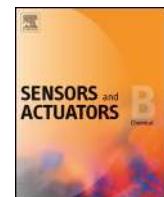




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Review Article

Detection of contaminants in water supply: A review on state-of-the-art monitoring technologies and their applications

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ABSTRACT

Water monitoring technologies are widely used for contaminants detection in wide variety of water ecology applications such as water treatment plant and water distribution system. A tremendous amount of research has been conducted over the past decades to develop robust and efficient techniques of contaminants detection with minimum operating cost and energy. Recent developments in spectroscopic techniques and biosensor approach have improved the detection sensitivities, quantitatively and qualitatively. The availability of *in-situ* measurements and multiple detection analyses has expanded the water monitoring applications in various advanced techniques including successful establishment in hand-held sensing devices which improves portability in real-time basis for the detection of contaminant, such as microorganisms, pesticides, heavy metal ions, inorganic and organic components. This paper intends to review the developments in water quality monitoring technologies for the detection of biological and chemical contaminants in accordance with instrumental limitations. Particularly, this review focuses on the most recently developed techniques for water contaminant detection applications. Several recommendations and prospective views on the developments in water quality assessments will also be included.

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List of Acronyms

AACC	American association of cereal chemist
AC	Alternating current
AI	Artificial intelligence
ANN	Artificial neural network
ATR	Attenuated total reflectance
BSA	Bovine serum albumin
CE	Capillary electrophoresis
CCD	Charge coupled device
CDOM	Chromophoric dissolved organic matter
DIS	Dielectric impedance spectroscopy
DMF	Digital microfluidic
DNA	Deoxyribonucleic acid
DOM	Dissolved organic matter
DTM	Dynamic threshold method
EC	Electrical conductivity
EIS	Electrical impedance spectroscopy
EPA	Environmental protection agency
EWOD	Electrowetting-on-dielectric
EWS	Early warning system
FDOM	Fluorescence dissolved organic matter
FIR	Far infrared
FISH	Fluorescence in situ hybridization
FPGA	Field-programmable gate array
FTIR	Fourier transform infrared spectroscopy
GCMS	Gas chromatography mass spectrometry
HCA	Hierarchical cluster analysis
HPLC	High performance liquid chromatography
ICPMS	Inductively coupled plasma mass spectrometry
IR	Infrared
LAMP	Loop-mediated isothermal amplification
LCMS	Liquid chromatography-mass spectrometry
LCOF	Liquid core optical fibre
LCW	Liquid core waveguide
LED	Light emitting diode
LOADEST	Load estimator
LOC	Lab-on-a-chip
LPFG	Long-period fibre grating
MIR	Mid infrared
MVE	Minimum volume ellipsoid
MPN	Most probable number
MS	Mass spectrometry
NIR	near infrared
ORP	Oxidation reduction potential
PBS	Phosphate buffered saline
PCA	Principal component analysis
PCR	Polymerase chain reaction
PFC	Perfluorinated compounds
POC	Point-of-care
PSA	Prostate specific antigen
SARS	Severe acute respiratory syndrome
SERS	Surface enhanced raman spectroscopy

SPA	Sensor placement approach
SPE	Solid phase extraction
SVM	Support vector machine
TDLAS	Tunable diode laser absorption spectroscopy
TN	Total nitrogen
TNT	2,4,6-trinitrotoluene
TOC	Total organic carbon
TU	Turbidity
UN	United nations
UNESCO	United nations of environment, scientific and cultural organization
UNICEF	United nation children's fund
USEPA	United state environmental protection agency
UV	Ultraviolet
VDA	Vector distance algorithm
WDS	Water distribution system
WHO	World health organization
μPADs	Microfluidic paper-based analytical device
μTAS	Micro total analysis system

1. Introduction

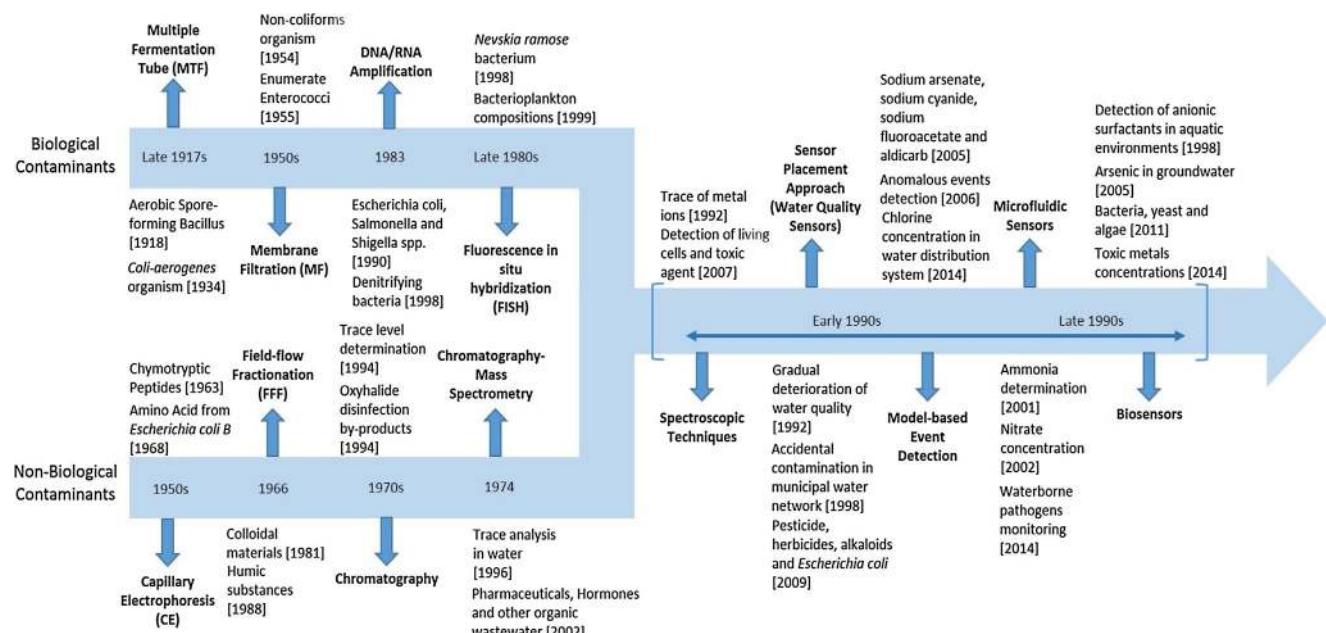
Waste production from agriculture, industrial sewage, animal and human activities are affecting the boundaries between clean water and wastewater, causing the reduction in the fresh water supply for human. Water ecology provides services such as food production, nutrient cycling, habitat provision, flood regulation, water purification and soil formation [1]. Biological and chemical contaminants in tap and drinking water, initiate the evolution of contagious diseases [2]. Therefore, fast and sensitive detection techniques are crucial to ensure safe and clean water supply. Unsafe water supply affects human health, causing contagious diseases such as hepatitis, influenza, SARS, pneumonia, gastric ulcers and pulmonary disease [3]. There are numerous non-biological contaminants existed in the water supply and some of the examples are silica, sodium, sulphur, ammonia and chlorine [4]. Other hazardous substance of heavy metals such as cadmium (Cd), lead (Pb), arsenic (As), mercury (Hg) and nickel (Ni) are also found in water supply [5]. These non-biological contaminants are among the commonly detected pollutants in urban areas that constitute a wide array of human activities.

The preservation of water quality has been regulated since the introduction of directive 91/271/EEC, which requires accurate treatment process targeting on organic contaminants, nitrogen and phosphorus [6]. In addition to these contaminants, other concern on the water quality includes the existence of microbiological contaminants in tap and drinking water at point of consumption. Derivation of pathogenic activity in water supply poses serious threats not only to human but also the entire water ecosystem. Pathogenic microorganisms can be categorised into bacteria (e.g., *Salmonella typhi*, *Vibrio cholera* and *Shigella*), viruses (e.g., *Poliovirus*) and protozoa (e.g., *Giardia lamblia* and *Cryptosporidium*). These types of microorganisms have been periodically detected in the water

Table 1

List of the most commonly found contaminants in water supply [9–13].

Parameter	Occurrence	Health Significance	Limit Value
Non-biological			
Ammonia	Results in microbiological activity	Irritations to eyes, nose and throats, non-deadly threats to human	0.5 mg/L
Arsenic	Dissolution of minerals from industrial	Very toxic to humans, high risk of skin cancers	10 µg/L
Barium	Natural occurring chemicals	Painful swallowing, ulcer	5 µg/L
Boron	Natural occurring chemicals, leach of rocks and soil	Kidney failure, depression	0.5 mg/L
Chlorine	Industrial effluents	Toxicity to humans, hazardous	5 mg/L
Chromium	Industrial processes	Skin irritation, damage kidney, liver	10 µg/L
Cadmium	Sediments of rock and soil	Hazardous to human, effect respiratory system and bone disease	3 µg/L
Lead	Leaching from ores, attack on water pipes	Toxic cumulative poison	10 mg/L
Mercury	Normally from industrial waste	Very toxic, human fatal	1 µg/L
Nickel	Chemical used in water treatments	Cancer of lungs and nose	20 µg/L
Nitrate	Presence from agricultural activities	Risk of lifetime cancer	3 mg/L
Sodium	Natural waters, abundant of rocks and soil	High-blood pressure, heart diseases	200 mg/L
Biological			
<i>Cryptosporidium</i>	Presence in human and animal waste	Infections, fever, stomachache, diarrhoea	630 mL/L
<i>Escherichia coli</i>	Sewages and similar waste	Pathogenic properties, effect human health	10 CFU/mL
Giardia	Presence in human and animal waste	Effect human health, rarely fatal	10 cysts/L
<i>Legionella</i>	Sediments of water	Risk of Legionnaire's disease and Pontiac fever	100 CFU/mL
Pesticide	Agricultural discharges, spillages	Eyes and ears infection	0.1 µg/L
<i>Pseudomonas</i>	Abundant in sewage	Hypertension if taken excess	500 CFU/mL

**Fig. 1.** Evolution of contaminant detection techniques in water analysis application.

samples of river, groundwater and drinking water [7,8]. Hence, minimizing the exposure of deadly diseases is important by providing early warning detections on the presence of pathogens [9]. According to World Health Organization (WHO), the most commonly found microorganisms in the drinking water sources are *Cryptosporidium*, *Legionella*, *Pseudomonas*, *Giardia* and *E. coli* [10–13]. A list of possible water supply contaminants that is based on standard guideline is summarized in Table 1.

Recently, analytical technologies in water monitoring have taken a variety of directions. There are several water monitoring techniques, including conventional instrumental analysis (laboratory-based analysis), sensor placement approach, model-based event detection, microfluidic devices, spectroscopic approach and biosensors. Selecting suitable detection technique(s) is strongly dependent on the purpose of intended detection analysis, whether it requires quantitative, qualitative or hybrid measurement. Biological and chemical sensors have been in great demand for use in water monitoring technology, and they

appear to be feasible for device integration and commercialization.

