WHO 2015, 2022b), and share similarities in method, purpose and goals. Using these approaches as a starting point, an approach for iWSSP in rural small-scale systems was developed by UBA and RIVM. Integration of both approaches started with aligning and integrating the steps, which resulted in an iterative process cycle to facilitate continuous improvements in drinking water and sanitation over time, visualising the circular nature of the iWSSP approach containing six steps (Figure 7.1). In this study, re-use of (treated) wastewater was not taken into account as this was not applied in the pilot sites. These iWSSP-steps are:

Step 1 – Prepare for iWSSP

Define the objectives and scope of the iWSSP, identify stakeholders and assemble a team. In this step, both drinking water supply and sanitation should be covered in the objectives and scope. The iWSSP team should be a multidisciplinary team including experts in the field of drinking water supply and sanitation as well as external stakeholders such as representatives of health authorities, environmental agencies and users.

Step 2 – Describe the drinking water supply and sanitation system

Accurately describe the drinking water supply and sanitation system and compare with the real situation in a site visit. In addition to WSP and SSP, available and newly collected data on drinking water supply and sanitation are combined to provide insights on interconnections between the systems. Develop combined information documentation such as maps of the drinking water supply and sanitation system.

Step 3 – Analyze hazards and hazardous events, identify existing control measures and assess risks

Identify biological, chemical, physical or radiological hazards and hazardous events and assess the risk based on severity, likelihood of occurrence, and the effectiveness of existing control measures. In this step, hazards and hazardous events for both drinking water supply and sanitation are identified. Combined information from step 2, such as maps of the drinking water supply and sanitation system, also allow teams to identify vulnerabilities and risks based on interconnections. A risk assessment is conducted in a way that the risk levels of both systems can be compared.

Step 4 – Develop and implement an incremental improvement plan

Develop a detailed improvement plan to address all significant risks requiring additional control. The improvement plan focuses on new or improved control measures that prevent, reduce or eliminate the identified risks of both drinking water supply and sanitation. Those measures can foster positive interactions across both the drinking water supply and the sanitation system.

Step 5 – Monitor control measures and verify the effectiveness of the iWSSP

Define an operational monitoring plan for important control measures and obtain evidence that the iWSSP as a whole is working effectively. In this step, operational monitoring covers both the drinking water supply and the sanitation system to check if these are operating as intended at any given point in time. The operational monitoring plan includes actions to eliminate the cause of a non-conformity (non-fulfilment of an operational target) and to prevent recurrence. Verify the effectiveness of the iWSSP by compliance monitoring, auditing or customer satisfactory surveys.

Step 6 – Develop supporting programmes and review plans

Develop supporting programmes that contribute to reaching the iWSSPs objectives and review the iWSSP on a regular basis.

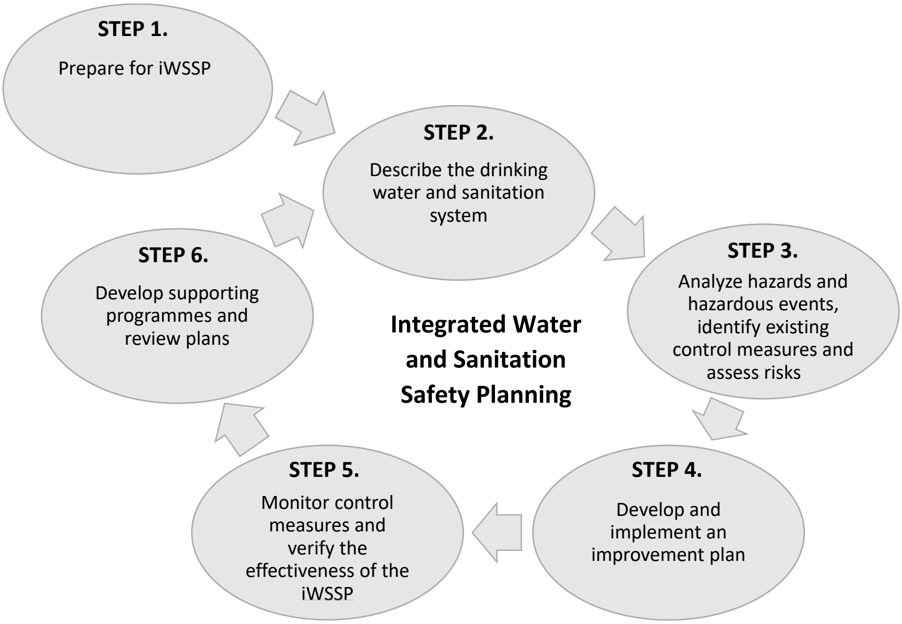

Figure 7.1. *Six steps of integrated water and sanitation safety planning.*

Capacity building

In addition to the development of the iWSSP approach, a series of capacity building activities took place to support piloting the approach:

Sensitization workshop

The sensitization workshop was held on 16th June 2021 in hybrid in-person and online format. This was hosted by IPH, Belgrade. The workshop was held to create enthusiasm and support for the implementation of iWSSP in small supplies in Serbia. IPH mapped relevant stakeholders for the sensitization workshop and implementation of iWSSP. Twelve national and local stakeholders involved in health, water and sanitation services, environmental protection and infrastructure were identified. The 30 workshop participants represented the identified national and regional stakeholders. Representatives from the WHO regional office for Europe demonstrated the importance of Water, Sanitation and Hygiene from a public health perspective. The Serbian Ministry of Agriculture, Forest and Water Management and IPH provided information on the status of drinking water and sanitation practices in Serbia. International experts from RIVM and UBA sensitized the participants to the concepts of WSP and SSP through lectures on the key steps and benefits of the approaches, and provided information about the project.

Development of training materials and supporting materials

To achieve an impact to both drinking water supply and sanitation systems under one integrated approach an acceptable balance between both systems was needed in all steps. To support capacity building and implementation of this iWSSP approach, a concept guidance document, templates and training materials were developed with the aim of integration of drinking water- and sanitation aspects.

Training and capacity building workshop

A three-day training- and capacity building workshop took place between the 21st and 23rd of September 2021 in hybrid format, hosted by IPH, Belgrade. The workshop was organized to build sufficient capacity within the implementing teams to execute iWSSPs at the pilot sites in rural Serbia, and to provide information for facilitators to support local iWSSP implementation. The capacity building workshop had nine participants from the drinking water and sanitation sector organizations in Serbia. Information was shared by RIVM and UBA on iWSSP and on the role of facilitators. In total, nine persons were fully trained (five facilitators, three implementers, one observer). The workshop included lectures, interactive exercises, training of facilitators, and exchange between attendees of contextual information about each of the pilot sites.

Piloting of iWSSP

Three pilot sites in two rural areas in Serbia were identified and selected for implementing iWSSP (Figure 7.2). Pilot sites were selected for differences in sanitation practices and management types for rural small drinking water supply systems, as well as institutional and community engagement (Table 7.1).

Figure 7.2. *Map of Serbia with the pilot sites: Sokolovica, Mladenovo and Pivnice (light gray dots)*.*

** This map is different compared with the map in the original paper.*

iWSSP implementation took place following capacity building activities over a period of nine months, from September 2021 to May 2022, piloting the developed iWSSP approach in all three pilot sites. Facilitators from the IPH of Serbia and regional branches of IPH, who had been trained at the capacity building workshop, together with the local communities, implemented the developed iWSSP approach. The implementation of iWSSP was supported by experts from RIVM.

Table 7.1. *Attributes of iWSSP pilot sites.*

	Pilot site		
Village	**Sokolovica**	**Pivnice**	**Mladenovo**
Municipality	Kursumliji	Backa Palanka	Backa Palanka
Population	440	3,300	2,600
Drinking water source	4 natural springs	2 deep wells	3 deep wells
Drinking water treatment	-	Chlorination	Chlorination
Drinking water system authority	Community	Public utility company	Public utility company
Sanitation system type	Septic tanks (on-site sanitation)	Septic tanks (on-site sanitation)	Centralised sewer system (80%) Septic tanks (on-site sanitation, 20%)
Sanitation system authority	Private households	Private companies	Public utility company
Land area	1098 km^2	579 km^2	
Relief	Mountains	Flat land	
Landforms	Forest and fields	Fields	
Land use	Agriculture (fruit growing, medical herbs for teas, mushrooms)	Industry (food and wood) and agriculture	

The progress of iWSSP implementation was monitored and documented regularly for each of the six steps. IPH and RIVM organised fortnightly online meetings throughout the implementation period to share experiences between the facilitators and to discuss progress made and challenges observed by the facilitators. To ensure that experiences, best practices, and challenges were shared between the pilot sites, two peer learning visits were conducted in April and May 2022. During these peer learning visits, teams from one rural area visited the drinking water and sanitation services of the other area and the teams discussed experiences, best practices, and challenges. In May 2022, during the second peer learning visit, two representatives of each iWSSP team were interviewed by RIVM, using a semi-structured interview, to gather experiences during the implementation.

Results

Capacity building

The sensitization workshop increased knowledge and improved understanding of participants on the main principles and steps in developing WSP and SSP, and the approaches' integration. Stakeholders from the drinking water and sanitation sectors recognized the need of introducing SSP in legislation, while WSP has already been addressed in the draft national law on water intended for human consumption. Furthermore, local stakeholders and pilot sites were enthusiastic to implement iWSSP and to become forerunners in the implementation of a new approach for small systems.

Training materials based on the iWSSP approach were developed to support capacity building. The materials contained background information on WSP and SSP, and on benefits of combining both approaches. The training materials emphasized the need of integrating WSP and SSP, ensuring that both drinking water- and sanitation system were addressed. This included the development of materials such as PowerPoint presentations and handouts, to explain the iWSSP approach and its six steps in detail. Templates, such as inspection forms, questionnaires, and tables, were developed to collect and document relevant information of the drinking water supply or sanitation system. Some examples of supporting materials were:

- the template for objectives contains objectives for both drinking water supply and sanitation (step 1);
- the guidance document stating that stakeholders should be identified from both drinking water supply and sanitation (step 1);
- a combined system description template to collect data on both systems (step 2);
- definitions of likelihood, severity and risk that matched for drinking water and sanitation (step 3, see Supplementary Material 1); and
- an Excel sheet was developed to document all relevant information regarding steps 3 – 5 in a systematic way for both drinking water and sanitation (Supplementary Material 2).

Materials were translated into Serbian for the implementation of iWSSP at the pilot sites. During the capacity building workshop, the six iWSSP-steps including the developed materials were explained in detail. Through familiarising with the developed templates, participants made a start in using these materials, and gained confidence in explaining them independently to professionals at the pilot sites in the subsequent practical implementation. Facilitators received additional information on their role and specific tasks to support them in their role in implementing iWSSP at the pilot sites. The capacity building workshop was well received by the facilitators, though it is challenging to conduct the training remotely due to the COVID-19 pandemic. The participant mentioned that seven hours a day is too intensive and tough, with a lot of content, for a digital training. The topics were dense and the presentations given in English, which was challenging for some participants. Training materials were well accepted by the facilitators as they found these materials very well structured and supported with respective tools for understanding and practicing what would be expected in the field work. The participants mentioned that the training and final implementation of iWSSP could contribute to improve conducting their work in a more systematic way. Overall, the participants were very content with the training workshop and eager to start iWSSP implementation at the pilot sites.

