



 **Opin vísindi**

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Title/Titill: Water safety plan enhancements with improved drinking water quality detection techniques

Year/Útgáfuár: 2020

Version/Útgáfa: Post-print (lokagerð höfundar)

**Please cite the original version:**

**Vinsamlega vísið til útgefnu greinarinnar:**

Gunnarsdottir, M. J., Gardarsson, S. M., Figueras, M. J., Puigdomènech, C., Juárez, R., Saucedo, G., . . . Hunter, P. (2020). Water safety plan enhancements with improved drinking water quality detection techniques. *Science of The Total Environment*, 698, 134185. doi:<https://doi.org/10.1016/j.scitotenv.2019.134185>

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# Water safety plan enhancements with improved drinking water quality detection techniques

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## Abstract

Drinking water quality has been regulated in most European countries for nearly two decades by the drinking water directive 98/83/EC. The directive is now under revision with the goal of meeting stricter demands for safe water for all citizens, as safe water has been recognized as a human right by the United Nations. An important change to the directive is the implementation of a risk-based approach in all regulated water supplies. The European Union Framework Seventh Programme Aquavalens project has developed several new detection technologies for pathogens and indicators and tested them in water supplies in seven European countries. One of the tasks of the project was to evaluate the impact of these new techniques on water safety and on water safety management. Data were collected on risk factors to water safety for five large supplies in Denmark, Germany, Spain and the UK, and for fifteen small water supplies in Scotland, Portugal and Serbia, via a questionnaire aiming to ascertain risk factors and the stage of implementation of Water Safety Plans, and via site-specific surveys known as Sanitary Site Inspection. Samples were collected from the water supplies from all stages of water production to delivery. Pathogens were detected in around 23% of the 470 samples tested. Fecal contamination was high in raw water and even in treated water at the small supplies. Old infrastructure was considered a challenge at all the water supplies. The results showed that some of the technique, if implemented as part of the water safety management, can detect rapidly the most common waterborne pathogens and fecal pollution indicators and therefore have a great early warning potential; can improve water safety for the consumer; can validate whether mitigation methods are working as intended; and can confirm the quality of the water at source and at the tap.

**Keywords:** Drinking water safety, Water Safety Plan performance, Risk factors in water supply

### 1. Introduction

Drinking water quality in the member states of the European Union and European Economic Area has been regulated by the Council Directive 98/83/EC (DWD) on the quality of water intended for human consumption since 1998. There is a consensus that compliance rates have improved and that it has had a positive effect on public health in Europe (Klaassens et al., 2016). As an example, there has been a significant reduction in the presence of the fecal indicator *E. coli* in drinking water (EC, 2014 & 2016). However, many studies have shown that the water quality at small water supplies is poorer than for large water supplies and information on their status is often lacking (EC, 2014; Beaudeau et al., 2010; Hulsmann, 2005; Pitkänen et al., 2011; Hendry & Akoumianaki, 2016; Gunnarsdottir et al., 2017a; Gunnarsdottir et al., 2016; Gunnarsdottir et al., 2015). Sixty-five million European citizens, or

around 8%, are estimated to be served by small water supplies and two million are without water service (Klaassens et al., 2016; Hulsmann, 2011).

The human right to water and sanitation was recognized by the United Nations General Assembly on July 28, 2010 and is reflected in the new UN Sustainable Development Goals (UN-SDGs) of September 2015. Goal 6 ensures universal access to safe and affordable drinking water for all by 2030 (Resolution 64/292; UN-SDGs, 2015). If the UN-SDGs with the human right to safe water are to be met in Europe, the water safety of the small supplies, that have limited surveillance and poor water quality, needs to be addressed. The first European Citizens Initiative (ECI) Right2Water was conducted in 2013-14, in accordance with the Lisbon Treaty. The ECI urge the European Commission to implement the human right to water into the drinking water directive and ensure that water remains a public service and public good. The ECI was signed by over 1.8 million European citizens across 13-member states.<sup>1</sup>

In 2003, the European Commission started to discuss the key elements that should be modified in the DWD such as current knowledge and advances in technology (Figueras and Borrego, 2010) and has recently published an evaluation report on the performance of the DWD (Klaassens et al., 2016). It was emphasized in the EC evaluation report that in the twenty years that have passed since the directive was written there have been various developments, including technology and identification of new contaminants, that together require updating of the DWD. For example, the implementation of a risk-based approach, such as the Water Safety Plan (WSP), can lead to a faster decision-making process in the case of incidents, which will improve water safety (Bartram et al., 2009; Figueras and Borrego, 2010; Gunnarsdottir et al., 2012a). The report also points out that the use of new methods, such as molecular methods in water quality testing, give results faster and are more sensitive and more specific than the current methods based on culturing. Furthermore, it is emphasized in the report that the implementation of the newly developed information and communication technologies could enhance water quality and performance of services.

A systematic preventive approach for managing risk to water safety, the WSP, is now internationally recognized as an important and modern method for reducing health risk from drinking water. This approach has been advocated by the World Health Organization (WHO) since 2004 and is now used in at least 93 countries around the world. It has also been adopted as policy or a regulatory requirement or being under development as such in 69 countries (WHO/IWA, 2017). This approach aims at shifting surveillance from control at the tap to preventive management for the whole water supply chain. The WSP implementation has been shown to improve drinking water quality and public health as well as being crucial in management (Summerill et al., 2010a & 2010b; Gunnarsdottir et al., 2012a; 2012b; Setty et al., 2017; Roeger & Tavares, 2018). The approach used in some European countries (e.g. Switzerland, Iceland, France, Slovenia, Norway and Sweden) to classify drinking water as food that needs to be protected in a systematic way has been shown to positively change the mindset of people working in the water sector (Baum & Bartram, 2017; Gunnarsdottir et al., 2012b). A recent amendment to the DWD allows reduction of sampling if a risk-based approach is used (EC, 2015). This acknowledges the merit of preventive management, such as WSP, to be included in formal legislation (Baum & Bartram, 2017).

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<sup>1</sup> ECI Right2Water: <http://www.right2water.eu/>.

The microorganisms which can cause waterborne outbreaks are not directly included in the DWD. The DWD only considers indicator parameters, whereas pathogens are only investigated when an outbreak is suspected or occurs. The main regulatory indicators for pathogens currently are the bacteria *E. coli* and Enterococci; both indicate presence of fecal contamination but may not necessarily reflect whether there is a threat to human health. However, other microorganisms such as viruses and parasites may be present in water in the absence of the indicator bacteria and can pose a risk to human health, particularly viruses, parasites and bacteria with very different survival strategies. Survival of pathogens in the environment depends on many factors, such as temperature, acidity and composition of the strata, and these factors are not the same for all classes of pathogens. Parasites live much longer than bacteria in water, and viruses travel longer in the strata, being much smaller in size (Yates et al., 1985; Figueras and Borrego, 2010). For example, in a norovirus outbreak infecting 100 people at a hotel in Northern Iceland in 2004, there were no indicator bacteria found, whereas water samples were registered as very strongly positive for Norovirus (NoV) GII. The cause of the outbreaks was a septic tank situated 80 m from the water well and upstream of groundwater flow to the well (Gunnarsdottir et al., 2013). Therefore, it is important to develop techniques to measure pathogens and suitable indicators instead of relying mostly on indicators of only one class (i.e. bacteria).