Previously, the detection of water contaminants has often been conducted manually in water laboratory facilities [14]. At the laboratory level, analyses are usually carried out by skilful personnel using high-end and cutting-edge technologies. Conventionally, multiple fermentation tube technique [15,16], filtration method [17,18], DNA amplification [19], fluorescence *in-situ* hybridization (FISH) techniques [20,21], capillary electrophoresis [22,23], field-flow fractionation [24,25], chromatography [26,27] and mass spectrometry [28,29] are the commonly used instruments to detect contaminants in the water samples. The potential benefits of laboratory-based analytical methods have been recognized for a long time, but studies have shown that they are not very efficient for on-site monitoring applications. With the technological advancement in the analytical chemistry, new techniques are developed through the introduction of advanced spectroscopy [30], model-based event detection [31], water quality sensors [32,33],

microfluidics [34–36] and biosensors [37–39]. Recently, wireless sensor network has been adapted in various detection techniques for portability. The evolution of water contaminant detection techniques is illustrated in Fig. 1.

Contaminant detection analysis is gaining importance in the water monitoring and environmental applications. An initial growth in water quality sensor fabrication and optimal sensor placement for event detection has been introduced over the past several years [40–43]. The synthesis of the aligned water sensors, also known as sensor placement approach, could improve the device detection performance by increasing the detection sensitivity [44,45]. Deploying sensors in water distribution system allow the measurements of temperature, pH, turbidity, conductivity, oxidation reduction potential (ORP), UV-254, nitrate-nitrogen and phosphate both on-line and off-line. These water quality parameters are important to the treated water could meet the limit of detection (LOD) set by the United State Environmental Protection Agency (USEPA) [46]. However, the installation of numerous sensors along the distribution system seems not very practical owing to high installation cost [47].

Determining the presence of contaminants in water requires accurate and fast response detection techniques. Intentional sabotage events such as water contamination in Japan [48], the incidence of mesophilic *Aeromonas* within a public drinking water supply in Scotland [49] and occurrence of *Aeromonas spp.* in tap water in Turkey [50] triggered awareness on the need to have high-accuracy sensors installed along the water distribution system (WDS). Since then, obtaining optimal sensor placement has been widely explored for the security of WDS by utilizing model-based event detection technique. Optimal sensor placements, detection likelihood, expected contaminant concentrations and affected populations could be predetermined using detection algorithm by obtaining signals from conventional water quality sensors.

A surrogate approach for contamination detection is also suggested by the EPA to determine irregularities of water quality parameters obtained from sensing mechanism for an early warning of possible presence of contaminants [51,52]. Obtaining an early detection system using multiple sensor data station on-site is more beneficial compared to the data from a single site [53]. Most of the water quality parameters are used as primary indicators for contamination events in WDS, which are obtained from an online database, such as CANARY Database [54–57]. The main objective of the event detection method is to: (1) identify the possibility of event occurred, (2) characterize the event into subgroup (e.g., spatial area, time duration and severity level), and (3) detect contamination as accurately and early as possible. Achieving optimal time responses are superior for early indications of potential contaminants in WDS [58]. To minimize the spread of outbreaks, rapid and sensitive detection of pathogens is important.

As many different types of contaminants could present in WDS, case-to-case approach is necessary for accurate qualitative and quantitative analysis. To tackle the issues, handheld detection devices such as microfluidics sensors, miniaturized biosensors as well as portable spectroscopy are widely considered. Nowadays, lab-on-a-chip platforms that require microscopic amount of fluids (10^{-9} – 10^{-18} l) are achievable for sampling, filtration, pre-concentration, separation and detection of biomolecules or analytes. Microfluidic composes channels with dimension up to hundreds of micrometers (μm). The integration of microfluidic sensors constitutes a multi-disciplinary theorems for sensing [59]. This kind of sensors is widely used in biomedical, science, genomics, forensics and environmental studies and for immunology or biochemistry applications [60].

The most frequently used vibrational spectroscopy instruments in water monitoring technology are Infrared (IR) and Raman spectrometers. Vibrational spectroscopy is based on the correspon-

dence of radiation absorptions to a discrete energy level, which is generated from the stretching or bending of atom molecule vibrations at frequencies of 1012–1014 Hz. In recent years, the integration of biosensor and spectroscopic techniques with digital microfluidic (DMF) devices is widely explored for the use in contaminant detections. The popularity of such integrated technique can be reflected by the number of technical papers published in the literature [61–64]. However, two major concerns regarding the DMF device are the complete elimination of analytes from sensing surface of DMF and disassembly of target analytes from the biosensor receptors for every sensing cycle [65].

This paper intends to provide an overview on the developments of water monitoring technologies for both biological and non-biological contaminants determination. As detection mechanism varies with water contaminants, extensive review on analysis and data mining is also provided. This review emphasizes the current trend in water monitoring technologies and compares their performances with conventionally used methods. Significant limitations and drawbacks of the techniques are also discussed and recommendations are provided for future development in the water quality monitoring. Nevertheless, some analytical methods such as photo-acoustic, ultrasonic and microwave spectroscopy are not included in this review as they are very limited in applications.

2. Discontinuous (sample-based) methods

2.1. Biological contaminants

Microbiological parameter in tap and drinking water focuses on the detection of various pathogens using indicator organisms. Transmission route disease, also known as faecal-oral route, occurs when pathogens in faecal particles are transmitted into oral cavity of another host. According to the European Commission Drinking Water Directive, indicator bacteria *E. coli* and *Intestinal enterococci* should not be present in 100 mL of water volume [66]. The proliferation of water monitoring technology has prompted awareness over the safety of microorganism contaminants to human health and environment. Despite the advancement of the other techniques, the conventional culture-based methods are still in-use for the detection of microbiological parameters in water [67].

2.1.1. Multiple tube fermentation (MTF) technique

Multiple tube fermentation (MTF) technique is one of the standard laboratory methods that can be used to detect microbiological activity in water samples. The MTF technique is executed in a three-stage procedure which is known as presumptive stage, confirmed stage and completed test [68]. Presumptive stage consists of a series of tube incubation process resulting in the formation of gas indicating positive presumptive test. Enumeration procedure for each bacterium sample is executed using suitable broth medium in the presumptive phase [69]. The inoculation process of testing tube samples should be performed instantly after any gas formation occurs. This procedure is known as confirmation stage.

According to EPA's *Standard Methods 9131 for Total Coliform: Multiple Tube Fermentation Technique* [70], completed test is finalized by gas formation and the presence of bacteria in the culture colonies. Detection of coliforms is often carried out by using MTF technique [71]. The concentration of bacteria in water samples is evaluated by examining a series of tubes containing suitable selective culture medium and dilutions of water sample. Formation of turbidity will occur accordingly due to the microbial growth and the results are expressed in terms of statistical estimation of the mean, known as the most probable number (MPN).

The presence of coliforms bacteria, known as 'indicator' organisms, is identified based on the MTF technique with an A-1 medium

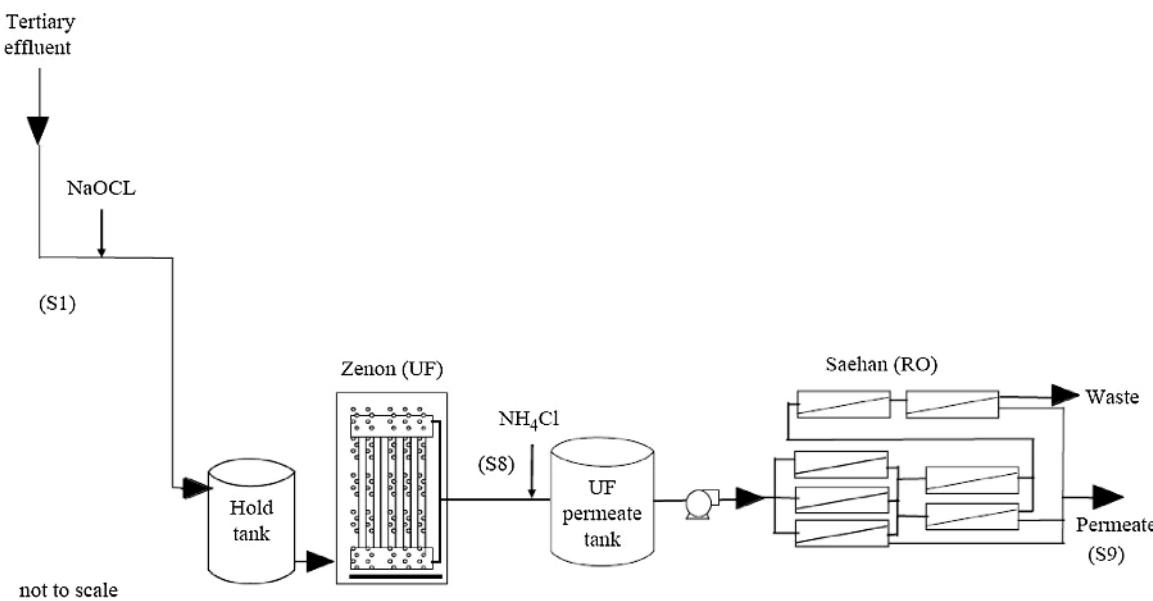


Fig. 2. Process flow diagram of a submerged Zenon ZeeWeed™ 1000 (ZW1000) ultrafiltration unit integrated with a multi-pass reverse osmosis unit (Synder et al. [101]).

for a MPN test procedure described in the *Standard Methods for the Examination of Water and Wastewater* [72]. Using this technique, various types of coliform bacteria such as *E. coli*, *Enterococci*, *Salmonella* and *Bacillus* could be easily detected in different water samples [73–76]. On the other hand, MTF technique was also found to be effective in the process of yeast isolation and identification of *E. coli*, *Enterococcus* spp. and *P. Aeruginosa* densities [77]. The result showed the positive correspondence between yeast densities and counts of standard indicator, suggesting that yeast may also be considered as an organism indicators of sewage contamination.

MTF method was also used to determine *E. coli* in the water samples and high detection rate (100%) could be achieved compared to only 75.5% shown by a membrane filter (MF) method [78]. The presence of faecal contaminants in the water samples is confirmed based on turbidity measurements, which are reflected by the increase of water temperature and decrease of dissolved oxygen (DO) content [79]. Reduction in DO content may reduce the survival rate of coliform bacteria in aquatic environments [80]. However, determining DO measurement in surface level water samples is considered irrelevant due to high production of oxygen on its surrounding that leads an indirect proportional relationship between turbidity, DO and faecal coliform [81].

2.1.2. Membrane filtration (MF) method

The MF technique is recognized by the USEPA and UNEP/WHO as a method for detecting biological contaminants of potable water. This technique is capable of isolating and eliminating discrete microbiology colonies in relatively large number of sample volume compared to the MTF technique [82–84]. Similar to the MTF technique, MF method is generally used for major indicator organisms [85]. In the past, both methods have been conducted at laboratory level, but due to the advancement of portable technologies, MF and MTF method are now able to be executed for on-site applications.