Piloting the iWSSP approach in small systems

The iWSSP approach was piloted in three pilot sites in two rural areas in Serbia. For each iWSSP step the experiences related to the process as well as technical information are described below.

Step 1: Prepare for iWSSP

The iWSSP teams Sokolovica (pilot site Sokolovica) and Backa Palanka (pilot sites Mladenovo and Pivnice) could be established relatively easy due to the stakeholder mapping and sensitization workshop. The teams contained expertise from both drinking water supply and sanitation. Besides the entities for drinking water supply and sanitation, the iWSSP team also included national and regional public health officers, local government, and local communities. In Backa Palanka, industries were located in the catchment, so representatives from industries were also included. In the selected pilot sites, no challenges were observed in mobilizing the iWSSP teams. The very good relation of IPH with the community and/or utilities probably contributed to the engagement, and furthermore, travel costs were compensated for iWSSP team members for participating in meetings and field visits. The objectives for iWSSP were set by the multidisciplinary iWSSP team and covered both drinking water supply and sanitation: to ensure safe drinking water by minimizing contamination of water sources and reducing or removing contaminants. Sub-objectives were set, such as ensuring continuous control of drinking water and the practice of wastewater disposal and education of legal and natural persons on safe ways of using/producing drinking water and wastewater disposal.

Step 2: Describe the drinking water supply and sanitation system

For the system description, information was available in diagrams and/or narrative form at the pilot sites. These were important starting points for the iWSSP teams to collect additional information to provide accurate, relevant, and up-to-date system descriptions, without starting from scratch. More data and information was available for drinking water and sanitation system, such as maps or flowcharts, when this was utility managed compared to community managed. Furthermore, the utility had more staff and expertise available to collect additional information then the community responsible for drinking water and sanitation systems. Collecting detailed information for sanitation systems was challenging, especially in case of on-site sanitation (septic tanks), as they are located on private premises. The iWSSP teams did not have authority to access private properties. It was technically challenging to create and produce maps on sanitation that include on-site sanitation facilities. It was difficult to create a complete overview of septic tanks in use, or septic tanks which were abandoned but still present. As the utility had more experiences with creating maps and flowcharts, more difficulties were observed for the community in creating (integrated) maps.

Integrating drinking water supply and sanitation system data into one unified map was found challenging. A combined map was not created, but to gather information on the interconnections between both systems, the separate maps were jointly examined. The iWSSP teams recognized the need for describing the systems in a comprehensive way. After filling in the system description template, one of the iWSSP teams also described their systems in a shorter way that would provide greatest contextual fit and practicability for use on location, e.g. to train new staff. The system description contributed to improved understanding of the drinking water supply and sanitation systems and resulted in accurate and up-to-date system descriptions in diagrams and narrative form.

Step 3: Analyse hazards and hazardous events, identify existing control measures and assess risks

The iWSSP teams conducted at least one field visit to each pilot site to gather and check information on the system description for the drinking water supply and sanitation systems. During the field visit the iWSSP teams identified hazardous events and existing control measures. The field visits also contributed to sensitizing and improving awareness and enthusiasm in the communities. Information gained from the field visits enabled iWSSP teams to finalize the system description from iWSSP step 2. For identifying hazards and hazardous events, the following documentation templates were used: sanitary inspection forms (WHO 2020), sanitation inspection forms (WHO 2022c), list of hazardous events (compilation developed for iWSSP within this project) and forms developed under the SMOSS project. Projects such as RADWQ and SMOSS already introduced templates to identify hazardous events that all facilitators were familiar with (IPH 2021; Jovanović *et al.*, 2017). Using these well-known forms was easier for the facilitators instead of using a new or additional list of hazardous events. For the three pilot sites, the iWSSP teams needed significant support from facilitators to fill in the template for hazardous events and assessing the risks for all identified hazardous events and related hazards in the drinking water supply and sanitation systems after considering existing control measures. The compilation of hazardous events was comprehensive, and due to the length of the list provided, both iWSSP teams became concerned about the perceived complexity. Adaptation of this template by facilitators through preselection and shortening the list of hazardous events resulted in greater confidence of the iWSSP teams in their ability to complete this task. In total, 77, 47 and 58 hazardous events from the compilation were identified to be present for respectively Sokolovica, Pivince and Mladenovo. In Sokolovica, 44 (57%) of the hazardous events were related to the drinking water supply, and 33 (43%) to the sanitation system. In Pivnice, a similar distribution between the hazardous events related to the drinking water supply and sanitation were observed, respectively 60% and 40%, and for Mladenovo, the number of hazardous events were equally distributed with 50% for both systems.

All hazardous events were compiled in one Excel table for which a template for iWSSP steps 3, 4 and 5 was provided, as shown in Supplementary Material 2. In this table, the risk assessment was documented, categorising each of the hazardous events and the related hazards as high, medium and low risks. Facilitators were needed to provide information on identifying hazardous events using the templates and to support the risk assessment. This emphasises the need for facilitators in implementing iWSSP, as not all communities are used to filling in questionnaires, especially not related to drinking water and sanitation, and as the risk assessment was too difficult for local communities to complete without external support.

Step 4: Develop and implement an incremental improvement plan

For all combinations of hazards and hazardous events for which the risk was medium or high, the iWSSP teams identified improvements and documented them in improvement plans included in the Excel table (see Supplementary Material 2). The improvement plans triggered the communities to initiate some immediate improvements performed there-and-then despite of no formal budgets within the project to do so, such as cleaning areas around storage reservoirs, locking fences around storage reservoirs, installing vent screens on aeration pipes at the storage reservoirs and changing the shape of storage reservoir aeration pipes by adding a U-turn pipe. These immediate improvements only took place within the drinking water supply, as the iWSSP team of Sokolovica had the mandate to adapt the system. Both iWSSP teams mainly identified possible mid- to long-term improvements for drinking water and sanitation systems. Examples for improvements in the drinking water were renewal or repair of the drinking water supply network, improving procurement of appropriate equipment, education of staff and raising awareness on source water protection to local self-government. For sanitation, examples of suggested improvements were the introduction of permits for the construction of septic tanks and education on the proper handling of waste in order to protect the health of people for both sanitation workers well as the general population. The outcomes of this study reflected to the short-term improvements in the public utility company (PUC) “Kursumlija” that support operation and management of community level water supply system, including Sokolovica. The concrete improvements refer to the overall operation and management of PUC in development of WSP for this particular water supply system and enchantment of control monitoring.

Step 5: Monitor control measures and verify the effectiveness of the iWSSP

Different types of monitoring were already conducted for drinking water quality and wastewater at all pilot sites. The drinking water utility or community conducted operational monitoring by measurements and/or observations, and IPH conducted compliance monitoring to check if the drinking water quality met the regulations. An operational monitoring plan for drinking water supply and sanitation was developed by the iWSSP teams. This was based on existing monitoring activities, and new measurements or observations focussing on identified control measures were added. For example, the drinking water utility in Sokolovica directly increased the frequency of operational monitoring after receiving a field kit for testing turbidity, pH, temperature, and conductivity. Regular checking of turbidity in the drinking water source and measuring residual chlorine in the pumping stations throughout the distribution network were implemented after ending the project. In this operational monitoring plan, visual inspections were addressed for both the drinking water supply and sanitation systems. In case of inspections of septic tanks, operational monitoring was added in the plan, but needed to be checked with the competent authority. Quality parameters were included for operational monitoring of the drinking water system.

Step 6: Develop supporting programmes and review plans

Many activities on supporting the implementation of iWSSP are undertaken by the drinking water utilities, communities and/or the local IPH as something that is done often and considered normal. Examples were calibration of equipment, collaboration between IPH and utilities in measurements, communication with local government, and analysis of public health related to waterborne diseases. iWSSP teams provided an overview of programmes that may support the iWSSP approach. The iWSSP teams then developed a plan on how to revise and review iWSSP in the future. During the project, it was possible to revise details about the drinking water and sanitation system such as adding new information to the system description from field visits, or changes to the iWSSP team as resignations occurred and new employees were hired.

iWSSP implementation was supported by many activities carried out by drinking water utilities, sanitation utilities, communities, or IPH Serbia, as well as the local authorities. Most of these were routine and commonly practiced activities and were therefore not always regarded as supporting programmes. Examples of supporting programmes were educating employees, the provision of health information on waterborne diseases to residents, collaboration between IPH Serbia and drinking water supplier on monitoring, continuous maintenance of defined drinking water supply and sanitation processes, and communication and awareness of the importance of drinking water supply and sanitation among legal entities, households, and other stakeholders.

The iWSSP teams agreed to meet every three months in the future, and to hold a mandatory meeting after an incident in the drinking water and sanitation system. The periodic review of the iWSSP was planned to take place annually.

Sharing expertise and best practices

Fortnightly meetings were scheduled between Serbian facilitators and the RIVM during the project period. Progress, experiences, and challenges were discussed in detail, and facilitators appreciated the regular meetings. The meetings supported the implementation as challenges could be troubleshooted and resolved. Frequent meetings provided structure for exchange and contributed positive momentum in the delivery of the project.

Peer learning visits were conducted to share expertise, lessons learned and experiences between the iWSSP teams and local communities of the three pilot sites. In April 2022, a peer learning visit took place in Sokolovica. Twelve people participated in this visit: Sokolovica iWSSP team (5); Backa Palanka iWSSP team (5); IPH (1) and RIVM (1). The mayor of Kursumlija and director of the drinking water supply system of Kursumlija attended at the end of the first day. In May 2022, a peer learning visit was arranged in Backa Palanka (Mladenovo and Pivnice). Fifteen people participated in this meeting: Sokolovica iWSSP team (5); Backa Palanka iWSSP team (9) and IPH (1). Due to practical reasons and COVID restrictions not all team-members could join the visits. Both drinking water supply and sanitation experts joined each visit. Participants of these visits found it useful to observe the processes in a different setting and were able to learn more about the drinking water and sanitation systems in other places, and how their peers dealt with (similar) challenges. Moreover, peer-learning visits triggered the iWSSP teams to critically review their risk assessments and monitoring plans. This subsequently contributed to improvements in their respective iWSSPs.

Discussion

The integrated approach was piloted in three small systems in rural Serbia. This approach was conducted to increase knowledge knowledge and understanding of the drinking water supply and sanitation system and its vulnerabilities among staff. Similar findings were observed by Van den Berg *et al*. (2019) who described that implementation of WSP contributed to greater understanding of the drinking water supply. By integrating drinking water and sanitation safety planning, improved safety can be achieved through a better understanding of both systems, how they are interrelated, and how they can influence each other.