A new proposal for revision of the DWD has been recast (Feb. 2018; Oct 2018)<sup>2</sup>. This is a follow-up on the ECI Right2Water initiative. The main changes in the proposal is that all water supplies that provide more than 50 m<sup>3</sup> a day (or 250 people) are to carry out a risk-based approach to water safety; new parameters are added (e.g. *Clostridium perfringens* spores, somatic coliphages, *Legionella*, per-fluorinated and endocrine disrupting compounds); and information on drinking water to consumers is to be increased considerably, using information and communication technology.

The objective of this research was evaluation of the impact of implementation of improved modern detection techniques for pathogens and microbial indicators developed in the EU FP7 Aquavalens project ([www.aquavalens.org](http://www.aquavalens.org)) on drinking water safety and WSP plan management.

## 2. Methods

The methods employed to achieve the objectives of this study used results from work done in the FP7 Aquavalens (AQV) project, mainly in three work packages; WP13 on WSP and water safety, WP10 testing pathogens in large scale water supplies, and WP11 testing pathogens in small water supplies (Gunnarsdottir et al., 2018, 2017b, 2017c; Eglitis et al., 2017; Puigdoménech et al., 2017; Monteiro & Santos, 2017; López-Avilés & Pedley, 2017a & 2017b).

To evaluate water safety data were gathered from the water supplies participating in the project via two questionnaires, results from monitoring over 478 samples with the new technique and verification control with cultural method on the same samples and results site-specific Sanitary Site Inspection (SSI) surveys performed for the small water supplies. For

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<sup>2</sup><http://ec.europa.eu/environment/water/water-drink/>;  
<http://www.europarl.europa.eu/sides/getDoc.do?pubRef=-//EP//TEXT+TA+P8-TA-2018-0397+0+DOC+XML+V0//EN&language=EN> [https://ec.europa.eu/info/law/better-regulation/initiatives/com-2017-753\\_en#initiative-givefeedback](https://ec.europa.eu/info/law/better-regulation/initiatives/com-2017-753_en#initiative-givefeedback)

comparison results from regular surveillance monitoring was also gathered for the sites. Data gathered is shown in Table 1.

**Table 1. Information on questionnaires, sanitary inspection and samples**

	Large water supplies	Small water supplies	Sum
<b>Number of water supplies:</b>			
No. of water supplies participating in AQV project	5	15	20
People served by the 20 water supplies	12 200 000	1045	12 201 045
No. of water supplies answering WP13 Questionnaire 1: Information about the water supplies, risk to water quality and WSP	5	15	20
No. of water supplies answering WP13 Questionnaire 2: Performance of AQV techniques	5	3	8
No. of WP11 Sanitary Site Inspection of the water supply site risk	0	15	15
No. of water supplies testing AQV techniques in WP10 and WP11	4	15	19
No. of surveillance monitoring sites 2013-2014	4	10	14
<b>No of samples:</b>			
No. of samples tested with AQV techniques in WP10 and WP11	215	263	478
No. of samples tested in AQV verification control in WP10 and WP11	177	153	330
No. of results gathered for two years of regular surveillance sampling (2013 and 2014)	2 906	134	3 040

Analyses of the impact from the new technique developed in the AQV project on WSP were done by using the WHO WSP manual (Bartram et al., 2009). The SSI surveys were designed to identify water supply site risks and were constructed based on WHO Guidelines for Drinking-Water Quality (1997).

The AQV technique tested included a single concentration procedure based on the use of the commercially available filter Rexeed™ 25 A for primary concentration with large volume filtration (10 to 1000 L). The volumes of concentrated Rexeed eluates varied between 200-700 mL. The Rexeed eluate were further concentrated for nucleic acid extraction to 2–5 mL using Centricon® 70 plus (140-210 ml), VivaSpin® 15R (50 ml) or PEG precipitation (600 ml). The nucleic acid extraction was performed using NucliSENS® or UNEX& OiaGen and the extracts were used for qPCR (20 µL) and FISH analyses (0,1-10 mL) (Puigdoménech et al., 2017). Before testing the newly developed methods for concentration, elution and extraction as well as detection technique were validated to secure their efficacy and to standardize their

use (Stange and Tiehm et al., 2015; 2016; Stange et al., 2016). Recovery rate was from 60% to over 80% depending on turbidity of the water (Hedui et al., 2015). The Rexeed filter coupled with the AQV protocol allows simultaneously concentration and recovery of pathogens of the three classes (bacteria, viruses and parasites), with significant economic gains.

The tested detection techniques included three off-line detection techniques; two molecular techniques qPCR (quantitative polymerase chain reaction) produced by two industrial partners Ceeram and GPS testing viruses, bacteria and parasites; one fluorescent in-situ hybridization (FISH) technique from Vermicon AG testing total cells (DAPI staining) and viable cells (EUB probe) as well as *E. coli* and thermophilic *Campylobacter* cells (these include *C. jejuni*, *C. coli* and *C. lari*); and one online system BACTcontrol, from the partner MicroLAN, measuring enzymatic activity with fluorescence that tested total activity and the indicator bacteria, total coliform bacteria and *E. coli*.

The techniques that were developed in the project were tested for one year (2016-2017) at **nineteen** water supplies in seven European countries, namely **four** large supplies in Denmark, Germany, Spain and UK, and **fifteen** small supplies located in Portugal, Scotland and Serbia.

The molecular techniques from Ceeram and GPS were tested on samples from the nineteen sites, large and small alike, whereas FISH and BACTcontrol were only tested at four large supplies. In all, testing was carried out for nineteen pathogens and indicators (Table 2). Results from BACTcontrol system and of Total cell counts and Total viable cell with FISH are not presented in this paper.

**Table 2. Pathogens and indicators tested with the AQV detection technique**

SME's	Type of technique and developed tools used	Pathogens and indicators tested	
		4 large water supplies (number of supplies tested)	15 small water supplies (number of supplies tested)
<b>Ceeram</b>	Molecular qPCR <b>qPCR-Kits for viruses:</b> norovirusGI@ceeramTools™ kit norovirusGII@ceeramTools™ kit hepatatisA@ceeramTools™ kit <b>qPCR-Kits for parasites:</b> cryptosporidium@ceeramTools™ kit giardia@ceeramTools™ kit	Norovirus GI and GII (All) Hepatitis A Virus (1) <i>Giardia</i> spp. (3) <i>Cryptosporidium</i> spp. (3)	Norovirus GI and GII (All) Hepatitis A virus (All) Hepatitis E virus (9) Enterovirus (6) <i>Giardia</i> spp. (All) <i>Cryptosporidium</i> spp. (All)
<b>GPS</b>	Molecular qPCR <b>qPCR-Kits for bacteria:</b> CamJej dtec-qPCR Test F100 EscCol dtec-qPCR Test F100 <b>qPCR-Kits for parasites:</b> CrySpp-dtec-qPCR Test F100 GiaInt dtec-qPCR Test F100	<i>Escherichia coli</i> (All) <i>Campylobacter jejuni</i> (All) <i>Salmonella</i> spp. (1) <i>Legionella pneumophila</i> (1) <i>Campylobacter</i> spp. (3) <i>Cryptosporidium</i> spp. (2) <i>Toxoplasma gondii</i> (1) <i>Giardia intestinalis</i> (1)	<i>Escherichia coli</i> (All) <i>Escherichia coli</i> 0157 (All) <i>Campylobacter coli</i> (All) <i>Campylobacter jejuni</i> (All)
<b>Vermicon</b>	FISH (Fluorescent in-situ hybridization) ScanVIT <sup>R</sup> Campylobacter SC ScanVIT <sup>R</sup> E.coli/coliforms SC	Total cell counts (All), Total viable cells (All), <i>Escherichia coli</i> cells (All), Thermophilic <i>Campylobacter</i> cells (All)	Not tested
<b>MicroLAN</b>	Online-platform for detection of bacteria measuring enzymatic activity with fluorescence of specific enzymes BACTcontrol system	Total enzymatic activity (2) <i>Escherichia coli</i> (1) Total coliform (1)	Not tested