Incubation of MF on solid and/or liquid selective media at appropriate temperature allows the growth and development of organism cultures providing a direct count of total coliforms colonies. The choices of temperatures depend on the type of bacteria indicator and the selective media. For instance, *P. aeruginosa*, *E. faecalis* and *Penicillium* were able to be detected using cellulose nitrate membrane filters which after being incubated for 48 h at 44 °C on the solid and liquid selective media [86]. The MF technique

has also been used for retaining virus, known as F-RNA coliphages via incubation using mixed cellulose nitrate and acetate membrane filters [87].

Work conducted by Grabow et al. [88] indicated that theoretical efficiencies of 100% are achievable with procedures governing to direct plaque assays on 100 mL samples and the presence-absence test on 500 mL samples. In addition, positively-charged filter media have been widely used for recovering bacterial viruses and phage. However, poor detection of viruses and phage was experienced following poor absorption and inactivation caused by extreme pH level exposures [88,89]. In general, microfiltration and ultrafiltration membranes are preferable to be used for the filtration purpose due to their appropriate range of surface pore sizes that could retain microorganism effectively [90]. Previous work has shown that the removal of bacterial contaminants in water could be carried out using microfiltration-based method by observing the *in-vitro* nitric oxide production and binding response of Limulus amebocyte lysate (LAL) assays [91].

Instead of using single type of membrane filter, simultaneous use of microfiltration and ultrafiltration membrane filters were attempted in recent years. Xiong et al. [92] has established the first quantitative assessments in the water samples using integrated method and reported the total organic carbon (TOC), total suspended solid (TSS) and turbidity of the samples to be 95–630 mg/L, 180–1300 mg/L and 150–1900 NTU, respectively. The integrated method has also been used to reduce peroxidase activity in red beet stalks in order to maintain natural pigment stability [93]. Results showed that it achieved more than 99.5% reduction of peroxidase activity and 99.9% reduction in turbidity.

Previous work showed the applications of membrane bioreactor (MBR) process based on aerobic fermentation method [94]. MBR combines the membrane filtration process with reactor that involves biological reactions. The efficiency to monitor microbial population in water sample depends on the attachment of bacteria onto inert material inducing high biomass [95,96]. Integration of bioprocess and MF method provides a better efficiency in the removal of bacteria as, demonstrated by Adan et al. [97] in which a derivation of photo catalysis-microfiltration hybrid system was used to remove *E. coli* from water source.

MF, selective medium broth and culture plate methods have become the standard (ISO 16654:2001) to monitor the presence of

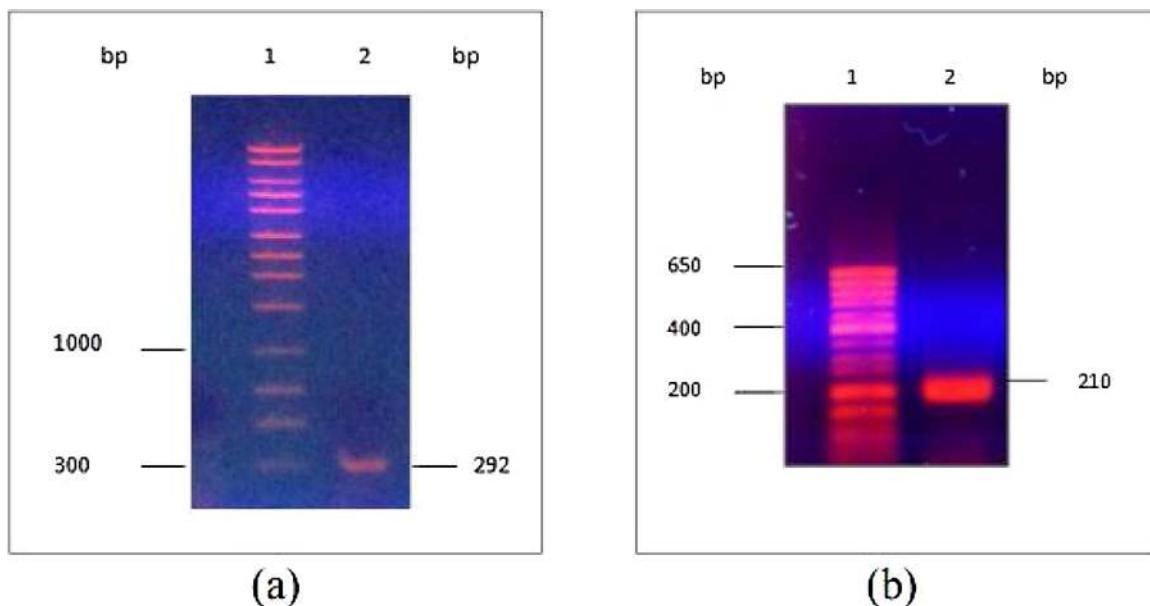


Fig. 3. A typical PCR amplification product optimized using *E. coli* O157:H7 strain. (a) Lane 1: 1 kb DNA ladder; Lane 2: 292 bp O157 gene amplified with Rfb F and R primers. (b) Lane 1: 50 bp DNA ladder; Lane 2: 210 bp SLT-I gene amplified with SLT-I F and R primers (Imtiaz et al. [109]).

Table 2
Primer sequences and positions [114].

Primer	Position	Target	Sequence
EUB f933	933–954	Bacteria, regions V6–V8	5'-GC-clamp-GCACAGCGGTGGAGCATGTGG-3'
EUB r1387	1387–1368	Bacteria, regions V6–V8	5'-GCCCGGGAACGTATTACCGG-3'
GC-clamp			5'-CCCGCGCGCGCGCGCGCGCGGGCGGGGCACGGGGG

E. coli and other pathogens in water [98]. However, the drawbacks of employing these methods are time-consuming, laborious and low sensitivity in detecting contaminants at low concentrations [99]. Limitation of selective culture or immunological methods due to lack of consistent differentiation in phenotypic traits may also affect detection accuracies [100]. The efficacy of various membranes was elaborated by Snyder et al. [101] in which MF is capable of reducing concentration of contaminants with specific properties. It was reported that the degree of contaminant removal was highly dependable on the characteristic of the membrane and the molecular properties of the targeted analytes. Microfiltration and ultrafiltration membranes showed the least value of contaminant removal whereas reverse osmosis membranes are capable of removing almost all investigated contaminants [101]. Fig. 2 illustrates a process that combined ultrafiltration and reverse osmosis as advanced treatment process for wastewater. The findings indicated that reverse osmosis membrane could remove almost all targeted contaminants, achieving values below the method reporting limits (MRL).

2.1.3. DNA/RNA amplification

DNA amplification is used to detect molecular biology by amplifying a single copy or a few copies of targeted DNA molecules to produce specific DNA particles *in vitro*. Polymerase chain reaction (PCR) invented by Kary B. Mullis was the first DNA amplification method designed to study particular DNA molecules [102]. Traditionally, replicating DNA sequence requires days or weeks to complete. But, with amplification of DNA sequence using PCR, it only takes several hours [103]. Because of its high detection sensitivity and level of amplification, PCR is capable of replicating minuscule amount of DNA sequences and is extremely useful in commercial uses, including genetic identification, forensics, quality control industrial applications and *in vitro* diagnostics.

In general, PCR amplification reaction constitutes three major elements: (1) a thermo-stable DNA polymerase, (2) a mixture of deoxynucleotide triphosphates (dNTPs) and (3) two oligonucleotide primers [104,105]. One cycle amplification denotes a series of temperature and time, hence amplifies the amount of targeted DNA sequence after reaction takes place. There are three steps in PCR protocols, i.e., denaturation at 93–95 °C for 1 min followed by 45 s annealing at 50–55 °C and 1–2 min elongation at 70–75 °C [106]. Modification on the standard PCR protocol could also be performed using different techniques, e.g., a multiplex PCR protocol for detection of *E. coli*, *Campylobacter* spp. and *Salmonella* spp. in both drinking and surface water [107–109].

Fig. 3 shows the PCR amplification product developed using oligonucleotide primers Rfb and SLT-I for the purpose of detecting *E. coli* O157 and *E. coli* virulence gene SLT-I in drinking water. Optimization of PCR protocol was tested with *E. coli* O157:H7 strain. Another example is conventional hot-start PCR technique for the detection of *Actinobacillus actinomycetemcomitans* by heating the reaction components under DNA melting temperature before mixing with polymerase to reduce non-specific priming amplification [110,111]. A touchdown PCR amplification was then established to identify the presence of bacteria in aquatic samples by gradually decreasing the primer annealing temperature in later cycles [112–114]. The analysis of bacterial water contaminant was performed using a universal conserved bacterial 16S rDNA sequence, which are specifically used for amplification of 16S rDNA fragments with GC-clamp-EUB f933 and EUB r1387 primers (Table 2).

Although the PCR method has a high detection successful rate, it is still associated with several limitations that include low sensitivity to certain classes of contaminants and reduction of amplification efficiencies in the case where inhibitors are present in water samples [115]. Over the past few decades, there has been a diversity of newly developed technologies to overcome these

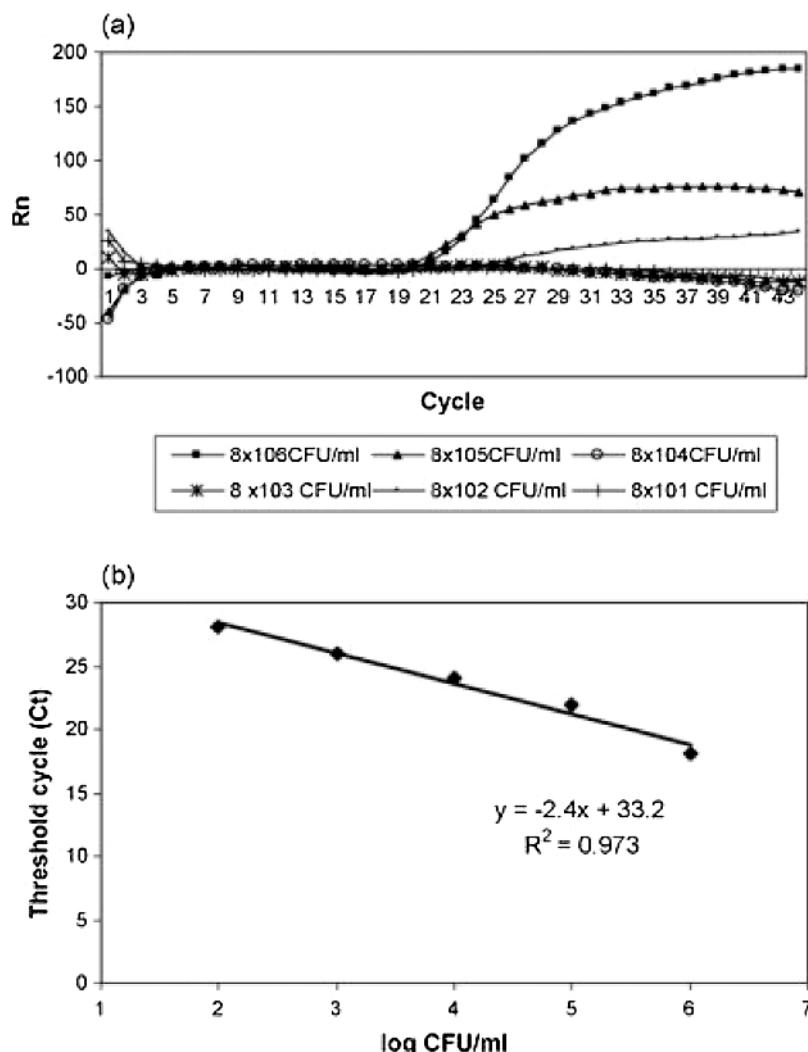


Fig. 4. (a) Sensitivity of real-time PCR assay consist of ten-fold serial dilutions of DNA template isolated from *E. coli* JM109 strain ATCC 43985 and (b) Linear curve for real-time PCR assay with wide range of initial target concentrations (from 10^2 to 10^7 CFU mL) (Sandhya et al. [119]).

limitations. Some of the examples are quantitative real-time PCR assays (qPCR), reverse transcription real-time PCR (RT-qPCR) protocol, loop-mediated isothermal amplification (LAMP) technologies, strand displacement amplification (SDA), ligase chain reaction (LCR), rolling circle amplification (RCA), helicase-dependant DNA amplification (HDA) and the most recently developed random amplified polymorphic DNA analysis (RAPD) [116,117].