The representatives of the iWSSP teams mentioned that they were positive on the multi-disciplinarity and multi-stakeholder involvement in the teams. The teams could provide an evaluation of the entire water cycle, including drinking water supply and sanitation. Similar findings were observed by Clavijo *et al*. (2020) and it was shown that the absence of collaboration between different stakeholders could be a barrier for iWSSP implementation (Murei *et al*., 2022). Due to the presence of diverse stakeholders in the iWSSP teams, improved communication and collaboration was observed between the often siloed drinking water and sanitation domains, which is also a benefit of WSP implementation (WHO 2017a).

WHO reported that policy and regulatory instruments serve as critical drivers for WSP implementation (WHO 2017a). Also, Schmiege *et al*. (2020) described that formal rules together with the conditions that affect the achievement of objectives (enabling environment) at policy level is required for effective country-wide scaling up of WSP implementation. In Serbia, WSP has already been addressed in a draft new law on water intended for human consumption, but SSP is not addressed in legal regulations. In this project, the drinking water and sanitation sectors recognized the need of introducing SSP in legislation. Specific policies and regulatory drivers strongly support implementation of iWSSP and scaling up (Clavijo *et al*., 2020), after this project has been completed. Inequality in financial power is described as one of the barriers for iWSSP implementation (Murei *et al*., 2022). Financial support is critical for successful WSP implementation in order to avoid additional burden on communities with limited financial capacity (WHO 2016). In this project, budget was available for capacity building, organizing meetings, transport, and external support to implement iWSSP at the pilot sites. Already some improvements have been made and intend to improve more in the near future. The short-term improvement made for operational monitoring showed its results and effectiveness during a recent emergency situation i.e. flooding in the region in June 2023, affecting drinking water sources with flooded water and raised river level. Based on improved monitoring, turbidity has been tested regularly allowing timely information for the operational team to react and temporary exclude certain sources with increased turbidity from the system. However, for larger improvements external funding sources are needed. Herschan *et al*. (2020) described that building on achievements and existing activities is cost-effective and sustainable. This was experienced in several iWSSP steps in this project. Therefore, attention should be placed on what is already done and practiced by the relevant stakeholders and how their existing practices fit into the iWSSP steps. During the implementation phase, it was possible to build on day-to-day practice to show that iWSSP could work in synergy with current routine processes without requiring extra human, material and financial resources. Outcomes of iWSSP can support management in better decision making for medium to long-term investments in the drinking water supply and sanitation system. In this way, it became clear that iWSSP builds upon established daily practices and strongly supports continuation of iWSSP after initial introduction, as also document for WSP (Herschan *et al*., 2020).

According to the representatives of the iWSSP teams it was useful to combine available information on drinking water supply and sanitation services to identify or better understand possible risks. However, we experienced that integrating drinking water supply and sanitation was difficult as the systems are managed in different ways and physical overlap is limited. Lack of comprehensive and integrated assessment of drinking water supply and sewerage at the pilot sites were observed. This was caused by the existing arrangement of systems and organization in public utility companies in which jobs on drinking water and sewerage are separated and do not have overlap. Furthermore, in some rural areas public drinking water supply systems are not managed by a utility, but by local communities, and nobody is responsible for the operation with on-site sanitation. At the pilot sites, less information was available on sanitation and less activities were observed related to sanitation. Implementing iWSSP in Sokolovica was difficult due to a lack of mandate for inspecting on-site sanitation systems to observe the system and identify hazards and hazardous events. Although Clavijo *et al*. (2020) suggested that using risk assessment tools, such as iWSSP, could provide a more standardized approach toward the assessment and management of on-site systems, we experienced that in pilot sites with only on-site sanitation, integration was less popular. Besides the use of an integrated approach in small supplies in rural areas it might be applicable in urban areas as well as described by Clavijo *et al*. (2020). Theoretically, it would be beneficial to combine both approaches especially in rural areas with limited resources. However, it is difficult to determine if iWSSP is more beneficial when compared with separate WSP and SSP implementation, as this was not investigated in this project. Therefore, it would be useful to compare pilot sites where both WSP and SSP have been implemented separately with pilot sites where an integrated approach was used. In this study, we did not assess the impact on water quality, sanitation practices and management. In future studies it would be useful to measure the impact and include auditing implementation.

In Serbia, climate change was identified as a significant threat to the provision of safe drinking water, given the high frequency of flooding in the last 15 years and the number of affected river basins (Anonymous 2017). Smaller drinking water supply systems in rural areas are particularly vulnerable to climate change. Notably, as these supplies often rely on a single water source, they are sensitive to torrential rainfall and flooding or droughts (Serbia 2014). Including climate change into integrated water and sanitation safety planning would be needed to create resilience to climate change. Huber *et al*. (submitted) provide options of accessing climate information to improve climate resilience of the drinking water supply and sanitation services. In this study, re-use of (treated) wastewater was not taken into account as this was not applied in the pilot sites. However, when water scarcity increases due to climate change or urbanisation re-use of water might be considered.

Training tools that are locally appropriate and available in local languages supports implementation of the WSP (Van den Berg *et al*., 2019; Schmiege *et al*., 2020). Before scaling up implementation of iWSSP in Serbia, it is recommended to revise and update the templates based on the feedback and experiences from this pilot project. It is recommended that templates be specific for small systems. They should also remain flexible enough to allow the integration of resources such as photos, tables, and written text. Templates should be generalised, which would allow better consideration of the variety of drinking water supply and sanitation systems present in small rural supplies, as well as in settings beyond Serbia. As extension of this project, training materials and supporting materials will be updated. In the future, the supporting materials can be improved based on new experiences and lessons learned incrementally. Implementation of iWSSP in small communities in rural areas requires the support of well-trained facilitators. This was achieved through a train-the-trainer approach in which key experts were trained as facilitators to such a level that enabled them to implement iWSSP, train others and advise on iWSSPs, which is in line with the requirements for sustainable uptake of WSP (Winkler *et al*., 2017; Schmiege *et al*., 2020). Local staff needed significant support from facilitators to implement iWSSP, for example, with filling in the templates in identifying hazardous events and assess the risks. External support from facilitators was also recognized by other WSP studies (Rahman and Paul, 2013; Sutherland and Payden, 2017). The facilitators showed high levels of integrity toward their responsibilities to support iWSSP implementation at the pilot sites. For scaling up, it is necessary that national IPH train other local IPH staff to support uptake of iWSSP as a novel approach that diverges from what is currently performed.

Conclusions

Under this project, an iWSSP approach was developed to integrate water and sanitation safety planning and piloted in rural areas in Serbia. Although the integrated approach seems to have potential, more information on the impact, benefits and feasibility of this integrated approach should be collected with future studies or applications at pilot sites. Future scaling up of this integrated approach would be beneficial as more guidance becomes available, especially practical easy-to-use guidance such as tools and templates which can be adapted and updated as needed. Furthermore, key experts play a crucial role in scaling up iWSSP and therefore sufficient experts (facilitators) should be trained to support local communities with implementation and to train others.

Study limitations

The main focus of this study was to develop an integrated approach of water and sanitation safety planning. This study did not include assessing the impact on water quality and management practices. In future studies it would be useful to measure the impact and include auditing implementation, as described for WSP. Although the pilot sites had different setting conditions, the number of pilot sites was too low to make a clear statement on the influence of the setting condition on the implementation. Close collaboration and regular meetings between the facilitators in the different iWSSP teams resulted in sharing challenges, lessons learned and best practices for all iWSSP steps. In this way the different settings were able to better deal with possible issues. Therefore, it is not possible to provide information to what extent the setting conditions affect the implementation.

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Chapter 8

General discussion

Risk management, including risk assessment and water quality monitoring, plays an important role in safely managing drinking water supply and sanitation services. In the chapters of this thesis, these topics were addressed for drinking water supply and sanitation services in different countries: Mozambique, Ethiopia, Serbia and the Netherlands (see Figure 8.1).

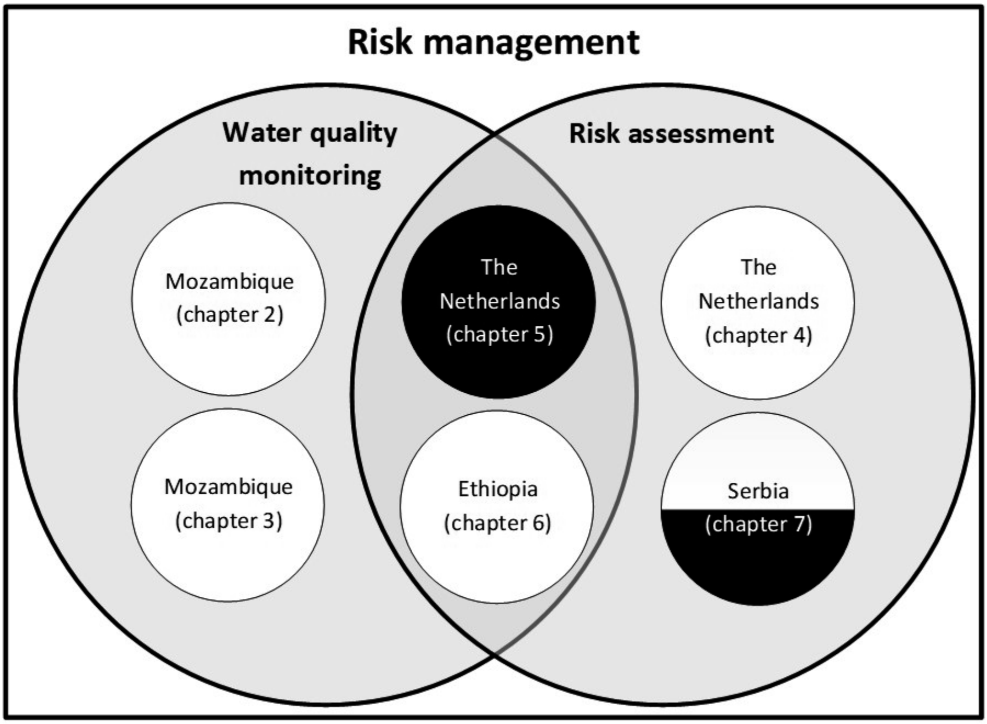

Figure 8.1. *Overview of the countries (chapters) in which risk assessment and water quality monitoring as part of risk management were addressed for drinking water supply (white circles) and sanitation services (black circles).*

Sanitation services are lagging behind in comparison to drinking water supply services

A call for action to promote prosperity while protecting the planet has been formulated in the sustainable development goals (SDGs) (UN 2015). SDG 6 'Clean water and sanitation' aims to ensure availability and sustainable management of drinking water and sanitation for all populations, respectively target 6.1 and 6.2 (UN 2015). An indicator for these targets is the proportion of a population using safely managed drinking water and sanitation services (UN 2015). To achieve universal coverage of basic drinking water and sanitation services by 2030, a dramatic acceleration in current rates of progress is needed in low-income countries (UNICEF and WHO, 2023). In the period 2015 – 2022, global coverage of safely managed drinking water and sanitation increased from 69% to 73% and 49% to 57%, respectively (UNICEF and WHO, 2023). In 2022, 2.2 billion people lacked safely managed drinking water, and 3.5 billion people lacked safely managed sanitation. At the current rate of progress, the world will reach 81% and 67% coverage by 2030, for drinking water and sanitation respectively (UNICEF and WHO, 2023; WHO 2023a). Especially low- and middle-income countries face difficulties in meeting high coverage of safely managed drinking water and sanitation.