Verification control of *E. coli* was measured according to ISO 9308-2. The results from regular surveillance monitoring were gathered for the water supplies participating in the testing from the local surveillance authorities, and for the two years 2013 and 2014. The surveillance monitoring of *E. coli* was performed with the conventional culturing methods (100 mL).

### 3. Results and discussion

This section is divided into five parts: 1) results from the survey of WSP performance at the large supplies; 2) general risk factors and challenges in twenty water supplies, both the large and the small supplies; 3) results from monitoring performed at the large supplies; 4) results from monitoring performed at the small supplies; and 5) evaluation of the impact the AQV techniques could have to improve WSP, if implemented.

#### 3.1. WSP Performance and benefits analyzed for large water supplies

The five large water supplies in four countries (Denmark, Germany, Spain and the UK) that participated in AQV answered WP13 Questionnaire 1. All supplies had developed and

implemented WSPs. It is mandatory to have a WSP in two of the countries, Denmark and the UK. Two sites had WSP certified as ISO 22000 and three had WSP developed in accordance with the guidance by WHO. All scored high in performance in all five components of WSP, as shown in Figure 1. Internal auditing was lacking at two supplies, and the WSP team was not active and periodic reviewing was lacking at one supply. The two that used ISO 22000 scored highest in the WSP process; the reason could be that ISO 22000 includes a requirement for regular external audits which, if violated, can lead to the loss of the ISO certification. Two of the small sites had WSP and six had recently done a risk assessment before being surveyed for the AQV project. However, none of the small supplies responded on WSP performance when answering Questionnaire 1.

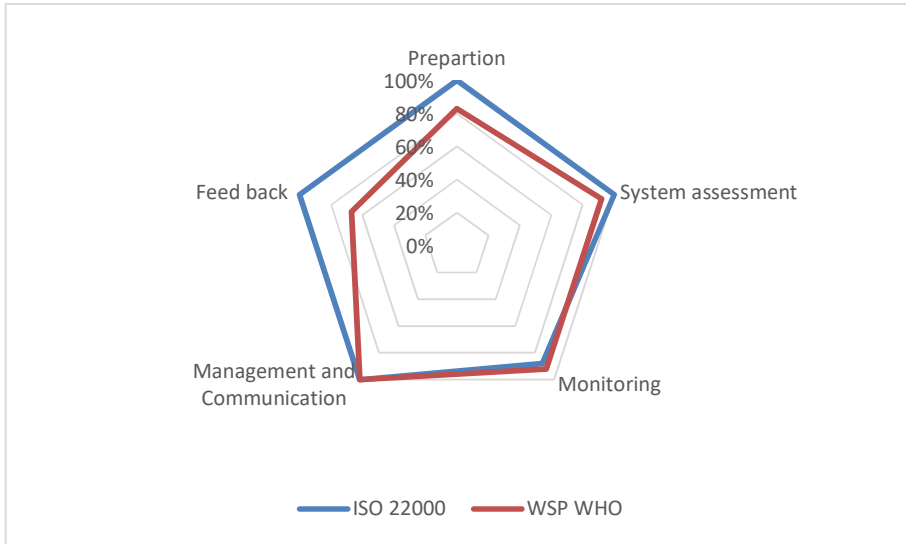


Figure 1. WSP performance in five large European water supplies in the 11 modules in the 5 main components of the WHO WSP (Fig. 9); preparation, system assessment, monitoring, management and communication and feedback.

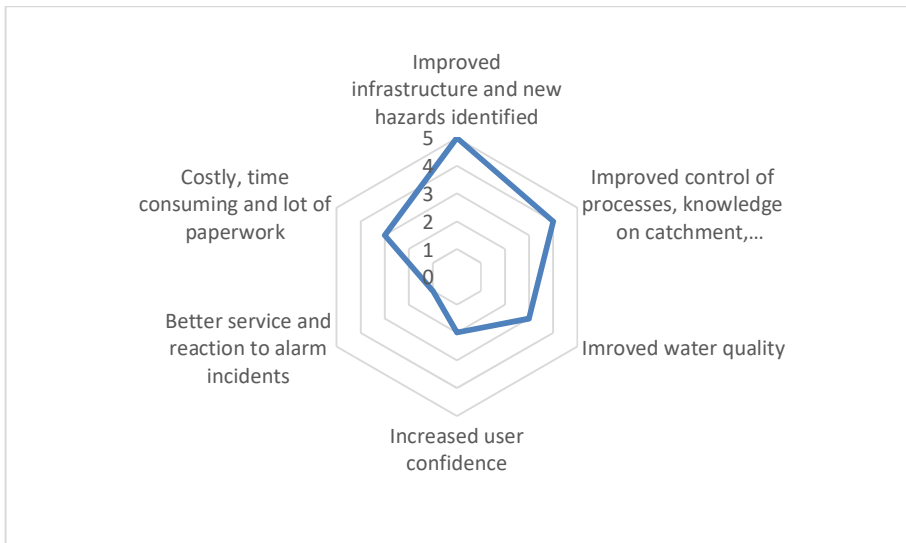


Figure 2. Main benefits and drawbacks reported for WSP from five European large water supplies



The main benefits stated with WSP were that infrastructure was improved and new hazards were identified (Figure 2). Improved control processes, water quality and knowledge of the status of the catchment were also considered beneficial. Regarding management, the main benefits experienced were that professionalism improved, and at two sites user confidence increased. Improved internal communication was also mentioned as a benefit by one respondent. The drawbacks cited by three supplies were that WSP is costly and time-consuming as well as involving a lot of paperwork. Two supplies considered WSP to have no drawbacks. The conclusion was that all five large water suppliers considered WSP as beneficial in many aspects that should result in safer water.

**3.2. Risk factors and challenges analyzed for twenty European large and small water supplies**

Risk factors to water safety were identified for all twenty water supplies. There were varied and significant risk-posing activities on the catchments of many of the water supplies, as shown in Figure 3. Most had some potential sources of faecal contamination within the catchment area (85%), i.e. sewage works, septic tanks and/or presence of animal fecal matter. Many supplies (70%) had agriculture, either cultivation, livestock, or both, practiced within their water catchment area. The presence of farm waste in the catchments was common for the small supplies, and two large supplies also had oil tanks in their catchment. All the large supplies had residential areas in the catchment and three of the small supplies also had some residential areas, and some risks also associated with the transport infrastructure and other activities at the catchments.