The qPCR automates both amplification and detection in quantitative measures. The simplified quantification is obtained through quantification cycles (Cqs) which are determined by fluorescence threshold or maximum second derivative [118]. Exponential phase in qPCR technique can be continuously observed for 30–50 Cqs and can be used to estimate the initial number of targeted DNA. The use of a qPCR assay to positively detect *E. coli* O157:H7 strains in drinking water was carried out using molecular beacons (MBs) oligonucleotide probes [119], InstaGeneTM matrix from Bio-Rad specially formulated 6% w/v Chelex resin [120], minor groove binding (MGB) probes with 6-FAM (6-carboxyfluorescein) [121] and propidium monoazide (PMA-based) qPCR assay [122]. Quantitative PCR assay provides the possibility of quantitative analysis for *E. coli* target by using formulated structural quantification curve as shown in Fig. 4. Such measure reduce the false positive results during analysis.

There have been several commercially designed real-time PCR assays for the detection of pathogens (e.g., *F. tularensis*, *B. anthracis* and *Y. pestis*) with high detection sensitivity and diversity of pathogen detection capabilities [123,124]. The qPCR techniques have been found useful for the detection of *Naegleria* sp. by referring to melting curve analysis SYTO9 and qPCR TaqMan assay [125–127]. Melting curve analysis is beneficial for the manipulation key conditions, including temperature interval and time delay before data are collected for each step. For example, melting curve that provides three informative peaks within temperature range of 79–86 °C (Fig. 5) can be used to distinguish species based on the positions and height of the peaks obtained.

Another extended version of standard PCR method is the RT-qPCR, which is a useful technique to identify specific messenger RNA (mRNA) as well as DNA from any type of living microorganism cells, either qualitative or quantitative measures [128–130]. This technique evolved tremendously after the introduction of hybridization on target DNA sequence using an oligonucleotide probe. The RT-qPCR technique involves the hybridization of oligonucleotide primer to produce a complementary DNA (cDNA). This process of deoxyribonuclease I (DNase I) is used to eliminate contaminated DNA that triggers false positive results. The application of RT-qPCR assay approach has been used in detecting pathogens such as mRNA in *E. coli* cells [131], family of Filoviridae

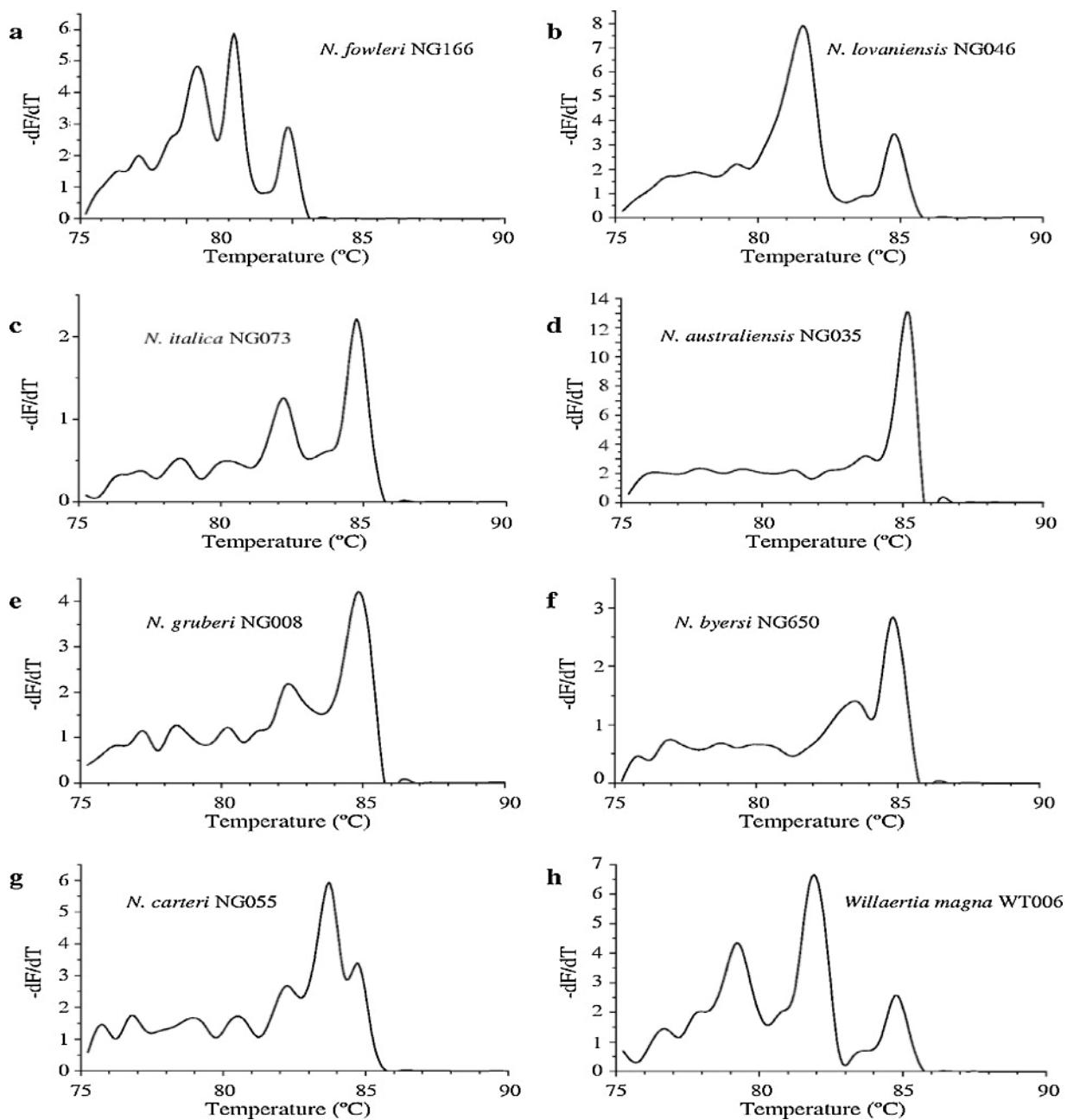


Fig. 5. Melting curve analysis of the 5.8S rDNA/ITS product of seven *Naegleria* species: (a) *N. fowleri*, (b) *N. lovaniensis*, (c) *N. italic*, (d) *N. australiensis*, (e) *N. gruberi*, (f) *N. byersi*, (g) *N. carteri* and (h) *Willaertia magna* (Robinson et al. [125]).

viruses and RNA transcription from Ebola viruses [132], cereulide-producing *Bacillus cereus* [133], RNA molecules of *Salmonella* [134] and rotaviruses and coronavirus in feces contaminations [135]. The reverse transcription PCR (RT-PCR) techniques proven to provide high efficiencies by amplifying both DNA and RNA sequence. Conventional PCR methods meanwhile only amplifies DNA. As reported by Wang et al. [136], RT-PCR has high detection sensitivity on bacterial quantity (as low as one bacterium) compared to those of PCR-based techniques.

Although PCR-based techniques could show significantly higher positive detection rate, performing accurate thermal cycling and utilization of sophisticated instrumentation (e.g., fluorescence measurement) require higher throughput and longer time [137–143]. Therefore, an alternative to PCR is isothermal-based amplification methods. This method can be carried out without undergoing repeated thermal denaturation procedure and does not

require sophisticated instruments [144]. Typically, loop-mediated isothermal amplification (LAMP) mechanism comprises two pairs of primers (inner and outer) and are dependable to strand displacement synthesis of DNA polymerase to produce loops amplifications [145]. LAMP has been widely used for diagnosis of biological specimens and is commercially available for environmental monitoring applications [146–148]. It has high sensitivity and rapid detection capability as well as greater ability to quantify several bacteria [149,150]. A pilot study was conducted to detect *Staphylococcus aureus*, *E. coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumonia*, *Stenotrophomonas maltophilia*, *Streptococcus pneumonia*, and *Acinetobacter baumannii* using quantitative LAMP (qLAMP) with better identification ($P < 0.001$) than that of traditional culture-based method [151]. The functionality of loop primers designed for LAMP assays improved the detection specificities and sensitivities by several magnitudes [152]. This was proven by Sotiriadou and Karanis

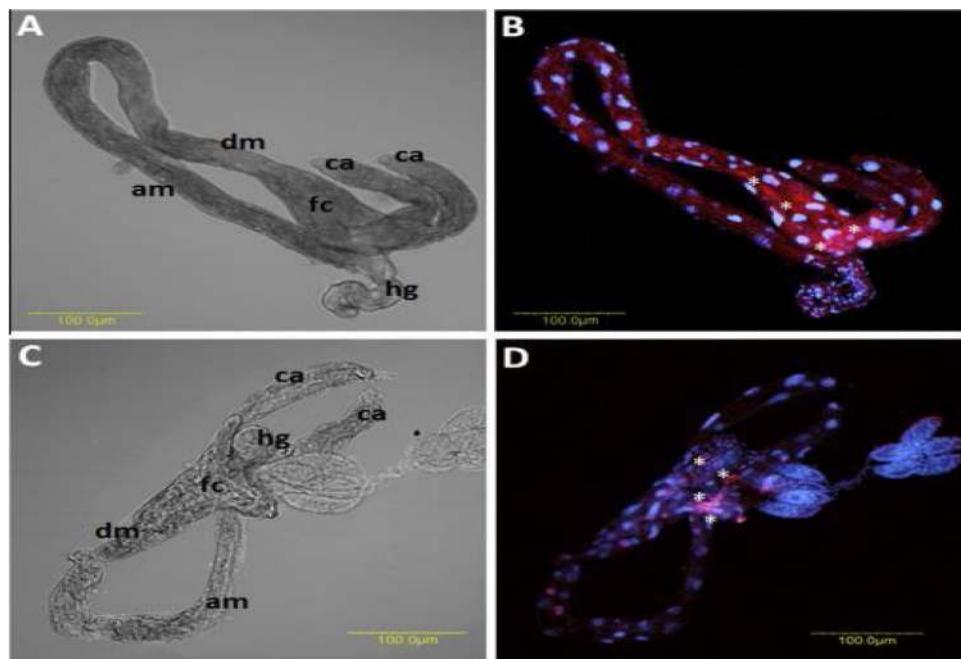


Fig. 6. mRNA localization of cyp6CM1 and ABC transporter genes in midguts of the whitefly *Bermisia tabaci* using FISH, (a) bright field of a *B. tabaci* midgut, (b) FISH fr mRNA localization on this midgut showing cyp6CM1 gene expression mainly in the filter chamber, (c) bright field of a *B. tabaci* midgut and (d) FISH for mRNA localization on this midgut showing an ABC transporter gene expression mainly in the filter chamber. (Definition – am: ascending midgut; dm: descending midgut; ca: caeca; fc: filter chamber and hg: hindgut) (Kliot et al. [169]).