Worldwide, many different risk management approaches are available to guarantee safe management of drinking water supply and sanitation services (Winkler *et al.*, 2017; Tsitsifli and Tsoukalas, 2021; Kombo Mpindou *et al.*, 2022; WHO 2022e, 2023a; Odjegba *et al.*, 2023). In the European context, legislation enforces the implementation of risk assessment and risk management and should be carried out from source to tap according to the European Drinking Water Directive 2020/2184 (EU 2020). In the proposed Urban Wastewater Treatment (recast) Directive 2022/0345 a risk-based approach was introduced for most of the proposed measures, which will help ensure that investments are taking place where they are needed (EU 2022a). In the pan-European region, the Protocol on Water and Health promotes and supports implementing risk management for drinking water supply and sanitation services (UNECE and WHO, 2021). The Dutch Global Health Strategy 2023-2030 addresses the contribution of the Netherlands to the improvement of water management for drinking water supply and sanitation services (MoFA and MoH, 2022). In 2017, WSP was implemented in more than 93 countries of which 46 reported having policy and/or regulatory instruments promoting or requiring WSPs. In another 23 countries such instruments are under development (WHO 2017a). On the other hand, uptake of risk-based approaches regarding sanitation services in legislation and practice is limited across the pan-European region (WHO 2022a). However, the SSP manual incorporated experiences of SSP in more than 25 countries across different regions (WHO 2022d). A much stronger focus on sanitation by national and local governments is needed to meet SDG6 (WHO 2022a; UNICEF and WHO, 2023). For example, we showed that in Serbia both on national and local scale, the focus was more on safe drinking water supply than on sanitation services (chapter 7). Integrating risk management frameworks, such as WSP and SSP, into one approach could contribute to increase focus on sanitation especially at the local level and/or where water sources are limited.

Integrating risk management frameworks for drinking water and sanitation

Drinking water supply and sanitation systems are closely related. Important interactions between the two systems may lead to risks (Pitkänen *et al.*, 2011; Narayan *et al.*, 2021). However, risk assessment and risk management frameworks have thus far addressed drinking water and sanitation separately. Some examples of integrated risk management approaches exist. For example, an integrated risk management approach for drinking water and sanitation services was applied in a metropolitan area in Latin America (Clavijo *et al.*, 2020) and in Belgium, risk management of treated domestic wastewater was integrated in the existing potable water production process (Dewettinck *et al.*, 2001). In smaller and more local contexts, drinking water and sanitation management are naturally interlinked, partly due to their close proximity. For drinking water supply and sanitation services for small systems in rural Serbia, we integrated the risk management approaches WSP and SSP (chapter 7). This study showed that integrating the risk management frameworks was difficult as the systems are managed in different ways and physical overlap is limited. However, it was very useful to combine available information on drinking water supply and sanitation services to identify or better understand possible risks. In theory, it would be good to combine both approaches especially in areas with limited resources. Combining approaches on drinking water supply and sanitation is likely to become even more important in the near future when water sources become more scarce. But in practice it is difficult to determine if an integrated water and sanitation safety plan (iWSSP) is more favourable when compared with separate WSP and SSP implementation. Further studies designing how such integration can be evaluated and roll-out of the iWSSP are needed.

Risk assessment and risk management should be continuous approaches

Many different risk management approaches for drinking water supply and sanitation services are available. The principles and goals of the approaches are similar: identifying and managing risks and documenting information in a structured way. Both SSP and WSP are based on the Stockholm Framework for preventive risk assessment and management of water-related diseases and use the methods and procedures of HACCP (Havelaar 1994; Fewtrell and Bartram, 2001). The European Standard "Security of drinking water supply — Guidelines for risk and crisis management" (EN 15975) supports the WSP approach. This standard contains similar topics to WSP and is subsequently derived from the HACCP approach (EN 2015). These approaches require a continuous need for updating, following up and improvement. Although the water suppliers and sanitation providers from the different countries that were included in this thesis were all well aware of possible risks to their systems, risk assessment was not always systematically updated or even well documented. The WSP and iWSSP approaches that were implemented in Ethiopia (chapter 6) and Serbia (chapter 7) focused on both continuity and documentation. To ensure continuity of risk management approaches, the approaches should be embedded in the day to day practices as well as the management structure, which was concluded from the study described in chapter 6 and 7.

If multiple risk assessment and risk management approaches are used, combining the results from these approaches is needed to share information and to provide an overview of all potential risks from source to tap or capture to disposal. This can be used as input for an overarching framework such as WSP or SSP. Individual utility approaches should not be limited to one risk management program as alternatives can be complementary (Setty *et al.*, 2019). Examples in which risk management approaches were incorporated or combined are:

- a Drinking Water Quality Risk Management Plan incorporated HACCP and ISO 9001 systems in Australia (Jayaratne 2008);
- multiple risk assessment and risk management approaches for drinking water supply are applied in the Netherlands (chapter 4);
- the methodological HACCP approach was adapted with the FMECA method of risk assessment for management of *Legionella* contamination in drinking water supply network installations in France (Germain and Dab, 2004); and
- WSP was integrated with the HACCP system in order to achieve the ISO 22000:2005 standard in Iran (Khaniki *et al.*, 2009).

Some challenges of combining multiple approaches are prioritizing the risks based on different risk assessments and centralizing information (chapter 4). We experienced that building onto existing approaches created more commitment and enthusiasm amongst suppliers of drinking water or sanitation services than starting from scratch (chapters 4 and 6). The revised WSP manual also addresses that in many cases, water suppliers already have information and documentation about their system, including system diagrams, and may have existing information on risks (WHO 2023a). Before implementing a new risk assessment and/or risk management approach it is essential to start identifying existing approaches, to investigate how information can be shared and if it would be favorable to implement a new approach. Water suppliers should review and update existing risk assessments regularly, ensuring that the information is up to date and covers all stages of the water supply. This updated information can then be integrated into the risk management approach(es), such as WSP, addressing gaps where required (WHO 2023a).

Water quality monitoring contributes to risk prevention

For decades, water quality monitoring was used to detect risks in order to protect public health. However, it became clear that monitoring was often too little and too late and, therefore, the focus moved from detecting to preventing risks (WHO 2017c, 2018b). Water quality monitoring still plays an important role in preventing risks, because it informs risk assessment and indirectly risk management. In the study described in chapter 5, risk assessment was used to identify wastewater treatment plants with a risk of spreading *Legionella* through the air or surface water. Based on the results of water quality monitoring, other risk factors, such as type of industry producing wastewater, could be identified to improve the risk assessment. Water quality data is required in quantitative risk assessments, such as QMRA, for example source water quality data, validation data and operational data. For other risk assessments, such as sanitary inspections, water quality data is not required, but could be combined with risk assessment, e.g. combining verification monitoring (surveillance) with sanitary inspections – the so called rapid assessment of drinking water quality (RADWQ). Sanitary inspections and microbial analyses of water convey distinct information, however, a perfect correlation between both methodologies is not expected (Kelly *et al.*, 2021). Furthermore, it should be noted that RADWQ provides a nationwide snapshot of the vulnerability of possible water sources for prioritization of investments and interventions, and is not meant for interpretation of a single source (WHO 2012). Water quality monitoring supports risk assessment and risk management. Therefore, it is necessary to understand the purpose of the monitoring and how the generated water quality data could be used in risk assessment and risk management (see chapter 1).

Operational monitoring was conducted by monitoring free chlorine at the drinking water treatment plant in Moamba, Mozambique (chapter 2). Results of operational monitoring provide information on possible risks by showing that control measures are not working as intended (WHO 2017c). The results showed the need of chlorine booster stations in the system to continuously supply drinking water with sufficient free chlorine, and inform consumers on short notice. Operational monitoring showed the need to act timely to reduce health risk.

In supplied drinking water in Mozambique, **verification monitoring** was performed (chapter 2 and 3). Although, no fecal contamination was detected in treated water leaving the water treatment plant, its presence was assumed subsequently entering the distribution system. In the distribution network, the free chlorine level did not meet the standards at all sampling points. The collected data can be used in risk assessment and risk management to take corrective actions to reduce the health risks and to review the risk management to improve the system to reduce or eliminate the risk in future. Failure to meet water quality targets should lead to a review of the risk assessment and risk management to take actions or improvements to protect public health (WHO 2017c). For the non-compliance in chapter 2 and 3, direct actions were communication with consumers, and improvements to reduce or eliminate the future risk were preventative maintenance and booster chlorination.

Outbreaks should trigger to review and revise the risk assessment and risk management of drinking water and sanitation. By identifying the source of contamination corrective actions can be taken to prevent further infections. After a *Legionella* **outbreak investigation** in which a biological wastewater treatment plant was identified as a common source for this outbreak (Loenenbach *et al.*, 2018) the study described in chapter 5 was conducted. This study contributed to the development of a guidance document to support owners of wastewater treatment plants to identify, interpret and control *Legionella* risks and changes in legislation.

For each purpose of water quality monitoring information is collected to be taken into account to review and revise the risk assessment and risk management approach, and thereby prevent infectious disease outbreaks in the future. By strengthening the link between different purposes of monitoring and risk assessment and risk management, as described in chapter 5, collected water quality data could be used to improve risk assessment and risk management, and thus reduce risks.

Managing risks should be applicable from global to local

Although risk assessment, risk management and water quality monitoring for drinking water and sanitation are applied worldwide, implementation at both global and local level may be challenging (Peletz *et al.*, 2018; Ferrero *et al.*, 2019; Kirschke *et al.*, 2020). Challenges can be general such as defining an enabling environment or prioritizing specific parameters, or specific capacity such as lacking financial, human or technical resources (Kirschke *et al.*, 2020). These challenges are described in more detail in the following paragraphs.

Legislation is a driver of the implementation of water and sanitation risk assessment, risk management and water quality monitoring (WHO 2017a). Many international and national laws and guidance documents exist, which are elements of the **enabling environment** that encourages the implementation of risk management (Baum and Bartram, 2018). However, this does not automatically result in implementation at all levels, from global to local drinking water supplies and sanitation services (WHO 2017a). While guidelines and regulations promote the uptake of risk management such as WSP, other conditions such as cultures and local norms and values also influence risk management practices (Amjad *et al.*, 2016). Local guidance and support is therefore needed in order to successfully implement risk management as well as adaptation of general guidance to legislation. For example, although national guidance for climate-resilient WSP in Ethiopia exists, more external support and guidance was needed to implement WSP at the local drinking water utilities (chapter 6). The WHO developed sanitary and sanitation inspection forms and guidance documents to support local communities and small supplies to identify and assess the risks in their small systems (WHO 2020a, 2022d). In Ethiopia and Serbia, we have experienced that the use of simple and easily accessible tools and templates in local language were required to increase understanding and positively contributed to the use of the materials (chapter 6 and 7). However, local communities may still need external support in using these materials (chapter 7). External support was also required in applying the semiquantitative risk assessment described for performing WSP and SSP in different settings (chapters 6 and 7) (WHO 2022e, 2023a). To support small supplies, WHO developed a field guide on WSP for rural communities (WHO 2022c). However, no field guide exists for implementing SSP in small systems. In chapter 7, iWSSP tools and templates were developed for supporting small systems in rural areas including required knowledge on SSP implementation.