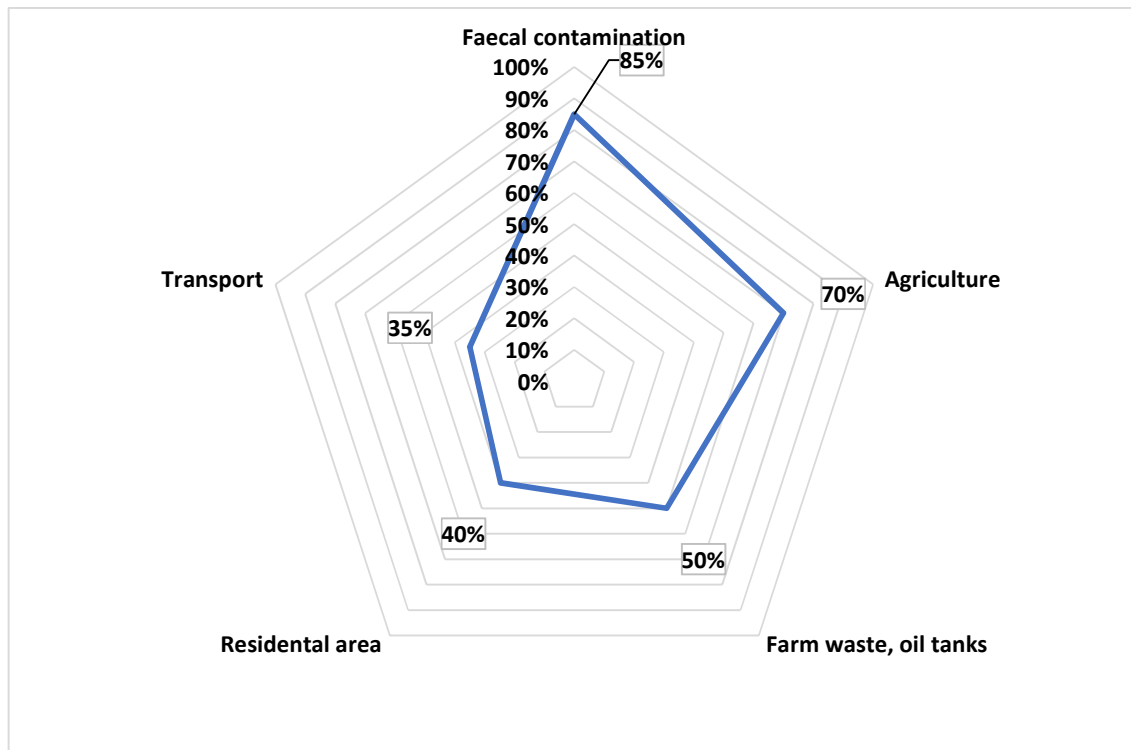


Figure 3. Polluting activity in the catchment for the twenty European large and small water supplies

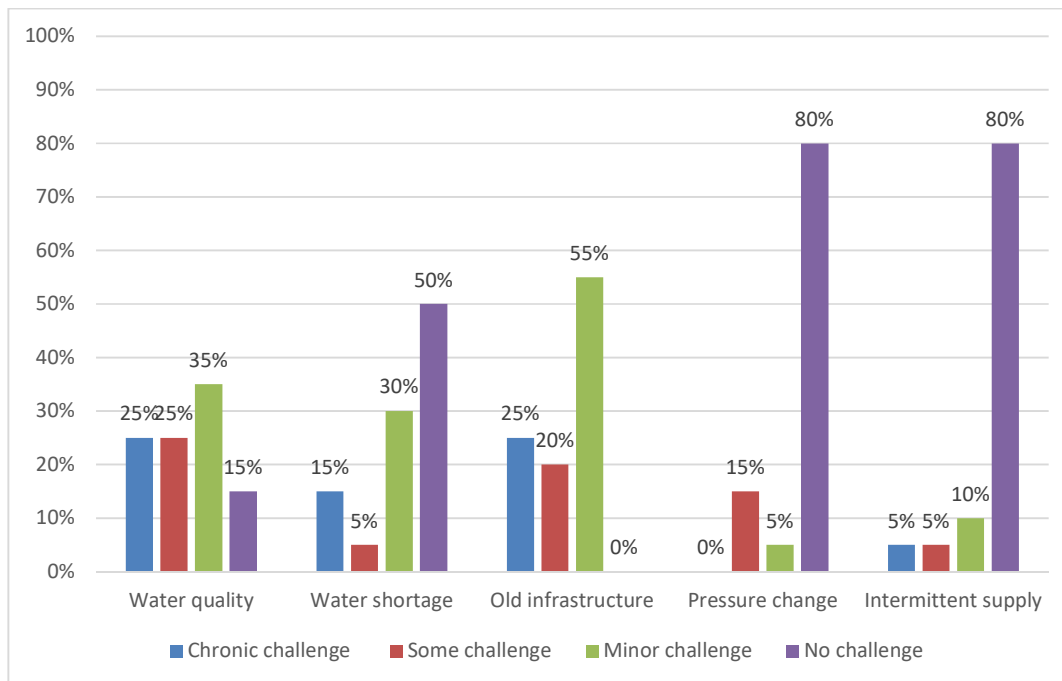
The pipes were old, especially at the large supplies, and more so in the transport pipelines from the water source to the urban areas compared to the distribution network. Pipe breaks were frequent in the large supplies (information was not available for the small supplies). Pipe

breaks per year in large supplies were on average 0.68 per km (Table 3). However, pipe breaks were much more frequent in the distribution network than in the transport pipelines (0.82 versus 0.07 pipe break per km). The explanation is most likely due to a higher stress on the infrastructure from traffic and other activity in the urban areas as well as more fluctuating pressure in the distribution network. This will increase the probability of pipe damage and increase leakages and the latter have been shown to cause ingress of contamination into pipes (Karim et al., 2003; Fox et al. 2016). The median pipe age in the large supplies was 51 years and 10 years in the small supplies. The oldest pipes in the large systems were reported as 99 years old, and one site in the small supplies reported that the pipes were 140 years old. Sewage was reported in the same ditch as drinking water pipes in two of the large supplies, increasing risk of fecal contamination. Leaking pipes increase risk of contamination. Only two large water supplies reported leakage, 8% and 17%. In the new DWD proposal there is a requirement of reporting and reducing leakage. In a recent EU report on leakage management it is stated that the average leakage from the supply network in the EU is 23% (EU, 2015).

**Table 3. Infrastructure data at the twenty European water supplies as indicator of water quality risk**

	Units	5 Large water supplies	15 Small water supplies
Source of water <sup>1</sup>	%	G = 42% S = 58%	G = 87% S = 13%
Sites with treatment	No.	4	10
Total length of pipelines	km	6 860	40
Total length of pipelines per person	km per person	0.76	38
Main pipe types	%	Ductile (33%), Cast iron (20%), PEH (16%), Asbestos (14%), Steel (6%), Concrete (5%), PVC (2%), Other (4%)	PVC, PEH and Cast iron <sup>2</sup>
Median pipe age	Years	51	10
Average pipe age	Years	54	28
Pipe breaks	No. per year	4 633	n.a. <sup>3</sup>
Pipe break frequency	Per km/year	0.68	n.a.