[153], by employing LAMP assays approach for the evaluation of *Toxoplasma* in water samples based on amplification of B1 and *TqOWP Toxoplasma* genes with 100% success detection rate. Separately, a gene amplification using hydroxyl naphthol blue could successfully detect *Naegleria fowleri* within 90 min reaction time with a Kappa coefficient of 0.855 [154].

Based on the previous discussion, one can realise that LAMP method is less expensive to perform as it involves no real-time thermal cycler. Furthermore, it has greater sensitivity and can be potentially deployed for on-site water contaminant detection. Gallas-Lindemann et al. [155] reported that *Giardia* spp. and *Cryptosporidium* spp. could be detected using LAMP assays with 42.7% and 43.6% detection rate, respectively compared to 33.5% and 41.9% shown by the conventional nested-PCR. Besides, the LAMP technique offers 100% detection sensitivity with LOD of 50 fg/mL compared to the conventional PCR method [156]. Integrating the extension method with the standard PCR could provide faster, less false positive indicators, better compatibility for detection of multiple pathogens [157–159]. Moreoever, LAMP was comparable to the qPCR method for surveillance of *Dehalococcoides* spp. in groundwater using six LAMP primers designed for each three RDase genes [160].

Enteric viruses generally yield between 105 and 1011 virus particles per gram of individual stool [161,162]. There is no direct relation between the occurrence of bacteria and enteric viruses, hence suggesting the need to separately evaluate the presence of viruses in water supply. Culture-based method is not the preferable approach for evaluating enteric viruses as it requires higher analysis cost and longer analysis period. It is also found to have complexity related to the permissive system of some non-cultivable viruses *in vitro* [163].

According to Kim et al. [164], molecular detection done by qPCR and qRT-PCR methods could overcome the issues regarding the sensitivity and analysis time. Huang et al. [165] and Jiang et al. [166] also agreed that in comparison to the conventional nested PCR approach, the qPCR method offers better efficiency (>95%) in quantifying enteric adenovirus serotype in environmental waters.

However, no method is completely perfect by taking into account the principle and procedure of each method. For instance, the detection of pathogenic viruses is obtained in a disinfection procedure, but this method is not suitable for the detection of coliform bacteria due to low concentration of bacteria indicator [167]. The generation of DNA-based amplification method has evolved due to demands in producing combined method of detection with higher specificity and rapidity. Probe based real time loop mediated isothermal amplification (RT LAMP) assay was then introduced for the quantification of *Salmonella* invasion gene (InVA), aiming to achieve significantly higher sensitivity [168].

2.1.4. Fluorescence *in situ* hybridization (FISH)

In situ hybridization is a technique that enables the detection, identification, localization and enumeration of microorganisms. Cellular component and targeted analytes can be visualized via fluorescence probe based on fluorescence *in situ* hybridization (FISH) technique. The FISH technique has been used in a wide variety of research fields such as cytogenetic, microbiology and genetic diagnostics applications [169]. There are a few important factors such as probe design and fluorophore selections that need to take into consideration before execution of FISH experiments. Commonly used probe is 15–30 nucleotides long that is labelled with fluorophore of 3' or 5' at each end. These probing designs are specifically used for the detection of microorganism and mRNAs as shown in Fig. 6. One of the earliest application to embrace the usage of FISH technique was microbial ecology. Similar to DNA amplification method, the most commonly used DNA probe is 16S rRNA sequences for the detection of bacteria in living tissues as well as in aquatic environment samples. In recent years, the FISH technique has been applied in microbiological monitoring field. However, FISH has low fluorescence signals which limits the detection factor to the specified microbial community [170]. To address this issue, a multi-labelled FISH technique is recommended so as simultaneous detection of microbial groups could be achieved by improving the fluorescence signals [171].

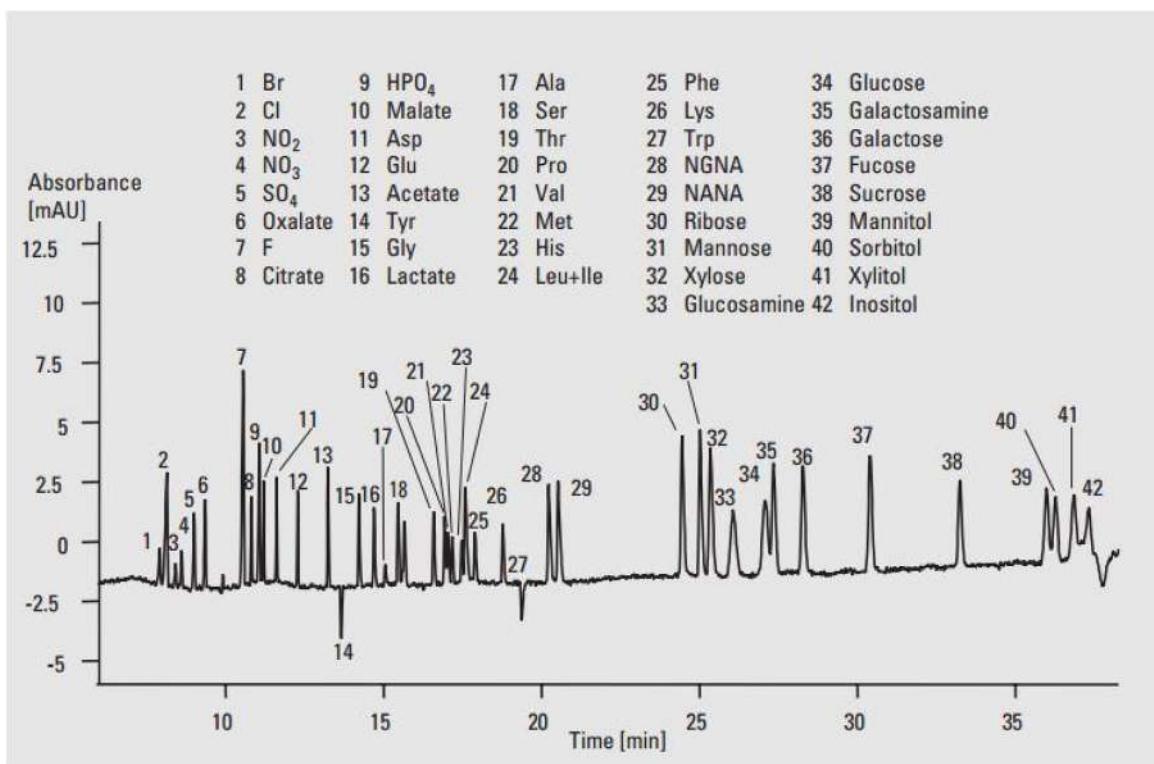


Fig. 7. A typical electropherogram of a 43-component (7 inorganic anions; 5 organic acids; 16 amino acids; 15 carbohydrates) anion standard mixture (Soga et al. [187]).

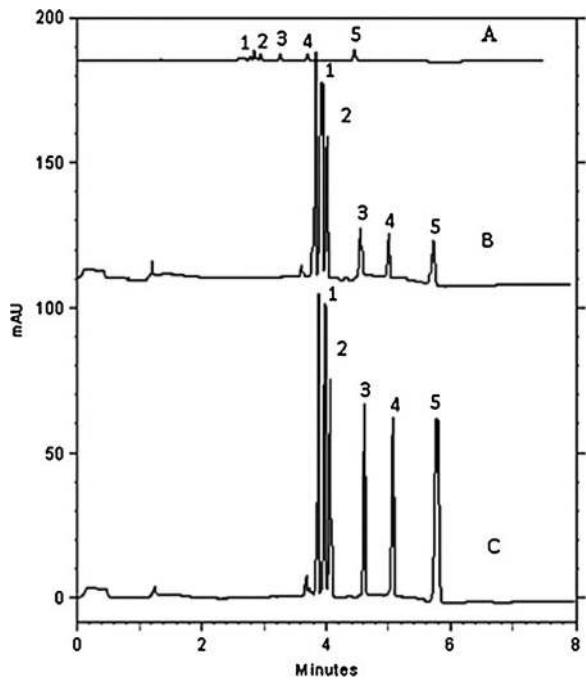


Fig. 8. Comparison of the electropherograms obtained by (A) conventional MEKC method (sampling: 1.0 µg/mL of the triazine herbicides in BGS, direct injection at 0.5 psi for 5 s), (B) the sweeping-MEKC method (sampling: 0.5 µg/mL in 50 mmol/L H₃PO₄ (pH 2.5), direct injection at 0.5 psi for 120 s) and (C) the combination of DLLME with the sweeping-MEKC method (sampling: starting from 5.0 mL of 10 ng/mL water sample for DLLME). Peak identifications: 1: prometon; 2: simetryn; 3: propazine; 4: atrazine; 5: simazine; u: unidentified peaks (Li et al. [192]).

Previous study reported the detection and quantification of β -Proteobacteria and Cytophaga-Flavobacterium cluster in an urban river after 3–7 days of formation [172]. A similar technique was

used to detect various members of Cytophaga-Flavobacterium cluster, classes of *proteobacteria* and members of *Planctomycetales* in an aquatic environment, ranging counts of 50% cells detection [173].