For water quality monitoring, the WHO Guidelines contain approximately 200 **specific parameters** (WHO 2017c, 2018a, 2021a). These guidelines are, however, not intended to be mandatory and the given parameters are not designed to be adopted as a complete set in national regulations or standards (WHO 2018a). However, many countries use the guidelines in setting national drinking water quality standards and in a survey 37 out of the 125 (30%) countries and territories directly reference the guidelines for Drinking Water Quality in their national standards (WHO 2021a). As a consequence, in many settings water suppliers and surveillance agencies do not meet regulatory requirements for testing that many parameters in many settings (Peletz *et al.*, 2016). The same was found for the drinking water suppliers in Mozambique (chapter 2), Ethiopia (chapter 6) and Serbia (chapter 7), which were carrying out water quality monitoring but could not cover testing for all national standards which were adopted from the WHO guidelines directly, showing the gap between legislation and practice. A sustainable way of water quality monitoring could be implemented when adapted to the local context and capacity. Depending on the resources and local settings, a selection should be made, such as prioritization of parameters, frequency of testing and type of analyses (WHO 2018a). Valuable information on (drinking) water can be gained using field testing kits with a minimum set of parameters, especially in remote areas (WHO 2012). In Mozambique (chapter 2), Ethiopia (chapter 6) and Serbia (chapter 7), drinking water was tested for the so called core parameters, *E. coli*, pH, turbidity and residual chlorine. In Ethiopia, we extended the monitoring of the drinking water supplies step by step by introducing new chemical analyses, and pathogen detection was only implemented at the national level due to capacity and available equipment. Mandatory standards should also allow for incremental improvement, for example the acceptable level of certain parameters can be made more strict over time or the amount of parameters can be increased (WHO 2019). To better fit with the local needs, monitoring parameters should be selected which are relevant for a specific area. This can be done based on risk assessment (WHO 2019). The European drinking water directive allows the list of monitoring parameters to be changed based on the outcome of the risk assessment of the drinking water supply system (EU 2020). Risk assessment, such as QMRA, helps the design of microbial water quality monitoring (Smeets *et al.*, 2010; WHO 2019).

In addition to an enabling environment and prioritizing parameters, implementation and carrying out risk assessment, risk management and water quality monitoring requires **capacity** (Peletz *et al.*, 2018; Ferrero *et al.*, 2019; Kirschke *et al.*, 2020). This includes aspects related to human capacity, funding and the availability of technical equipment (Peletz *et al.*, 2018; Ferrero *et al.*, 2019). Most institutions (water suppliers and surveillance agencies) reported on human capacity, such as knowledge and experience in water quality testing (Peletz *et al.,* 2018). However, funding and human capacity for consistent operational and surveillance monitoring of rural and informal urban supplies were limited (Rahman *et al.,* 2011). The drinking water suppliers described in chapters 2, 5 and 7, had staff members with dedicated responsibilities carrying out water quality testing. However, community managed drinking water supplies and sanitation services had less human capacity and mainly relied on surveillance agencies such as public health institutes (chapter 7). To preserve knowledge, it is important that staff turnover is low (Peletz *et al.*, 2018). In Ethiopia and Mozambique, staff turnover was very low for water quality monitoring (chapters 2 and 6). However, in case of higher turnover this had a noticeable negative impact on implementation of water safety planning, because knowledge was lost (chapter 6). Capacity building for water safety planning is a general requirement for ongoing sustainability of local safe water supplies (Ferrero *et al.,* 2019). Apart from that, technical equipment is important for carrying out water quality monitoring and risk assessment. However, for water quality monitoring of pathogens, sensitive analytical tools are either not available, or their applicability is limited. For example, because of technical complexity, the need to develop analytical proficiency, high costs and time taken to obtain results (WHO 2017c). In Ethiopia for example, not all materials could be purchased for monitoring of pathogens, such as *Cryptosporidium* and enteroviruses. Even for more common and simple methods used in operational monitoring, availability of materials was challenging (chapter 6). Another challenge for water quality monitoring was the availability of a vehicle for sampling of the distribution network, which was also described by Peletz *et al.* (2016). Limited availability of vehicles meant that it was challenging to collect samples for monitoring and to conduct field visits for confirming the system description from source to tap and identifying possible risks (chapter 6).

Risk management approaches for safe drinking water and sanitation should be future-proof

Drinking water and sanitation can play a role in spreading emerging threats, such as antimicrobial resistance or micropollutants. This depends to a higher or larger extent on local settings. Emerging challenges, such as climate change, urbanization and emerging threats, should be incorporated in the existing risk assessment and risk management approaches to ensure resilience to these challenges. Risk assessment and risk management approaches are continuous processes and should be reviewed and revised on regular basis taking into account emerging threats (WHO 2022e, 2023a). This will result in continued provision of safe drinking water and sanitation services, now and in the future. The following paragraphs describe how risk management approaches (such as WSP and SSP) could be able to deal with climate change, antimicrobial resistance and vector-borne diseases as examples of emerging challenges.

Climate change

Climate change has become one of the most significant global challenges that compromises drinking water supply and sanitation services posing a public health threat (Semenza and Paz, 2021). Climate change is expected to alter the frequency and severity of extreme weather events with possible consequences for the safety of drinking water supplies. The effects of climate variability and change also affect the operation of sanitation systems and can increase associated health risks and environmental contamination (Howard *et al.*, 2016; WHO 2022a). Waterborne infections pose an increased risk due to global warming and progressive climate change (Dupke *et al.*, 2023). For example, increases in drought frequency and/or duration potentially alter *Vibrio cholerae* outbreaks in future, potentially increasing the cholera burden when countermeasures such as improved sanitation infrastructure are lacking (Charnley *et al.*, 2022). Currently, a multi-country outbreak of cholera is ongoing with 29 countries having reported cholera cases in 2023 (WHO 2023b). In Mozambique, we found *Vibrio cholera* in surface water used for the production of drinking water, treated water and at households which may lead to more health risks in future due to climate change effects (chapter 3). In 2017, WHO published climate-resilient water safety plans: managing health risks associated with climate variability and change. In Ethiopia, implementation of climate-resilient water safety plans was done in two water supplies (chapter 3). However, addressing climate change requires new skill sets, data origination from additional sources and external expertise to understand how climate aspects are expected to change. Global experiences and case studies from Ethiopian urban supplies provide information on how aspects of climate change are included into water safety planning (Rickert *et al.*, 2019). In order to facilitate taking a holistic approach that considers not only the water supply but also sanitation, guidance is needed on how to consider the climate change effects. For example, the revised WSP and SSP guidance integrated considerations of climate variability and climate change and how these can be managed (WHO 2022e, 2023a). The Protocol on Water and Health focuses on establishing climate-resilient water and sanitation services and strengthening climate considerations in water and sanitation policy making in the pan-European region (UNECE and WHO EURO, 2021). International action to enhance resilience to and mitigate climate change for drinking water and sanitation services is a focus area in the Dutch Global Health Strategy (MoFA and MoH, 2022). This shows that the attention for management of climate change impacts on drinking water and sanitation systems is increasing, and that adaptation to climate change effects is needed. In addition to climate change itself, climate mitigation and adaptation measures may influence drinking water supply and sanitation services.

Antibiotic resistance

Globally, there are increasing reports of antibiotic resistant bacteria, not only restricted to clinical settings but also from environmental samples, especially water (Huijbers *et al.*, 2015). It is suggested that lack of sanitation might be more closely related to antimicrobial resistance than to the reported use of antimicrobials (Hendriksen *et al.*, 2019; WHO, FAO, UNEP, WOAH, 2023). However, the role of the environment as a reservoir of antimicrobial resistance is not fully understood (WHO, FAO, UNEP, WOAH, 2023). Wastewater treatment plants are hotspots for resistance development as well as a source for dissemination of resistance genes and antibiotic resistant bacteria (Bengtsson-Palme *et al.*, 2016). Wastewater treatment plants play a role in mitigating antimicrobial resistance (Nguyen *et al.*, 2021). Specific wastewater treatment processes remove antibiotic resistance which demonstrates the need to improve treatment, in order to limit the emergence and spread of antibiotic resistance (Blaak *et al.,* 2015; Ramalho *et al.,* 2022). However, more information is needed on the relation between antibiotic resistance genes and pathogenic species, and assessing removal efficiency of genes in wastewater treatment plants (Nguyen *et al.*, 2021). In Mozambique, antibiotic resistant bacteria were detected in inlet, treated and household water in a small drinking water supply spreading antibiotic resistant bacteria within the population (chapter 3). To reduce the transmission of antibiotic resistant bacteria via water, measures need to be taken to prevent contamination of surface and drinking water with antibiotic resistant bacteria, to eliminate contamination and to routinely test the water for antibiotic resistant bacteria (Coleman *et al.*, 2013). Antibiotic resistant bacteria can be reduced by advanced wastewater treatment processes such as ozone, UV, ultrafiltration, and chlorination or by treatment of manure (Huijbers *et al.*, 2015). Existing management approaches, such as SSP and WSP, should include the risks of and possible interventions for antibiotic resistant bacteria and more broadly for antimicrobial resistance including viruses and fungi.

Vector-borne diseases

Globally, vector-borne diseases account for over 17% of all infectious diseases (WHO 2017b). Various vectors are associated with the aquatic environment, which provides the habitat for breeding (e.g. mosquitoes and blackflies) or for their entire lifecycle (e.g. snails and water fleas). Drinking water and sanitation may render health risks of viral diseases by mosquitos, for example due to unplanned urbanization, lack of reliable piped water supply and inadequate solid waste or excreta management (WHO 2017b). Guidelines on drinking water and sanitation largely focus on risk management for diarrheal diseases and hardly or not at all on issues related to the aquatic ecological requirements of vectors, or the conditions conducive to intermediate hosts (WHO 2017c, 2018b). Because of the possible health risks due to vector-borne diseases, it is advisable to include vector control experts in risk management approaches for drinking water and sanitation to better understand the possible health risks of vectors and how to prevent and control this (Overgaard *et al.*, 2021).

Future perspective in managing microbial risks

One Water approach

The integrated iWSSP approach can be extended to incorporate other risk assessment and risk management approaches for all other local water applications, so-called One Water approach (Mukheibir *et al.*, 2014), or at least information between risk assessment and risk management approaches should be shared as they influence one another (see Figure 8.2). The One Water approach describes water as a circular and integrated resource. Instead of managing the different applications of water separately, the One Water approach encourages managing all water in an integrated way to secure a healthy and sustainable future for local and global communities (Mukheibir *et al.*, 2014). Water sources may be used for multiple purposes. The pressure on some sources may increase due to water scarcity in future. Intervention is needed as one or two applications of water (sources) can impair others or promote risks.