1) G= groundwater, S= surface water from river and/or lake; 2) Information on length of each type of pipe not available; 3) n.a. information not available.



**Figure 4. Main challenges in twenty European large and small water supplies**

Figure 4 shows the main challenges regarding water quality reported at the twenty water supplies. These included old infrastructure at all water supplies (100%), water quality (85%) and water shortage (50%). To a lesser extent there were challenges with pressure changes and intermittent supply (20%) that occurred almost only in the small supplies. Five of the twenty water supplies have had to cope with old infrastructure as a chronic challenge (25%), and pipe breaks and the resulting leaks were likely to have posed a risk to water safety. Challenges with pressure changes and intermittent supply pose an increased risk to water quality, especially in old pipe systems and if in the same ditch as sewage pipes. This reveals that there is a need to improve resource efficiency in Europe with improved leakage control and renewing the infrastructure, preferably done through requirements set by the DWD and then transported into national legislation.

Based on the data collected, it can be concluded that the sources of fecal contamination for the studied water supplies can either be from direct water runoff, including fecal matter within the catchment, or fecal matter entering into the pipe networks via cracks in the aging infrastructure, or where sewage pipes are in close proximity to drinking water pipes, or where there is some cross-connection to the sewage system. This situation with aging infrastructure and fecal contamination at the catchment or in the system could, to some extent, be representative of the situation in the water sector in Europe. Summarizing causes of twenty-nine examples of waterborne outbreaks in the developed countries, Hruday & Hrudays (2014) revealed that pathogenic outbreaks were divided equally into source contamination and contamination happening in the network, the latter often caused by accidental cross-contamination.

### 3.3 Test results for the large supplies

In the large supplies there were 104 instances of pathogens found in samples with the AQV techniques. The ones most frequently detected were norovirus (52 of 53 viruses detected)

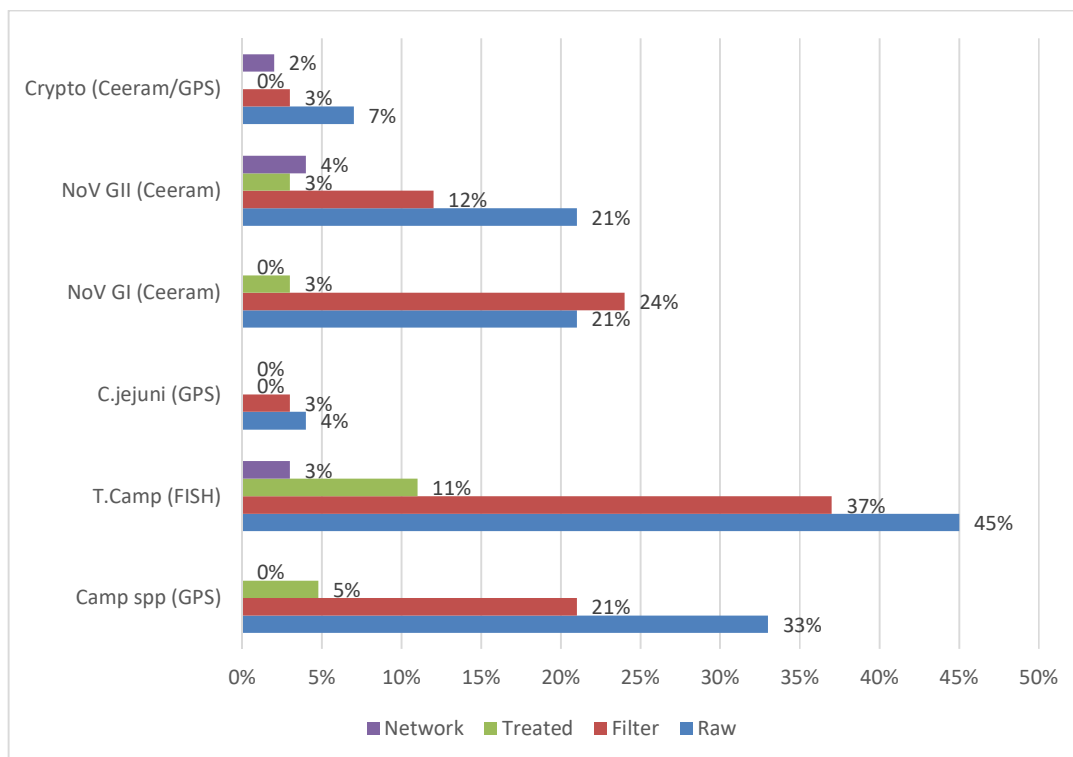
and *Campylobacter* (39 of 47 bacteria detected). All classes of pathogens were detected in raw and processed water, though mainly viruses and bacteria, as shown in Table 4. There were also few sporadic instances of pathogens in treated water leaving the treatment station and in the distribution network. In all, 24% of the samples were detected with pathogens, though mostly in raw and processed water, 40% and 31% respectively (Table 4).

**Table 4. Pathogens tested at the 4 large water supplies**

	No. of samples tested for pathogens	No. of samples with pathogens	% of sample with pathogens	No. of pathogen instances	Classes of pathogens		
					Bacteria	Virus	Parasites
<b>Raw water</b>	57	23	40%	47	20	25	2
<b>Processed water*</b>	67	21	31%	48	23	24	1
<b>Treated water</b>	39	4	10%	5	3	2	0
<b>Network</b>	54	4	6%	4	1	2	1
<b>Total</b>	217	52	24%	104	47	53	4

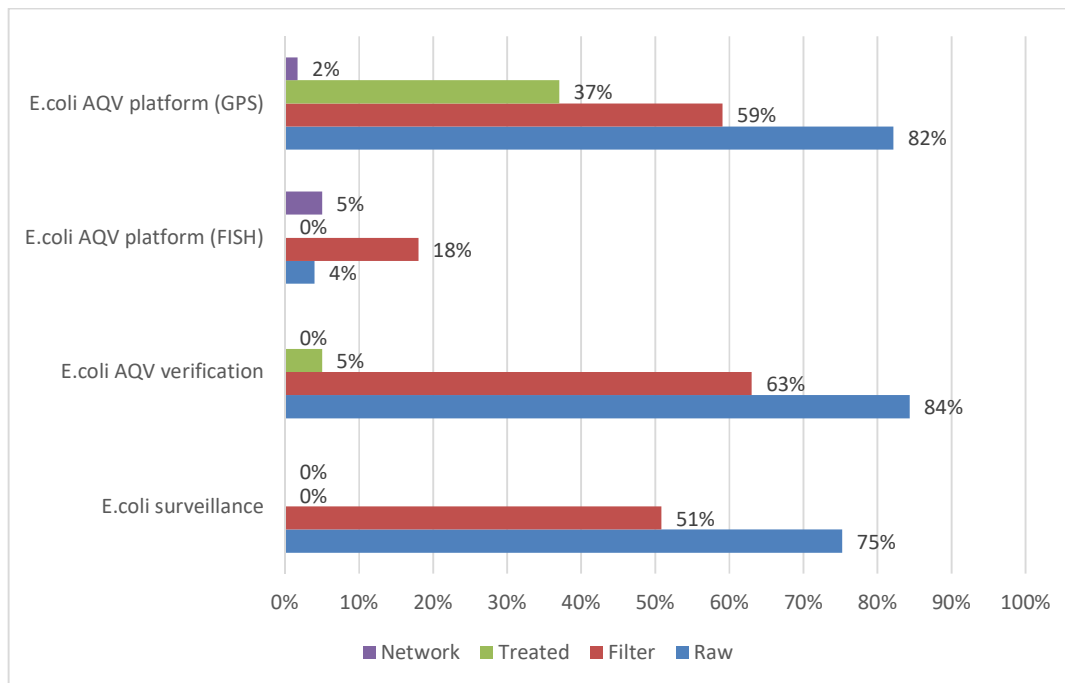
\*Processed water includes treatments such as flocculation/sedimentation, sand filtration, dissolved air flotation, and GAC filtration with a prior ozonation at the different demonstration sites.