A FISH-probe using *Bacillus subtilis* 16s rRNA has been also reported with the aim of distinguishing targeted nucleotides between 465 and 483 genes [174]. However, the results showed that FISH method was not able to identify strains, i.e., *B. altitudinis*, *B. cereus*, *B. gibsonii*, *B. pimulus* and *B. megaterium*. The development of FISH technique over the past several years has increased its usage in determining various types of mRNA and DNA molecules [175]. Fluorescent nanomaterials, also known by quantum dots (QDs), have been introduced to improve fluorescent-brightness, photochemical cohesion and coherence emission spectra [176–178]. The QD-FISH method offers the ability to detect specific target genes. For instance, synthesis of biotin-streptavidin deoxyuridine triphosphate (dUTP) labelled DNA probes via PCR using dNTP mixture enable the detection of *Ectromelia virus* (ECTV), a member of the *Poxviridae* family [179], resulting a genome detection of 80% after 36 h of post-infection with significant visual of red fluorescence. The detection of green micro-algae, *U. prolifera* using FISH was also carried out targeting the 5S rDNA of *U. prolifera*, *Ulva linza* and *Ulva flexuosa* molecular genes using 5S-1 and 5S-2 probes [180]. Six species of green algae were also tested, however only *U. prolifera* could be labelled by both specific probes. Because of the complex structure of bio-analytes, direct detection and quantification of single-cell bacteria using FISH are rather difficult. Therefore, an additional combination of flax desegregation protocol with quantitative FISH technique is recommended [181].

2.2. Non-biological contaminants

Generally, there are two categories of non-biological water contaminants. Chemical contaminants consist of elements or compound, such as volatile organic chemicals, disinfection by-products and synthetic organic chemicals which can be occurred naturally or

man-made. Whereas, radiological contaminants are from an unstable atom that emits radiation, for instance plutonium and uranium [182]. Another water contaminants that are starting to raise awareness are engineered nanoparticles/nanomaterial. Example of them are metallic nanoparticles (e.g., Ag, Au, and Fe), oxides (e.g., CeO₂, TiO₂ and ZnO) and quantum dots (e.g., ZnS). Although nanomaterials are beneficial for many industrial applications, the release of them may unintentionally promote hazardous occupations to environment and posses health risk to humans.

According to the EPA's Chemical Contaminants – CCL 4 that was recently drafted, it is found that non-biological contaminants are possibly present in tap and drinking water [183]. The majority of organic water contaminants are from industrial activities, environmental degradation, agricultural run-off and naturally-occurring elements. Whereas, inorganic water contaminants are the derivation of natural minerals resulted from erosions and runoff. With respect to the analysis of non-biological contaminants, chemical parameters such as pH, hardness, temperature, dissolved organic nitrogen, total organic carbon (TOC) and chemical oxygen demand (COD) are always considered. According to the WHO Guidelines for Drinking Water Quality (Fourth Edition), a derivation of the tolerable daily intake (TDI) should be taken into account when involving drinking water municipal [184].

2.2.1. Capillary electrophoresis (CE)

Capillary electrophoresis (CE) is a separation technique used to analyzes molecular polarity and atomic radius based on ions electrophoretic mobility. The movements of analytes through electrolyte solutions are directly proportional to the applied voltage, where high electric field leads to faster mobility. CE has the ability to perform separation in capillaries with diameters in mm and in micro to nano-fluidic channels. The CE technique is often related as capillary zone electrophoresis (CZE), however, there are other CE-based methods which include capillary gel electrophoresis (CGE), capillary isoelectric focusing (CIEF) and micellar electrokinetic chromatography (MEKC) [185]. There have been several studies related to the determination of NH⁴⁺, Na⁺, K⁺, Mg²⁺ and Ca²⁺ ions in environmental samples using the CE detection method [185].

The determination of existing unions (nucleotides, metacetylenediaminetetraacetic acid, haloacetics, etc.) in aquatic environments is achievable by analyzing their electrophoretic mobility within detection wavelength of 350/20 nm. Researcher have proposed a method consisting 50-μm straight capillaries with baseline noise modification to determine existing unions [186]. This method was found to be useful for the screening analysis of anions in liquid samples. Anions separation was executed simultaneously using a highly alkaline pH condition to attract a negative charge, triggering migration towards anode as shown in Fig. 7. Because of this, the existing anions in aquatic environment can then be analysed based on their electrophoretic mobility within selective wavelength [187]. CE technique has also been reported in peptide analysis, qualitatively and quantitatively [188].

Analysis on trace chloroanilines in water samples was developed by Pan et al. [189] using CE technique and the method could achieve LOD between 0.01 and 0.1 ng/L for eight aniline compounds within 25 min of detection. Under the optimum conditions, the enrichment factors were obtained within the range of 51–239 and exhibited linear calibration over three orders of magnitude ($r > 0.998$). Water contaminated by herbicide species is contagious to human health and potentially reachable to toxic levels. The identification and quantification of herbicides can be obtained using an extended CE method coupled with low voltage eigenmode expansions (EME) modelling technique [190]. The pre-concentration and detection of environmental pollutants, such as 2,4-dichlorophenoxyacetic acid (2,4-D), 4-(2,4-dichlorophenoxy)butanoic acid (2,4-DB), and 3,6-dichloro-2-methoxybenzoic acid in

water samples were executed using a Box-Behnken design (BBD) and response surface methodology (RSM) related to extraction efficiency. Because of this, herbicides could be detected using a novel MEKC method [191]. The composition of 25 mM borate, 15 mM phosphate, 40 mM sodium dodecylsulfate (SDS) and 3% (v/v) of 1-propanol at pH 6.5 was used as an optimum buffer. A successful LOD ranging from 0.02 up to 0.04 ng/g and LOQ of 0.1 ng/g was reported within the optimized conditions.

A similar method with the combination of online sweeping preconcentration in MEKC method was developed for the detection of five triazine herbicides in water samples [192]. However, under optimized condition, the LOD was slightly different with value shown in a broader range (0.05–0.10 ng/mL) in comparison to the conventional MEKC method as presented in Fig. 8. Due to vast usage of animal-based fertilizers in agriculture, the contamination of water with estrogenic compounds cannot be prevented. These estrogenic compounds were found present in mineral and wastewater samples with an alarming rate. The adjoint detection method proposed by D'Orazio et al. [193] using ammonium perfluorooctanoate (APFO) – based MEKC was effective to detect 12 estrogenic analytes with LOD ranging from 0.04 to 1.10 μg/L.

Since contaminants in water can exist in nano-scale measures, application of nanoparticles together with CE techniques has been presented in order to achieve a safe and sustainable water supply. Sensitivity of analytes detection could be improved using electrophoretic mobility integrated near the inlet capillary. Whether the analytes bind specifically to the sensitive capillary, deployments of several arrays of these capillaries are required for simultaneous analysis. Because of the complexity of equipment arrangements, its industrial implementation is still ambiguous.

The commonly used techniques to distinguish nanoparticles are based on either gel electrophoresis or capillary electrophoresis [194–196]. Detection of engineered nanoparticles (ENPs) such as bioconjugated quantum dots, have been demonstrated using polyacrylamide gel electrophoresis (PAGE). However, due to small pore size of polyacrylamide (PA) gels (<10 nm), the separation method is not practical. Hence, Hanauer et al. [197] introduced the separation techniques for nanoparticles with applications of agarose gel electrophoresis (AGE), with pore size of agrose gel ranging between 10 and 100 nm. The work presented a derivation of silver and gold nanoparticles using polyethylene glycols that, acted as electrophoretic mobility controller. However, findings turned out to be unsatisfactory for gold nanoparticles separation at common CE conditions in comparison to the ICP-MS and UV detection methods [198].

Bioconjugated quantum dots [199,200] and protein-nanoparticle interactions [201] have also been found to be able to distinguish environmental samples using CE. The detection of nanoparticles continued to be favourable in metal and metal oxide nanoparticles separation by using various inorganic buffers as electrolytes [194–196]. Using CE sodium dodecyl sulphate, various nanoparticles, such as Au, Au, Pt and Pd, were able to be detected with resolution as low as 5 nm [202,203]. The CE technique is getting more and more popular and a number of publications on the modification and integration of various CE-based detection methods could be found in literature. These advanced methods intend to overcome some limitations of CE instrument, such as unsymmetrical peak identification [204], poor mobility time repeatability [205], low separation resolution [206] and limited injection efficiency ranging only from 10⁻³ to 10⁻⁷ μL [207].

2.2.2. Gas/Liquid chromatography-Mass spectrometry (MS)

Mass spectrometry (MS) is an analytical tool used to measure molecular mass of targeted sample. Mass spectrometer used for environmental analysis is commonly coupled with a separation

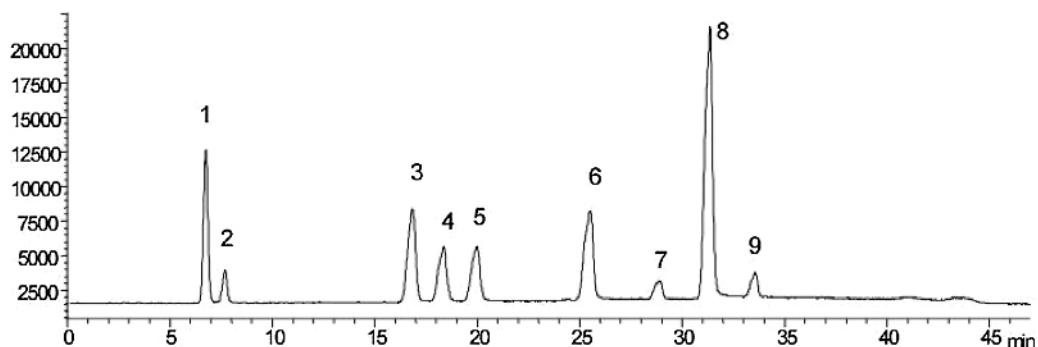


Fig. 9. Chromatogram obtained for the separation of pesticide standards using liquid chromatography: (1) carbendazim, (2) dimethoate, (3) simazine, (4) tebruthiuron, (5) carbaryl, (6) atrazine, (7) diuron, (8) ametryne adnd (9) linuron (Queiroz et al. [210]).