For other water applications, such as shellfish production and recreation, risk assessment and risk management approaches are also described in guidance documents (WHO 2010, 2021b). For priority bathing sites, recreational water safety plans should be developed and implemented (WHO 2021b). Monitoring and surveillance activities are fundamental risk management components to understanding the human health risks associated with contaminated shellfish (WHO 2010). In Europe, the EU Bathing Water Directive (2006/7/EC) introduces bathing water profiles, including the identification of potential risks and risk management (EU 2006a). The EC Shellfish Waters Directive (2006/113/EC) describes sanitary surveys as an approach to inventory possible risks likely to be a source of contamination of the production area (EU 2006b). Related to the EU Directive, regulation 854/2004 highlights the necessity for competent authorities to establish microbiological monitoring programs for shellfish harvesting areas, including carrying out sanitary surveys (EU 2004).

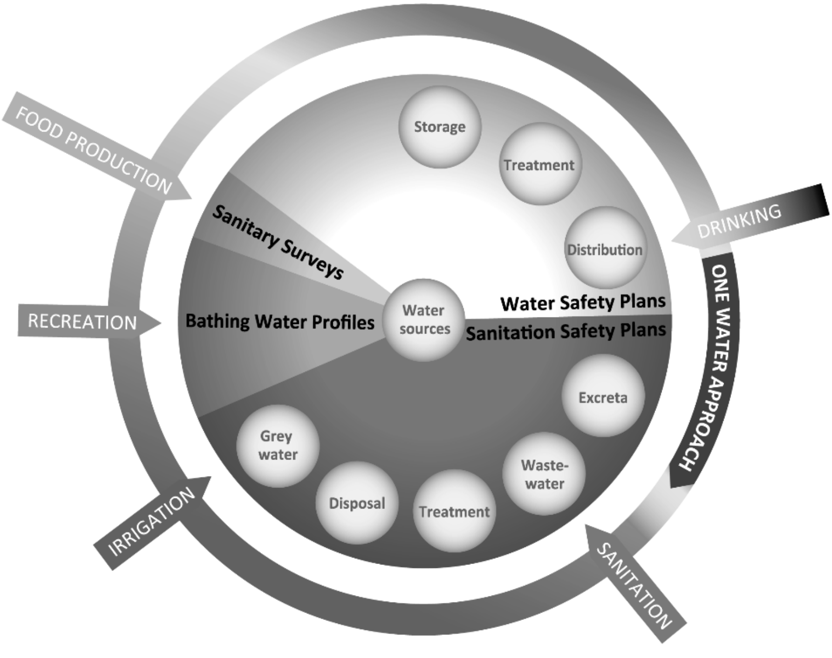

Figure 8.2. *One Water approach containing multiple risk assessment and management approaches for different water applications further complementing the integrated water and sanitation safety plan approach.*

When treated wastewater is reused for irrigation purposes, the competent authority shall ensure that a water reuse risk management plan is established for producing, supplying and using reclaimed water (EU 2022b). Risk assessment and risk management approaches are used to ensure food safety. These risk assessments take into account the water source and the quality used for the food production (FAO and WHO 2007, 2014). FAO and WHO (2019, 2021b) developed guidance on the microbiological criteria and parameters to determine if water can be used for food production and processing. This guidance describes the use of water, varying from wastewater to drinking water, for food production based on the purpose, so-called ‘fit-for-purpose’ principal (FAO and WHO 2019, 2021b). Water scarcity is one of the growing concerns and might result in an increase of reuse of treated wastewater to solve the issues of the water crisis (Kesari *et al.*, 2021). This might also include re-use of wastewater for different applications, such as bathing water or drinking water. The One Water approach will aid in protecting the public health from waterborne pathogens, considering all possible water systems (Fitzmorris-Brisolara *et al.*, 2022).

Extending the use of monitoring data for risk assessment and risk management

Many purposes of monitoring are available providing an enormous amount of water quality data. The use of water quality data could be improved and extended to better support risk assessment and risk management.

In many countries and laboratories, water quality monitoring is carried out to test whether the water quality is in compliance with legislation, and data is stored in logbooks and mainly used for this purpose. This is what we also observed in the laboratories in Mozambique (chapter 2) and Ethiopia (chapter 6). Water quality monitoring data may encompass an enormous additional amount of information. If this data is not shared or transferred to other domains, it will hardly or even never be analyzed or used to support risk assessment. For example, by carrying out trend analyses of the water quality data, information will be gathered on several parameters over the years and seasonal variations (Kostyla *et al.,* 2015). Furthermore, identified trends or fluctuations can be used in risk assessment and risk management and consequently support preventing future exceedances of the threshold values. In future, it might be worth investigating the use of artificial intelligence or machine learning to support trend analysis of water quality data and predict drinkability, as was shown for groundwater by Panigrahi *et al.* (2023).

Research can be done for all different monitoring purposes. It is important that researchers involve stakeholders in the research set-up and the outcomes of the research. In Mozambique (chapter 3) and Ethiopia (chapter 6), results of pathogen detection were shared with the drinking water utilities to understand the risks and act on them. In Mozambique (chapter 2), research was based on the most effective way of chlorination and intermittent supply. Based on the results of this research project, the drinking water utility adapted their approach to increase the drinking water quality. In the Netherlands, an example of proactive governance was shown even before data was gathered. Relevant stakeholders were involved in the set-up of the research and positive *Legionella* results at wastewater treatment plants during the study were directly communicated for follow-up with the owner and it was investigated whether additional control measures are required to protect public health (chapter 5). Scientific studies are relevant for regulatory actions, and to limit the time between research and regulatory actions a semi-automated methodology using literature mining was developed to identify the first scientific study which reports the presence of a contaminant in the aquatic environment (Hartmann *et al.*, 2019). This approach could also be applied for risk assessment by identifying risks of possible threats.

Not only water quality data, but also monitoring data from other sectors, can provide essential information for risk assessment and risk management for drinking water supply and sanitation. Examples are food, human and environmental surveillance. **Food surveillance** data together with product and process evaluations can aid the identification of hazard–food combinations (FAO and WHO, 2021a). Information on specific hazards occurring in food may be related to water used in the production (FAO and WHO, 2019), and provides information on this water source which might be relevant for other water applications, such as drinking water production. Younger *et al.* (2022) showed that sewage overflows occur frequently in England and impact the shellfish waters. Detection of pathogens in humans (**human surveillance**) can be used as an early warning system for drinking water supply and sanitation services to identify their role in possible transmission. In resource-limited areas, detection of *Vibrio cholerae* is frequently based on clinical signs and symptoms, and can also be diagnosed using laboratory testing in human stool samples (Chowdhury *et al.*, 2022). More recently, during the COVID-19 pandemic risk assessments were applied to identify the role of drinking water and sanitation in transmitting SARS-CoV-2 after the virus was detected in humans (Bogler *et al.*, 2020; WHO 2020b). The COVID-19 pandemic showed that **environmental surveillance**, in this case wastewater surveillance, for an emerging pathogen can be set up relatively quickly (WHO 2022b). Wastewater surveillance is not new and already existed for other pathogens, such as poliovirus and antimicrobial resistance. SARS-CoV-2 surveillance in wastewater could be a sensitive surveillance system and early warning tool (Lodder and De Roda Husman, 2020), and therewith provide information for identifying risks related to drinking water and sanitation. Furthermore wastewater surveillance can provide very cost-effective data to complement clinical surveillance, especially in surveillance limited regions (WHO 2022b).

To make use of the data from different monitoring or surveillance systems in an efficient way, data gathering and analyses needs to be harmonized and proactive governance would be beneficial.

Reduction of waterborne infectious diseases by risk management

This thesis focused on water quality monitoring, risk assessment and risk management for drinking water supply and sanitation systems. The different studies contributed to the limitation of exposure of people in different countries to unsafe drinking water or unsafe sanitation practices and therefore support public health protection. In this thesis, we did not investigate to what extent risk management reduced the transmission of waterborne infectious diseases. Although it is known that improving service levels towards safely managed drinking water or sanitation can dramatically improve health by reducing diarrheal disease deaths, future research on how risk management contributes to reduce waterborne infectious diseases is needed.

Conclusions

The main conclusion from this thesis is that both risk assessment and water quality monitoring contribute to risk prevention and therefore should be well embedded in the risk management approach. It is recommended that water quality monitoring and risk assessment should be better combined to strengthen both risk management and its implementation. A risk assessment should form the fundament for the selection of water quality parameters, the frequency of the analyses and the method of analysis. Extending the use of water quality monitoring data, derived from the various types of monitoring applied, could better support risk assessment and risk management, and thereby further reduce infectious disease outbreaks in the future.

It is concluded that integration or combination of risk management and risk assessment approaches is possible and useful. This can be done within the One Water approach, such as drinking water or sanitation, but also across additional water applications. Risk management for drinking water or sanitation services needs not be limited to one approach as other methodologies can be complementary. If multiple risk management or risk assessment approaches are used for drinking water or sanitation services, it is important that data are shared between the different methodologies and the approaches are reviewed regularly to be up-to-date. The regular reviews should take into account new challenges and the risks associated with them including how to manage these risks. In this way, health risks related to drinking water and sanitation now and in the future could be addressed proactively. In this thesis, risk management for drinking water and sanitation services were integrated into one approach (iWSSP). In the future, water scarcity and extreme weather events will be an increasing problem on every continent due to climate change and a growing population. Available water is needed for various purposes, such as drinking water supply, irrigation water, and recreation. There will be a competition between all these purposes of water. To be climate resilient and prepared for a growing population, an integrated and inclusive approach to the management of available water should be adopted.

Finally, to improve the implementation and application of risk management in different settings in order to improve public health the gap between legislation and practice should be closed. This thesis showed that although many international and national laws, frameworks and protocols, regulations and guidance exist to ensure access to safely managed drinking water and sanitation services, local needs could be better addressed and challenges in different areas should be taken into account. For water quality monitoring a selection should be made based on the resources and local needs for parameters, frequency of testing and type of analyses. Local guidance, tools and support from experts are needed to implement risk management as well as adaptation of general guidance to national frameworks

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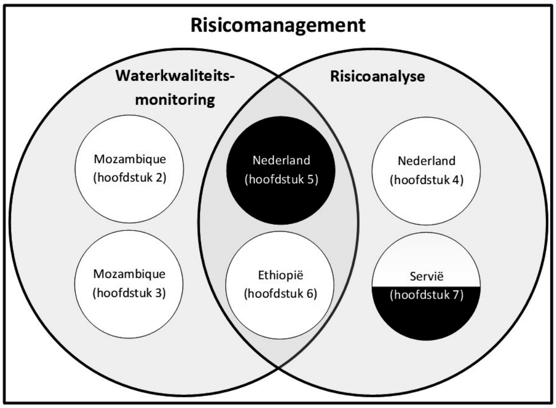
Samenvatting

Summary in Dutch

Besmet drinkwater en het ontbreken van goede sanitaire voorzieningen zijn belangrijke oorzaken voor de verspreiding van wateroverdraagbare infectieziekten. Veel factoren, zoals verouderde infrastructuur, verstedelijking en klimaatverandering, zorgen ervoor dat de toegang tot veilig drinkwater en sanitaire voorzieningen wordt bedreigd. Leveranciers van drinkwater en sanitaire voorzieningen willen gevaren en de bijbehorende risico's herkennen en begrijpen. Met die informatie kunnen ze de risico's beter onder controle houden. Risicomanagement is belangrijk voor veilig drinkwater en sanitaire voorzieningen door op een systematische manier risico's in kaart te brengen en te beheersen. Risicomanagement omvat een risicoanalyse en monitoring van de waterkwaliteit. Met behulp van risicoanalyse kunnen microbiële gevaren worden geïdentificeerd en risico's worden geschat. Op basis daarvan kunnen infectieziekten worden voorkomen en helpt risicoanalyse de volksgezondheid te beschermen. Monitoring van de kwaliteit van drinkwater en afvalwater is belangrijk door het (vroegtijdig) signaleren van gevaren zodat hierop proactief kan worden gehandeld. Hierdoor draagt waterkwaliteitsmonitoring bij aan veilig drinkwater en sanitaire voorzieningen.