Results from the pathogen monitoring with the AQV techniques, qPCR Ceeram, qPCR GPS and FISH in the large supplies are shown in Figure 5. NoV GI and GII were detected in 12 to 24% of the samples in raw and processed water, and some in treated water and in the network in very low concentrations (<1 GU/L). Most pathogens were found in untreated surface water and less often in groundwater. *Cryptosporidium* was found sporadically in raw and processed water, and in the network. *Giardia* was not detected with the AQV molecular techniques in the large supplies but several times with the AQV improved conventional verification method (IMS, Immunomagnetic separation) in raw and processed water (not shown in Fig. 5). This indicates problems of qPCR of detecting parasites that require development or refinement of the method.



**Figure 5. Pathogens tested and detected with the AQV techniques in the 4 large European water supplies as a percentage of sample tested**

Detection of pathogens was to some extent site-specific. For instance, norovirus was only found at two of the four supplies tested, both using surface water source and mostly in raw water and processed water, whereas *Campylobacter* spp, *C. jejuni* or thermophilic *Campylobacter* were found at three sites. *Cryptosporidium* was only found at one site (four instances), and at all stages. It also must be noted that the amount of testing for pathogens was not the same at all sites as at one site there was more testing, especially in the network. At this site, tests for additional pathogens, such as Hepatitis A (HAV), *Salmonella* and *Legionella* (*L. pneumophila*), were also performed, and the results from these additional tests are not included in Figure 5. HAV was only detected in one sample in raw water, *Salmonella* was found in one processed water sample, and *Legionella* in seven samples (one in raw water and six in processed water). The same large water supply was also tested for *Toxoplasma gondii*, but it was not detected in any samples.



**Figure 6. Detection of *E. coli* with AQV techniques, AQV verification and in regular surveillance monitoring in 4 large European water supplies as a percentage of sample tested**

The results from monitoring *E. coli*, as an indicator of fecal contamination, with the AQV techniques (qPCR kits of GPS) in the large supplies, showed a high percentage of positive samples in both raw and processed water (82% and 59%), and even in treated water (37%), and on one occasion in the network at a very low level (< 50 GU/L), as shown in Figure 6. That should result in consideration of disinfection methods at the distribution networks. Similar results for *E. coli* were obtained with the AQV verification control in raw and processed water but were lower in treated water. This could indicate that the AQV technique is more sensitive when PCR inhibitors are present at lower levels, in contrast to raw surface water. However, an important challenge associated with the molecular qPCR detection is that there is no distinction between live and dead cells. This is another factor possibly contributing towards the difference between the *E. coli* detected with AQV technique (GPS) and with the conventional verification method, or 32% (37% minus 5%). It is likely that pathogens inactivated by various disinfection processes are not detected with the culture method, but their genetic fragments are detected by the qPCR method. Much lower detection of *E. coli* was found with the FISH method and gave unreliable results with the lowest *E. coli* detected in raw water (Figure 6). Figure 6 also shows that somewhat lower detection was found with regular surveillance than with the GPS and AQV verification methods where no *E. coli* were found in treated water in regular surveillance monitoring.

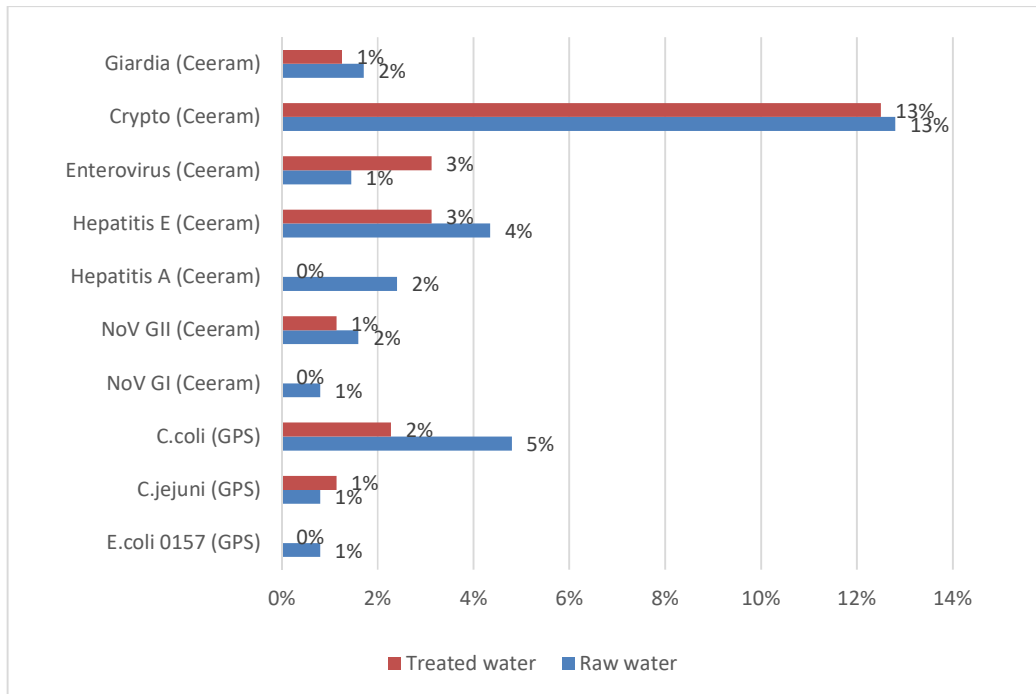
### 3.4 Test results for the small supplies

In the small water supplies, there were 61 instances of pathogens found with the two AQV techniques (qPCR Ceeram and qPCR GPS). The most frequently found pathogens were *Cryptosporidium* (28 of the 31 parasite instances) and *Campylobacter coli* (11 of 18 bacterial incidents detected). All pathogen classes were detected in both raw and treated water, although approximately half were parasites (Table 5).