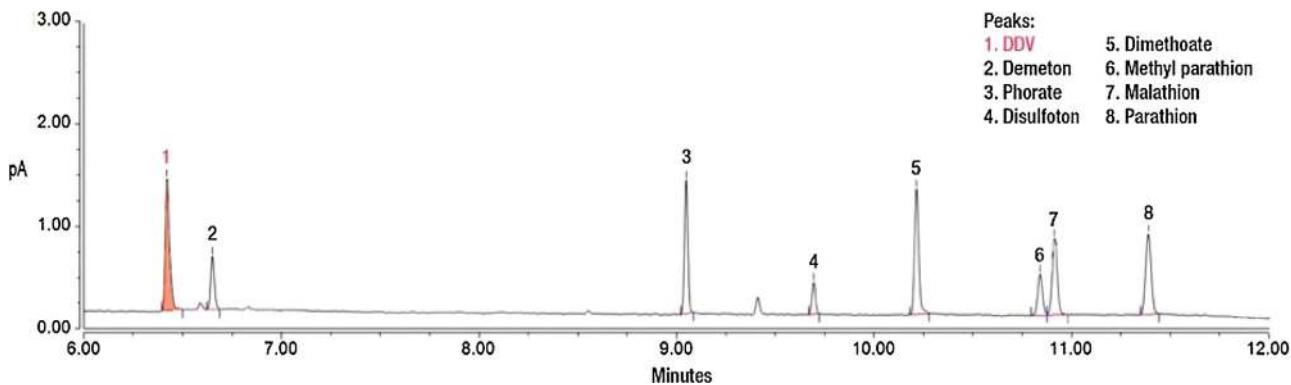


Fig. 10. Chromatogram obtained for separation of pesticides using mixed standard solution (gas chromatograph) (Qian et al. [211]).

method such as gas chromatography and liquid chromatography [208]. Recently, there have been combinational methods for determination of non-biological contaminants in water samples. For instance, Albishri et al. [209] used a UV-based reversed phase liquid chromatography with integration of liquid phase micro extraction for the determination of five organophosphorus pesticides with concentration of 0.01–0.1 ng/mL in tap, well and lake water samples. The derivation of pesticides in water samples has been detected by using a novel approach of sensitive ultrasound-assisted temperature-controlled ionic liquid (IL) liquid –phase microextraction combined with reversed-phase liquid chromatography. Five organophosphorus pesticides were investigated by varying the IL type, IL volume, ionic strength, sonication time, heating/cooling temperature, centrifugal time and speed. In comparison to the conventional liquid chromatography technique, the proposed method improved the extraction efficiency up to 98%. A selected group of pesticides in tap and drinking water was also distinguished using liquid chromatography in which pesticide dimethoate, carbaryl, simazine, atrazine, ametryne, tebuthiuron, diuron and linuron were perfectly isolated (Fig. 9) [210]. A series of gas chromatography (GC) with a nitrogen-phosphorus detector (NPD) has also been reported with the capability of detecting trace amount of eight different type of pesticides presented in drinking water [211]. Despite providing high detection sensitivity (Fig. 10), GC analysis is considered unsuitable for non-volatile and high molar mass compound such as pesticides [210].

Quantifications of nicotine in tap water and wastewater at trace levels were performed using a novel gas chromatography mass spectrometry (GC-MS) with liquid–liquid extraction process and a satisfactory LOD of 2.6 ng/mL was reported [212]. A study on detection of benzene, toluene, ethylbenzene and xylenes in the water sample was conducted by Franendez et al. [213] using the magnetic solid-phase extraction (SPE) method prior to GC-MS technique. The

experiments showed LOD of 0.3 µg/L for benzene and 3 µg/L for other compounds.

Disinfection by-products (DBPs) are very likely to be found in drinking water and are strongly linked to cancer [214,215]. To date, profound usage of GC-MS for the determination of DBPs is due to the wide range of available mass spectral library databases [216]. Unfortunately, GS-MS has limited detection sensitivity, which can only detect compounds with low molecular weight (<800 g/mol) [217]. Hence, a new technique that combined multiple solid phase extraction (SPE), dual-column liquid chromatography high resolution-LCMS and precursor ion elimination (PIE) was proposed by Richardson et al. [218]. Verstraeten et al. [219] and Erickson et al. [220] also employed such technique to analyse public water samples that contained hormones, pharmaceutical personal care products (PPCPs), polyfluoroalkyl substances and herbicides. Barnes et al. [221] validated the occurrence of pharmaceutical contaminants using LC-MS and reported that the concentrations of sulfamethoxazole and carbamazepine that exceeded 0.1 ng/L were recorded in 9 wells while another 5 wells showed 0.07 ng/L concentration. The determination of pharmaceutical contaminants could also be found in work of Llorca et al. [222] in which LC-MS was used to detect 33 analytes. Other works on the detection of sulfamethoxazole and carbamazepine in drinking water and groundwater can be found elsewhere [221–224].

Separately, perfluorinated chemicals (PFCs) are a large chemical compound that are used in wide variety of heavy industries, such as aerospace, automotive, buildings and construction, due to its ability to reduce friction [225]. Unlike other chemical excess, PFCs are frequently released into the aquatic environment due to the massive usage in industrial activities and food productions. As previously reported, LC-MS technique could be used to analyze the content of water samples containing perfluorooctanoic acid (PFOA)

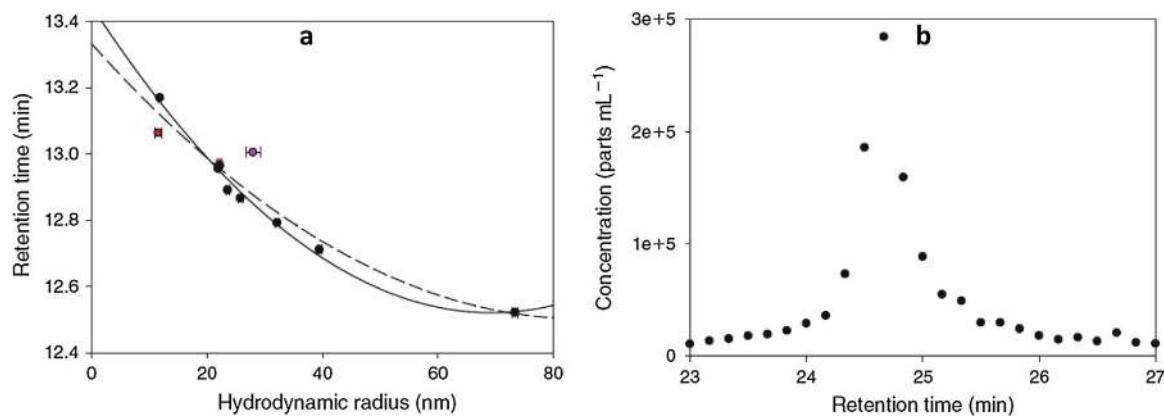


Fig. 11. (a) Calibration curve based on the polystyrene (black points), gold (red point) and silver (pink points) standards and (b) Partial chromatogram for river water sample spiked with 4 µg/L of nAg following separation by HDC and detection using SP-ICP-MS (Proulx et al. [235]). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

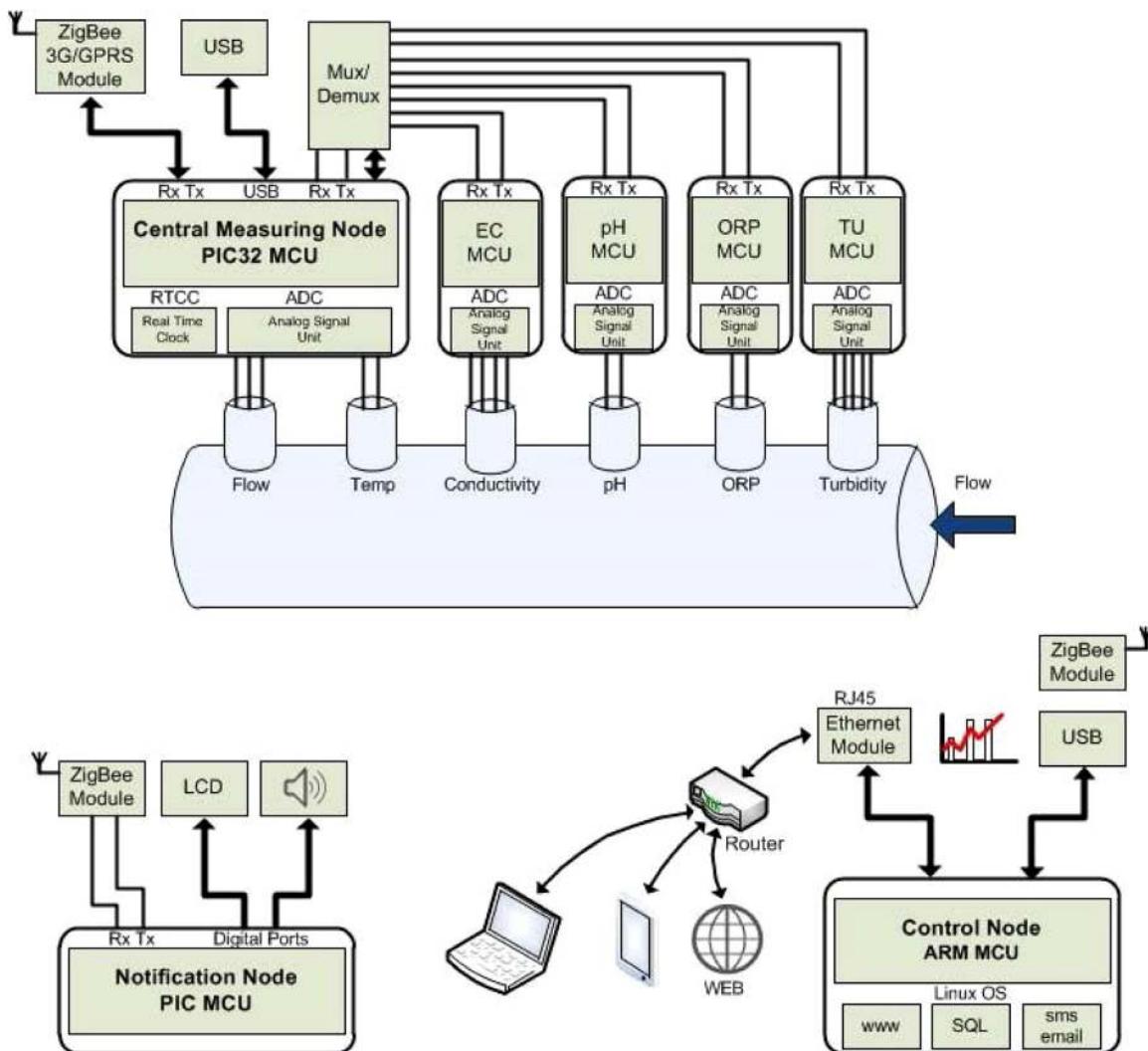


Fig. 12. System architecture sensor placement approach comprising three main subsystems: PIC32 MCU based board used for central measurements, a central node for data transmission via internet, charts and email/message alerts and water quality sensors installation (Lambrou et al. [249]).

and perfluorooctane sulfonic acid (PFOS) with detection limit as low 10 ng/L [226].