Dit proefschrift richt zich op risicomanagement voor drinkwater en sanitaire voorzieningen in verschillende landen met als doel wateroverdraagbare infectieziekten te verminderen (Figuur S.1). Het algehele doel van dit proefschrift is om waterkwaliteitsmonitoring en risicoanalyse beter te integreren in risicomanagement. Een ander doel is om te onderzoeken in hoeverre verschillende risicomanagementbenaderingen kunnen worden gecombineerd voor drinkwater en/of sanitaire voorzieningen. Tenslotte wordt onderzocht in hoeverre risicomanagement bestendigheid kan bieden tegen toekomstige veranderingen, zoals klimaatverandering en verstedelijking.



Figuur S.1. *Overzicht van de landen (hoofdstukken) waarin risicoanalyse en waterkwaliteitsmonitoring als onderdeel van risicomanagement zijn uitgevoerd voor drinkwater (witte cirkels) en sanitaire voorzieningen (zwarte cirkels).*

Waterkwaliteitsmonitoring werd uitgevoerd bij een klein drinkwatersysteem in Mozambique (**hoofdstukken 2 en 3**). De impact van veranderingen in operationele strategieën, zoals hogere chloordosering, aanpassing aan de leveringsduur en *first-flush*, op de microbiologische waterkwaliteit wordt beschreven in **hoofdstuk 2**. Op basis van de resultaten van monitoring van de waterkwaliteit werd de beste strategie bepaald. Door de chloordosering te verhogen verbeterde de microbiologische drinkwaterkwaliteit, terwijl een aangepaste leveringsduur en first-flush geen duidelijk effect lieten zien. Het aantonen van *E. coli* in het drinkwater op huishoudelijk niveau kan wijzen op een herbesmetting in het distributiesysteem of op onveilige hygiënische handelingen.

Het onderzoek naar de microbiologische kwaliteit van het geleverde drinkwater wordt beschreven in **hoofdstuk 3**. Watermonsters van bron tot kraan werden geanalyseerd op de aanwezigheid van fecale indicatoren (*E. coli*), pathogenen (*Vibrio cholerae*, *Salmonella* spp. en *Campylobacter* spp.) en antibioticaresistentie (cefotaxime resistente *E. coli*). In 79 procent van de monsters werd na de zuivering geen *E. coli* meer aangetoond. Naast *E. coli* werd in het gezuiverde water *Campylobacter* (10 procent) en *Vibrio cholerae* (11 procent) aangetoond. In het distributiesysteem namen de aantallen positieve monsters toe, vermoedelijk door herbesmetting. Fecale verontreiniging en pathogenen in het drinkwatersysteem wijzen op een gezondheidsrisico.

Er bestaan veel verschillende benaderingen voor risicoanalyse en -management. Voordat een nieuwe aanpak wordt ingevoerd is het zinvol om te onderzoeken welke bestaande methoden worden toegepast, hoe informatie tussen verschillende methoden kan worden gedeeld en of het nuttig is om een nieuwe aanpak in te voeren. In plaats van een waterveiligheidsplan (WSP) als een nieuwe aanpak bij drinkwaterbedrijven in Nederland te introduceren werd onderzoek gedaan naar bestaande activiteiten voor risicoanalyse en -management. Zo kon worden bepaald in hoeverre deze activiteiten de elf elementen van een WSP dekken (**hoofdstuk 4**). Door de zes, in Nederland, wettelijk vereiste methoden te gebruiken, worden alle elementen van een WSP gedekt. Deze wettelijke vereisten worden door de drinkwaterbedrijven nog aangevuld met activiteiten op sector- en waterbedrijfsniveau, zoals hygiënecodes en standaard operationele procedures. Bij gebruik van meerdere risicoanalyse- en risicomanagementmethoden is het van belang om informatie te delen en te combineren binnen de verschillende methoden. Hoe geïdentificeerde (microbiologische) risico’s uit verschillende risicoanalyses met elkaar kunnen worden geïnterpreteerd en geprioriteerd moet nog worden onderzocht.

In **hoofdstuk 5** wordt een inventarisatie beschreven van alle communale en industriële afvalwaterzuiveringsinstallaties (AWZI’s) in Nederland met hun relevante kenmerken. Daartoe werd een risicomatrix opgesteld op basis van type zuivering, temperatuur van het proceswater, beluchting en type industrie. De risicomatrix werd gebruikt om te bepalen welke AWZI's een middelmatig tot hoog risico op legionellavermeerdering en -verspreiding hebben. Deze AWZI's werden vervolgens onderzocht op de aanwezigheid van *Legionella*. Van de bemonsterde AWZI's bleek dat 18 procent positief was voor *Legionella* spp. Wanneer *Legionella* in het afvalwater werd aangetroffen, werd door de verantwoordelijke autoriteit direct contact opgenomen met de eigenaar om de uitstoot van *Legionella* te verminderen om de volksgezondheid te beschermen. Dit gebeurde bijvoorbeeld door beluchtingstanks te overdekken. Deze studie heeft ook bijgedragen aan de ontwikkeling van een handreiking om eigenaren van AWZI's te ondersteunen bij het identificeren, interpreteren en beheersen van risico's op legionellavermeerdering en -verspreiding. Ook heeft het Ministerie van Infrastructuur en Watermanagement het opgepakt om op te nemen in Nederlandse regelgeving.

Klimaatverandering zet de beschikbaarheid, kwaliteit en veiligheid van drinkwater in Ethiopië verder onder druk. Een klimaatbestendig WSP is bij twee grote drinkwaterbedrijven in Ethiopië geïmplementeerd (**hoofdstuk 6**). Om een dergelijk WSP beter te ondersteunen is in deze studie de waterkwaliteitsmonitoring uitgebreid bij de drinkwaterbedrijven en bij het nationaal instituut voor volksgezondheid. Op basis van mogelijke risico's werden parameters geprioriteerd voor waterkwaliteitsmonitoring. Denk aan het meten van *Cryptosporidium*, fluoride en arseen in ruw water om inzicht te krijgen naar de ernst van het risico. Monitoring van *E. coli* en vrij chloor in het distributienetwerk toonde aan in welke mate risico's werden beheerst, zoals herbesmetting in het distributienetwerk of onvoldoende chloreren. Door trends in waterkwaliteitsdata te analyseren werden de risico's in de loop der jaren, maar ook seizoenschommelingen, inzichtelijk. Door waterkwaliteitsmonitoring en WSP continue te koppelen, konden de verkregen data beter worden gebruikt. Dit moet nog verder worden ingevoerd.

Wanneer veiligheidsplanning voor drinkwater en sanitaire voorzieningen in kleinschalige systemen los van elkaar wordt gebruikt, kan dat een uitdaging zijn als menselijke, financiële en administratieve middelen beperkt zijn. Daarom is een geïntegreerde aanpak voor water- en sanitatieveiligheidsplannen ontwikkeld (iWSSP) (**hoofdstuk 7**). Ook werden tools en trainingsmateriaal ontwikkeld om deze nieuwe aanpak in de praktijk in te voeren. Deze geïntegreerde aanpak werd getest in drie kleine systemen in landelijk Servië. Door iWSSP op deze drie locaties in te voeren, is de kennis van de systemen voor drinkwater en sanitatie vergroot bij de betrokken personen. De ervaringen voor capaciteitsopbouw op het gebied van waterkwaliteitsmonitoring, risicoanalyse en risicomanagement in Ethiopië en Servië worden beschreven in de **hoofdstukken 6 en 7**. Hoewel er misschien wetgeving is, was er behoefte aan een betere aansluiting op de lokale context in de vorm van begeleiding, capaciteitsopbouw en instrumenten. Dat gold vooral in kleine systemen.

In **hoofdstuk 8** worden de resultaten bediscussieerd en toekomstperspectieven gegeven. De studies in dit proefschrift hebben eraan bijgedragen dat mensen in verschillende landen minder aan ziekteverwekkers zijn blootgesteld via onveilig drinkwater of sanitaire voorzieningen. De studies helpen daarom de volksgezondheid te beschermen. In dit proefschrift hebben we de mate waarin wateroverdraagbare infectieziekten worden verminderd door risicomanagement niet onderzocht. Er bleek nog ruimte te zijn om de implementatie en toepassing van risicomanagement te verbeteren. Ten eerste, zijn er veel internationale en nationale regelgeving en richtlijnen voor risicomanagement om veilig drinkwater en sanitaire voorzieningen te garanderen. Om ervoor te zorgen dat risicomanagement zowel in stedelijk als landelijk gebied wordt gebruikt, en ook in alle landen, moet de kloof tussen wetgeving en praktijk zo klein mogelijk zijn. Het is daarbij belangrijk rekening te houden met de lokale behoeften en de uitdagingen in verschillende gebieden. Zo kan voor monitoring van de waterkwaliteit een selectie worden gemaakt op basis van de middelen en de lokale behoeften. Ten tweede kunnen risicoanalyse en waterkwaliteitsmonitoring beter met elkaar worden verbonden. Een risicoanalyse vormt de basis voor de selectie van parameters, de frequentie van de analyses en de analysemethode. Door de waterkwaliteit te monitoren worden gegevens verkregen voor de risicoanalyse en risicomanagement.

Uit de thesis blijkt dat zowel risicoanalyse als waterkwaliteitsmonitoring bijdragen aan het voorkomen van microbiële risico’s. Beide moeten daarom goed worden ingebed in risicomanagement. Het is mogelijk om aanpakken voor risicomanagement met elkaar te combineren of zelfs te integreren. Dit kan op het gebied van één doeleinde van water, zoals drinkwater of sanitatie, of voor meerdere. Voor drinkwater of sanitaire voorzieningen hoeft risicomanagement niet beperkt te blijven tot één benadering, aangezien verschillende aanpakken elkaar kunnen aanvullen. Als er meerdere benaderingen zijn, is het van belang dat gegevens tussen de benaderingen worden gedeeld en de aanpakken regelmatig worden bijgewerkt. Op deze manier kunnen nieuwe uitdagingen en risico’s worden geïdentificeerd en beheerst. Hierdoor worden gezondheidsrisico’s gerelateerd aan drinkwater en sanitaire voorzieningen zo klein mogelijk, nu en in de toekomst. In dit proefschrift is risicomanagement voor drinkwater en sanitaire voorzieningen geïntegreerd in één aanpak (iWSSP). In de toekomst zal op elk continent de waterschaarste toenemen onder andere door klimaatverandering en een groeiende bevolking. Om weerbaar te zijn tegen klimaatverandering en een groeiende bevolking zal iWSSP in de toekomst kunnen worden uitgebreid met risicomanagement aanpakken voor andere doeleinden van water dan drinkwater en sanitatie. Afhankelijk van de kwaliteit en mogelijke gezondheidsrisico’s kan het water zo goed en efficiënt mogelijk worden gebruikt (*fit-for-purpose*).