**Table 5. Pathogens tested at the 15 small water supplies**

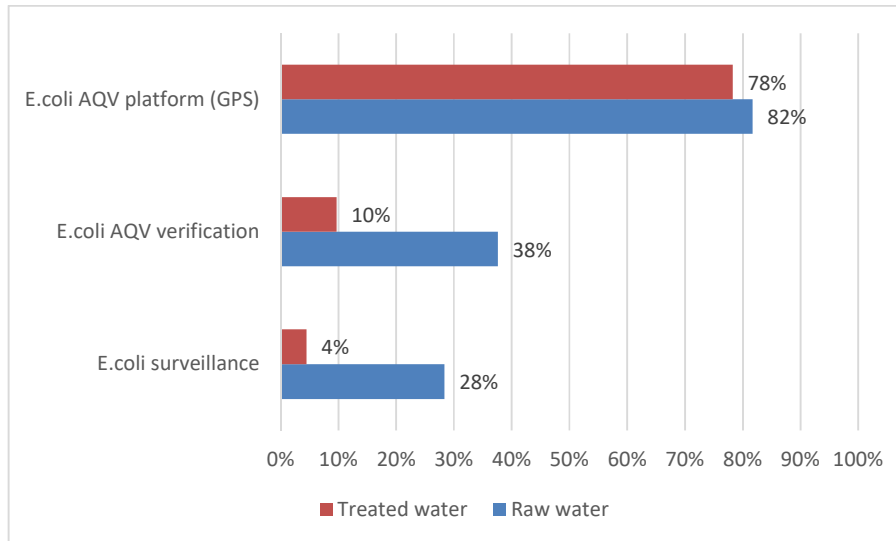
	No. of samples tested for pathogens	No. of samples with pathogens	% of samples with pathogens	No. of pathogen instances	Classes of pathogens		
					Bacteria	Virus	Parasites
Raw water	159	35	22%	37	10	9	18
Treated water	92	21	23%	24	8	3	13
<b>Total</b>	<b>251</b>	<b>56</b>	<b>22%</b>	<b>61</b>	<b>18</b>	<b>12</b>	<b>31</b>

There were fewer instances in raw water of the pathogens in the small supplies compared to the large supplies (Fig. 7). The reason could be that the raw water was mostly groundwater in the small supplies, 87% compared to 42% in the large supplies (see Table 3). Another explanation for the higher instance rate of pathogens in raw water at the large supplies could be the denser population in the catchment of the urban areas. However, pathogens were more frequent in treated water in the small supplies compared with large water supplies, 23% and 10%, respectively. This reflects the water quality problems of small water supplies discussed in the introduction. The length of the pipeline infrastructure is also much longer per user in the small supplies than in the large supplies (Table 3). This reveals the relatively higher investment cost and operational cost needed for managing the distribution systems in small water supplies as well as the higher risk of contamination and illustrates the challenges that small water suppliers must deal with.



**Figure 7. Pathogens detected with the AQV technique in 12 small European water supplies out of 15 tested as percentage of samples tested. No pathogens were detected in one of the three countries participating in the AQV trial**

As in the large supplies, pathogens in the small supplies were site-specific and country specific. Of the three countries in this study, *Cryptosporidium* was the dominant pathogen in one country and *Campylobacter* in another. No pathogens were detected in the third country (with three small water supply sites tested) though *E. coli* was detected in all samples from the three test sites in this country with AQV GPS and with verification testing. This could indicate some error in the approach used. Enterovirus was only monitored in one country, at six test sites. HEV was monitored in two countries, at nine sites.



**Figure 8. Detection of *E. coli* with AQV new technique, AQV verification in 15 European small water supplies<sup>3</sup> and regular surveillance monitoring in 10 small water supplies<sup>3</sup> as a percentage of samples tested**

The detection of *E. coli* was high in samples from the small water supplies with the AQV techniques, in around 80% of samples, in both raw and treated water at all fifteen small supplies combined, as shown in Figure 8. It was much higher than detected with the culture verification method (done in parallel with the AQV testing) and even more than detected through regular surveillance monitoring. Ten of the fifteen small supplies participating in the study had disinfection treatment, either UV or chlorination. However, all ten demonstrated a high detection of *E. coli*, both with AQV technique (GPS) and verification, revealing insufficient treatment. Results from regular surveillance monitoring showed much lower non-compliance in *E. coli* in the 134 samples from 10 sites than done with verification method done by WP11 indicating that the latter is more sensitive.

### 3.4. Impact of new monitoring techniques on Water Safety Planning

The WSP framework presented in the WHO WSP manuals consists of five main components: 1) the preparation stage; 2) system assessment; 3) monitoring performance; 4) management and communication; and 5) feedback and improvement. These components are divided into eleven modules, as shown in Figure 9 (Bartram et al., 2009).

<sup>3</sup> Surveillance monitoring was only available for 10 of the 15 small water supplies tested.



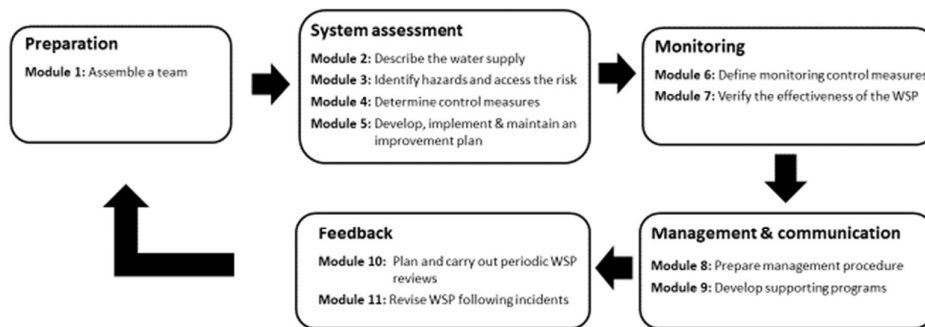


Figure 9. Overview of the 11 modules described in the WHO-WSPs manual (Bartram et al., 2009)

Improved knowledge of water quality and on the presence of pathogens in water will have an impact on water safety management, such as WSP, in many ways. The AQV testing showed that fecal contamination and certain pathogens are frequent but not at all sites. This calls for improved control and preventive measures at the water source as a part of WSP and supports the objective of the Water Framework Directive to gain previous water quality status of aquifers. Pathogens and fecal contamination were also high in treated water at the small supplies, emphasizing the need to improve treatment with training and guidelines as well as for improved control.

The results from employing the AQV techniques revealed that the pathogens were both site- and country-specific. This local or countrywide knowledge should be included in the risk assessment for individual water supplies. The possible impact from improved monitoring with the AQV techniques is summarized in Table 6, along with the impact on each of the five components and discussed in following sub-sections.

Table 6. Summary of possible impact from faster and improved monitoring on each module in WSP

Phase	Module	Impact on WSP
Preparation	<b>Module 1</b> Assemble a WSP team	Knowledge of presence and impact of pathogens will have to be added to the WSP team skills, as well as basic knowledge of the new detection technique and of ICT to inform consumers.
	<b>Module 2</b> Describe the water supply system	Knowledge of the presence of microbes will assist in identifying water quality status and status of infrastructure.
System assessment	<b>Module 3</b> Risk assessment	Knowledge of pathogenic occurrence will assist in risk assessment and give more accurate risk scoring.
	<b>Module 4</b> Determine control measures	Establishing the pathogen load in raw water will identify the adequacy of treatment. Identification of pollution will support necessary control measures, e.g. agreement with stakeholders on catchment.
	<b>Module 5</b> Improvement plan	Identification of pollution will support and prioritize improvement plan as renewal of infrastructure and improved treatment.
Monitoring	<b>Module 6</b> Monitoring effectiveness of control measures	Monitoring of common waterborne pathogens validates control measures. Fast off-line molecular monitoring and online monitoring of microbes will increase water safety. It will also assist in treatment processes.
	<b>Module 7</b> Verification external	Validation of regular external surveillance monitoring
Management and communication	<b>Module 8</b> Management procedure	Revised SOPs for treatment process are needed with improved management with online telematics monitoring. With the new possibility in ICT consumers can be informed more promptly of water quality status and boil advisory if needed.
	<b>Module 9</b> Supporting program	Improved training of staff is needed to adapt to this new technique and guidelines for running treatment station as UV.
Feedback	<b>Module 10</b> Periodic review	New information on pathogenic occurrence will be included in periodic review and confirm performance.
	<b>Module 11</b> Revise following incident/near misses	Improved and faster simultaneous monitoring of many pathogens will assist in case of incidents, emergencies or near misses.

### 3.4.1 Preparation

The preparation phase includes assembling a team responsible for the WSP and setting the agenda for the team. The implementation of the AQV techniques would require increased knowledge by the WSP team. Knowledge and the significance of the presence and impact of pathogens, as well as performance of treatment to reduce them, should be added to the WSP team skills. They should also understand the advantages and limitation of the monitoring techniques. The possibilities for the information and communication techniques to increase information to the consumers should also be a part of the team expertise.

#### *3.4.2. System assessment*

The second phase Assesses the system, describing it from catchment to consumers' tap and identifying places where water quality problems could arise (defined as critical control points, CCP), and performs the risk assessment, deciding on actions needed to prevent pollution, and carrying them out. Improved monitoring would increase knowledge of relevant pathogens and hence assist in identifying water quality problems in the system and verifying current risk assessment. It will also establish pathogen loads in source water that will support necessary control measures as regular cleaning of tanks and improve plans renewal of infrastructure to mitigate risk. Furthermore, as previously discussed, the results from the small supplies show high fecal contamination in the source waters tested, and even in treated waters tested, so risk assessment and preventive management should be applied in all supplies or improved if they are already in place and have not identified problems with pathogens. Improved monitoring also has the potential to help in microbiological management of treatment processes and to prioritize any necessary improvement plans.

#### *3.4.3. Monitoring*

The third phase is monitoring the performance of control measures, both with operational monitoring and external regulatory surveillance. The implementation of the molecular methods, which can potentially detect multiple pathogens quicker than the respective culture methods, has an important early warning potential in preventive management. The AQV techniques monitoring pathogens and indicators may also be used to validate if WSP, with its control measures, is working as it should in all stages of the water delivery. It will also validate external regular surveillance testing of indicators. The online AQV techniques has the potential to give early warning (in a few hours) of elevated levels of total activity, presence of coliform bacteria or *E. coli*, for example in case of surface water intrusion into groundwater and thus prevent any large spread of contamination, either by closing wells or boreholes or improving treatment.

#### *3.4.4. Management and communication*

The fourth phase addresses management, including support programs with training, management procedures and communication to users and stakeholders. The implementation of the AQV techniques is expected to improve treatment management and procedures in the treatment process. Procedures in managing water resources will also change with improved knowledge of pathogens and early warning of any change in water quality. This study found that treatment at the small supplies is often inadequate, and thus guidelines and training of staff to use instrumentation and treat water should be an essential part of WSP, especially for small water supplies serving the public. Considering the great progress in information and communication techniques, there are now great possibilities to provide consumers with more timely information about their drinking water. This will enhance water quality and performance of services. Improved communication to the public and other stakeholders is high on the agenda of the European Commission, and, therefore, the availability of more rigorous results that could be communicated to the EU citizens is very relevant.

#### *3.4.5. Feedback*

The fifth phase addresses feedback, both regular and in the case of incidents or near misses/close calls. Improved monitoring of pathogens and indicators will lead to better knowledge of sporadic incidents of pathogens that will assist in feedback and support revision of risk assessment. Knowledge of the status of pathogens will also support external auditing of WSP.

#### 4. Conclusions

Improved knowledge of water quality and of the presence of pathogens in water have the potential of a positive impact on WSP management in many ways. The AQV techniques can validate whether the control measures that have been implemented as part of WSP are working as they are intended and confirm the quality of the source water. For example, the AQV online monitoring techniques has the potential to provide early warning (1-2 hours) of elevated levels of fecal contamination (by measuring total enzymatic activity, total coliforms or *E. coli*) in source water, which then can inform operational actions, for example, immediate closing down of water sources, where needed, and thus preventing contamination of drinking water. The techniques can also be important in providing information about the impact from natural hazards to water quality, for example, extreme weather events which are expected to become more frequent as a result of climate change. These techniques can be combined with up-to-date information technology in order to provide crucial information to the consumer that could lead to increased confidence and trust in the safety of the water.

The results from employing the AQV techniques in field trials in multiple water supplies across Europe showed presence of pathogens in water. Pathogens were detected in 24% of samples from the large supplies, mostly in raw water (40%) and less in treated water (10%). In samples from the small supplies 22% had pathogens, equally in raw and treated water, and half of them parasites. The current revision of the DWD adds a requirement for monitoring of additional indicator organisms, more specifically for the pathogen classes of viruses and parasites: somatic coliphages and *Clostridium perfringens* spores.

The monitoring results showed significant fecal contamination in water, even in treated water, at the small supplies. This emphasizes the need for risk-based management at the small supplies, as is specified in the current DWD revision proposal. The AQV project also revealed the need to include appropriate guidance and training related to treatment in the small supplies, e.g. UV treatment. Old infrastructure and fecal contamination via catchment runoff are important challenges, as was demonstrated in the frequent fecal contamination found in raw water and the frequent pipe breaks reported, which emphasize the need for leak control and systematic risk-based renewal of infrastructure, as also specified in the new proposal for the DWD.

The results of the testing showed that monitoring with some of the molecular methods allows fast and reliable detection of some of the most common waterborne pathogens, and that monitoring for levels of fecal pollution has a significant early warning potential in preventive management. Some of the molecular methods trialed are likely to require further development or refinement, and some methods, in their current state, are likely to be more feasible in a limited number of water types. Some of the new methods allow for obtaining results from monitoring faster, which can be important for informing operational actions if a positive detection is obtained in the water tested.

The current DWD has currently been in use for nearly twenty years and has improved water quality for most European citizens. However, many still live with unregulated or poorly regulated water, as demonstrated in the case of the small supplies, or even complete lack of access to safe drinking water. The human right to water and sanitation has been recognized by the UN and the goal is that before 2030 everyone should have access to safe and affordable drinking water. The EU has also recognized the human right to water in the new proposal for revision of the DWD inspired by the Right2Water initiative. The AQV project, with its emphasis

on water safety plan management and tracing pollution with advanced and fast technologies, can assist in achieving the goals of the EU DWD and national regulations on safe water for all.

## Acknowledgements

This work was supported by the European Union Seventh Framework Programme under the AQUAVALENS project (Grant Agreement number 311846).

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