Summary

Unsafe drinking water and sanitation are major causes of the spread of waterborne diseases in a community. Drinking water supply and sanitation services face threats, such as aging infrastructure, urbanization and climate changes, which may affect the transmission of microbial hazards. Drinking water suppliers and sanitation providers need to know and understand these hazards and the associated risks these hazards pose to the drinking water supply and sanitation system and manage these risks well. To this extent, risk management is crucial for the identification and management of risks in a structured and continuous way. Risk management here includes risk assessment and water quality monitoring. In risk assessment, microbial hazards can be identified and risks can be estimated. Risk assessment can be used to develop control measures to reduces the risks. This may result in prevention of diseases, thereby contributing to public health protection. Monitoring the quality of drinking water and wastewater are key elements to ensure drinking water and sanitation safety by providing (early) signals of hazards for remediate actions.

This thesis will focus on risk management for drinking water supply and sanitation systems to prevent waterborne infectious diseases in different countries and resource settings (see Figure S.2). The main objective of this thesis is to better integrate water quality monitoring and risk assessment into risk management approaches. Another objective is to integrate risk management approaches for drinking water supply and/or sanitation services in order to reduce waterborne infectious diseases. The final objective is to investigate to which extent risk management methods create resilience to future changes, such as climate change and urbanization.

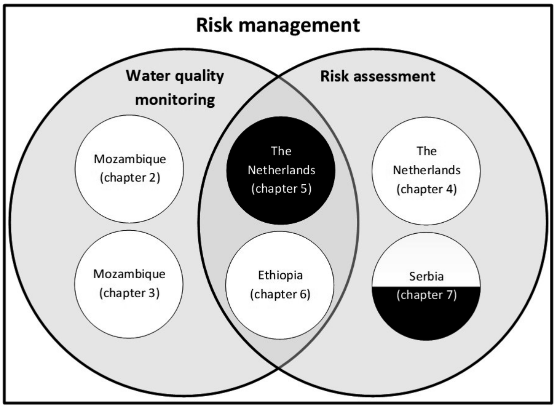


Figure S.2. *Overview of the countries (chapters) in which risk assessment and water quality monitoring as part of risk management were addressed for drinking water supply (white circles) and sanitation services (black circles).*

Water quality monitoring is an important tool to identify possible risks and validate the effectiveness of control measures. In Mozambique, water quality monitoring was carried out in a small drinking water supply (**chapter 2** and **chapter 3**). The impact of changes in operational strategies, namely increased chlorine dosage, increased supply duration and first-flush, on the microbial water quality was investigated in **chapter 2**. Based on the results of water quality monitoring, the best strategy could be determined. Increasing chlorine dosage ensured good microbiological drinking water quality but changing the number of supply cycles had no such effect. The detection of *E. coli* contamination in drinking water at household level could point at recontamination in the distribution or unsafe hygienic practices.

The microbiological quality of drinking water supplied was assessed by analyzing different water samples for the presence of fecal indicators (*E. coli*) and pathogens (*Vibrio cholerae*, *Salmonella* spp. and *Campylobacter* spp.) and an antibiotic resistance determinant (cefotaxime resistant *E. coli*) (**chapter 3**). No *E .coli* could be detected in 79% of the samples after treatment. Besides *E .coli*, the pathogens *Campylobacter* (10%) and *Vibrio cholerae* (11%) were detected in the treated water. In the distribution system, the number of positive samples increased, presumably due to recontamination. The presence of fecal contamination and pathogens in the drinking water system indicates a health risk.

There are many different risk assessment and risk management approaches. Before implementing a new approach it is essential to start by identifying existing approaches, investigating how information can be shared and if it would be beneficial to implement a new one. Instead of implementing a water safety plan (WSP) as a new approach for drinking water utilities in the Netherlands, existing risk assessment and risk management approaches were identified (**chapter 4**). The results showed that the six legally required risk assessment and risk management approaches cover the eleven elements of the WHO WSP approach. These legal requirements are complemented by additional activities at sector and water company level such as codes of practice and standard operating procedures. The outcomes of all approaches and activities combined provide information from source to tap. Nevertheless, when using multiple risk assessment and risk management approaches it is crucial to share and combine information derived from the different activities.

An inventory of all communal and industrial wastewater treatment plants (WWTPs) with their characteristics was carried out in the Netherlands (**chapter 5**). In this study, a risk matrix was drafted to assess wastewater treatment plants at risk for *Legionella* growth and emission based on the risk criteria type of treatment, temperature of process water, aeration and type of industry. The risk matrix was applied to assess the risk of the identified wastewater treatment plants. Analyzing wastewater collected at WWTPs with moderate to high risk for *Legionella* growth and emission showed that 18% of the sampled WWTPs were positive for *Legionella* spp. If *Legionella* was detected in the wastewater, the responsible authority directly contacted the owner to reduce emission, for example by covering aeration tanks, and protect human health. The work also contributed to the development of guidance to support owners of wastewater treatment plants to identify, interpret, and control risk for *Legionella* growth and emission. Furthermore, the Dutch Ministry of Infrastructure and Water Management is developing legislation for *Legionella* prevention from WWTPs.

Climate change puts intense pressure on the availability and quality of water. A climate resilient WSP was implemented in the urban drinking water supplies of Addis Ababa and Adama in Ethiopia (**chapter 6**). In this study, water quality monitoring was extended at the utilities and at the national level to support the WSP. Based on the risks identified with the WSP, water quality monitoring was optimized by prioritizing parameters. Examples are measuring *Cryptosporidium*, fluoride and arsenic in raw water to provide information on the severity of the risks. Monitoring *E. coli* and free chlorine in the distribution network, showed to which extent the risks of recontamination in the distribution network or insufficient chlorination, were controlled. Implementing trend analyses on existing water quality data provided insight on risks over the years, as well as seasonal fluctuations. By continuously linking water quality monitoring and climate resilient WSP, utilization of the collected data was optimized, and both approaches benefit from linking these activities.

Applying risk management approaches for drinking water and sanitation separately might be challenging for small systems or communities with limited human, financial and administrative resources. An integrated water and sanitation safety planning (iWSSP) approach was developed together with guidance and training material for practical application of this novel approach (**chapter 7**). The integrated approach was piloted in three small systems in rural Serbia. Implementing iWSSP at the pilot sites contributed to a better understanding of both drinking water supply and sanitation system. It also resulted in increased awareness, knowledge and understanding among staff of drinking water supply and sanitation services. The experiences for capacity building on water quality monitoring, risk assessment and risk management in Ethiopia and Serbia are described in **chapters 6 and 7**. Even though legislation might be in place, better connection to the local context was needed in the form of guidance, capacity building and tools. This was especially the case in small settings.

In **chapter 8** the results are discussed and future perspectives are given. The different studies in this thesis contributed to limit exposure of people in different countries to unsafe drinking water or unsafe sanitation practices and therefore support public health protection. In this thesis, we did not investigate to what extent risk management, including risk assessment and water quality monitoring, reduced waterborne infectious diseases. It has become clear that there is still room to improve the implementation and application of risk management in different settings. Firstly, many international and national regulations and risk management guidelines exist to guarantee safe drinking water and sanitation services. To ensure that risk management is applied both urban and rural, as well as in all countries, it is necessary to minimize the gap between legislation and practice. Taking into account local needs and challenges in different areas plays an important role. For the monitoring of water quality, a selection can be made based on resources and local needs. Secondly, water quality monitoring and risk assessment can be better linked, to strengthen both. A risk assessment could form the fundament for the selection of parameters, the frequency of the analyses and the method of analysis. Water quality monitoring contributes to safe drinking water and sanitation, which is reinforced by the use of water quality data to improve risk assessment and risk management

The main conclusion from this thesis is that both risk assessment and water quality monitoring contribute to risk prevention and therefore should be well embedded in the risk management approach. It is possible and useful to integrate or combine risk management approaches. This can be done for a single water application, such as drinking water or sanitation, but also for multiple water applications. Risk management for drinking water or sanitation services needs not be limited to one approach as other methodologies can be complementary. If multiple approaches are used, sharing data between the different methodologies and reviewing the approaches regularly to be up-to-date is needed to address and manage new challenges. In this way, health risks related to drinking water and sanitation now and in the future are minimized. In this thesis, risk management for drinking water and sanitation services were integrated into one approach (iWSSP). In the future, water scarcity will be an increasing problem on every continent due to climate change and a growing population. To be resilient to these challenges, an integrated and inclusive approach to the management of available water should be adopted. iWSSP can be extended by integrating or combining risk management for other water applications in order to use the water correctly and efficiently (fit-for-purpose), depending on the quality and possible health risks.

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Curriculum vitae

Harold van den Berg was born on 3 August 1978 in Helmond in the Netherlands. He grew up in Gemert, where he completed secondary school in 1996 at Macropedius College. In 1996, he started to study Biology and Medical Laboratory Research at Fontys Hogescholen for Applied Science and Technology in Eindhoven. After he received his BSc degree in 2000, he started working at the National Institute for Public Health and the Environment (RIVM) at the Microbiological Laboratory for Health Protection (MGB). As a research technician, he worked on the detection of bacteria, viruses and parasites in environmental samples, especially water. Since 2004, he has been a member of the response unit for biological incidents in The Netherlands. In 2007, MGB was renamed Laboratory of Zoonoses and Environmental Microbiology (Z&O). And since that time, his work has become more internationally oriented. He has worked in various international projects for capacity building on water quality monitoring, such as Jordan, Ethiopia, Mozambique, Tajikistan and Surinam. As a member of the WHO Collaborating Centre for 'Risk Assessment of Pathogens in Food and Water', since 2014, his focus is on strengthening national capacity and scaling up the uptake of risk-based management approaches and water quality monitoring. He has contributed to the implementation of water safety plans in several countries, such as Ethiopia, Serbia, Surinam and Bosnia and Herzegovina. Since 2015, he has been a member of the Dutch working group 'Infection Risk for quantitative microbial risk assessment'. In 2018, he officially became researcher at the department Environment of Z&O continuing his work focused on the detection of various waterborne pathogens, risk assessment and risk management for drinking water and sanitation services. In 2021, he became a member of the Dutch Standards Subcommittee 390 020 06 'Environmental quality – Microbiological parameters' and in 2023 he became chair of this subcommittee.

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