Field-flow fractionation (FFF) technique with higher analytes sensitivity and selectivity is a family of analytical separation technique used to extract detailed information on chemical com-

position, functionality and molecular architecture. It was initially introduced by Calvin Giddings to separate macromolecules and colloids [227]. The working principle of FFF is due to the use of external field that is applied perpendicularly to the direction of phase flow within a capillary to derive analytes separation. In comparison to

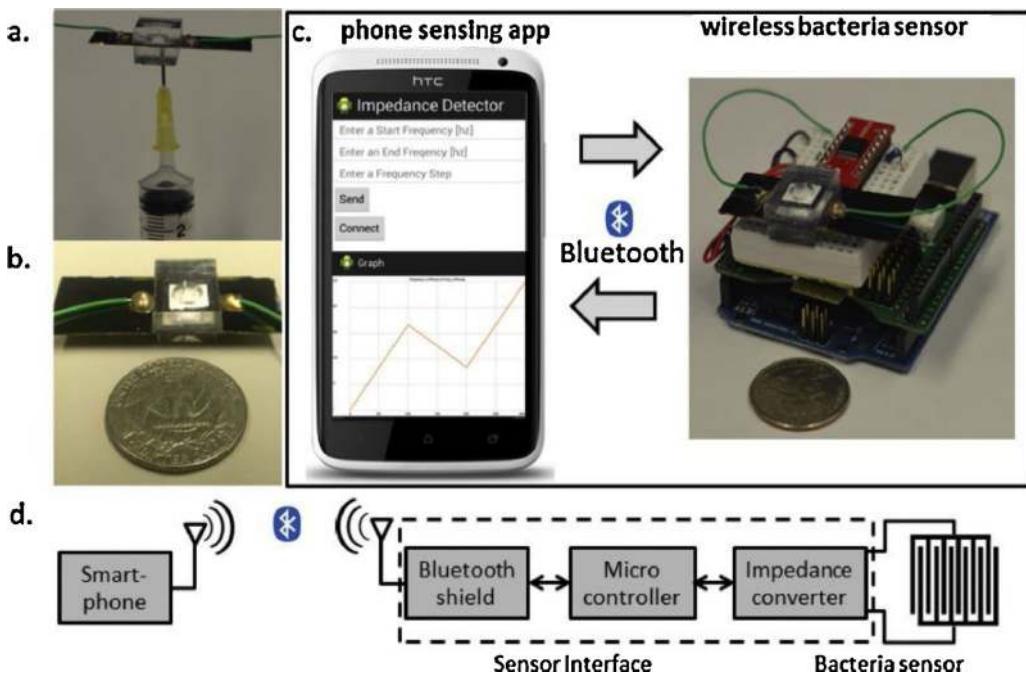


Fig. 13. Wireless mobile phone bacteria sensing system, (a) syringe injection of water sample into sensor package, (b) EIS bacteria sensor package, (c) schematic diagram of smartphone sensing app and wireless bacteria sensor and (d) schematic diagram of wireless sensing system (Jiang et al. [274]).

traditional chromatography approach, FFF is beneficial to those of analytes detection techniques in terms of effective separation components, minimum shear degradation, ultra high resolution, adjustable separation ability and mild operation condition which allows fragile analytes analysis [228–230]. Measurements of colloidal phosphorus in natural waters were demonstrated using an asymmetric FFF technique integrated with high resolution of ICP-MS and membrane filtration [231]. This separation technique comprises two categories, which are centrifugal force-based sedimentation (SdFFF) and perpendicular flow-based (FIMFF) that is mainly used for discrimination of engineered nanomaterial (ENMs) in aquatic environments. High density particles, such as metallic nanoparticles with a relatively large size were detected by SdFFF [232–234] owing to their ability to achieve higher resolution during separations.

The use of analytical techniques to detect and quantify ENPs in environment is limited due to the complex matrices of samples and extremely low concentrations of nanoparticles. A novel approach based on coupling hydrodynamic chromatography (HDC) and FFF has been proposed to separate polystyrene, silver and gold nanoparticles from environment samples [235]. Measurement of hydrodynamic radii of nanoparticles and retention time was conducted using calibration curves and an exceptional polynomial fit from on-line detectors (DLS, SLS) with size ranging between 20 and 80 nm ($R^2 = 0.98$) could be obtained (Fig. 11(a)). Comparison of nanoparticle hydrodynamic radii was further made using manufacturer's off-line detectors (DLS, AUC, SP-ICP-MS) with detection of particle radius of 20.3 ± 0.6 nm at 24.7 min as shown in Fig. 11(b).

By combining methods for differentiation of nanomaterial characterization, the possibility of assessing silver nanoparticles in water samples is achievable. As reported by Antonio et al. [236], the proposed combined technique (asymmetric flow field-flow fractionation, ICP-MS and UV-vis) enable the agglomeration process of silver nanoparticles in artificial seawater. Several works have been carried out to investigate the stability and detection of nanoparticles, including Ag, Au, Se, TiO₂, and ZnO in natural systems [237–239]. The existence of nanoparticles in the aqueous environments must be taken into account since the main factors causing

the derivation are due to surrounding effects, such as temperature, light, oxygenation and total surface area [240,241]. In addition, separation techniques related to inorganic engineered nanomaterial are currently expanding with the aim of achieving nano domain analysis.

A method of coupling ICP-MS with FFF technique was used by Lyven et al. [242] to differentiate iron- and carbon-based colloidal carriers based on the particle size difference. Peak deconvolution analysis was used to quantify and estimate the distribution between organic carbon- and iron-rich colloids and the results indicated the consistencies of chemical properties from two carrier colloids [243]. Although chemical chromatography (HPLC, GC, GC/MS) could offer identification and quantification of analytes, it is only able to detect specific contaminants [244]. Besides, it often involves multi-stage protocols and suffers a number of biases, such as loss of absorption due to the high reactivity [245]. Nonetheless, it must be pointed out that the chemical chromatography is still the preferable method for chemical identification.

3. In-line sensor-based monitoring

Continuous monitoring for microbiological contaminants, especially for chemical contaminants, is a challenging task due to the presence of variety of contaminants at low concentrations [246]. As discussed in the previous sections, standard/sample-based laboratory methods for detection of various water contaminants are often based on the discontinuous approach (off-line analysis). Hence, a sensor-based detection methods such as sensor placement approach (SPA), microfluidics, spectroscopic techniques and biosensors have been tremendously evolved over the last decade.

3.1. Sensor placements approach (water quality sensors)

In contrast to conventional analytical methods, deployment of multiple sensor station in the distribution system is an alternative approach to detect contaminants in a simultaneous manner. In recent years, multi-parametric sensor arrays have emerged to be less cost-oriented and user-friendly to monitor quality of water

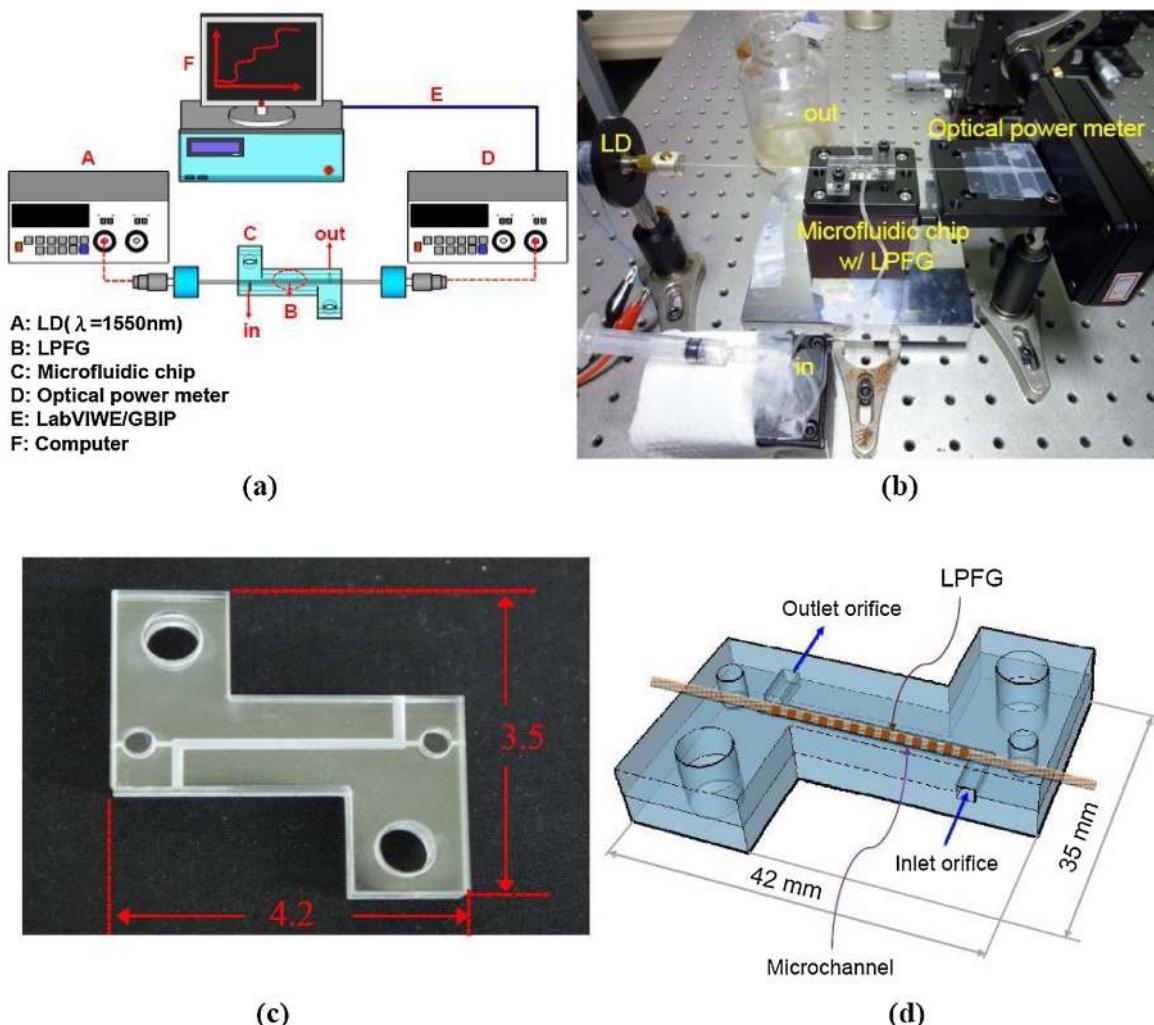


Fig. 14. (a) Schematic diagram of experimental setup for LPFG-based microfluidic chip system, (b) Actual setup of LPFG-based microfluidic system, (c) Microfluidic chip and (d) 3D illustration of the structure and fluidic operation (Wang [275]).

ecology systems [247]. The performance of multiple sensor stations was evaluated by Jeffry et al. [248] using real-time contaminants drinking water and the sensor was reported to be able to detect the existence of aldicarb, glyphosate, colchicines and nicotine in water samples. The data analysis was conducted based on a combination of both distance-based statistics and contaminant transitions.

An alternative low-cost sensor network was developed by Lambrou et al. [249] using multiple electrochemical optical sensors to detect *E. coli* and As in real-time distribution system. The system comprised of six different types of water quality parameter sensors that were able to determine water flow, temperature, conductivity, pH, ORP and turbidity simultaneously as shown in Fig. 12. As water distribution system is increasingly polluted with low concentration hazardous chemical, there is an urgent need for rapid detection.

Similar method was also reported in the work of Che et al. [250], but it was found that EC and UV-254 sensor failed to detect contaminants, owing to the possible hidden responses and fluctuations from water source. Instead of using single sensor researchers were preferable to determine water quality based on multiple sensor placement approach [251–253]. Since obtaining optimal sensor station requires certain expertise, Berry et al. [254] and Tratchman [255] introduced a complex optimization tools, TEVA-SPOT and PipelineNet that employed EPANET to provide guidance for simulation in water distribution systems. The sensor placement

method for water contaminants detection however is relatively complicated [256,257].

Researchers have different views regarding the use of multiple water quality sensor approach. Hypothetically, contaminants could present at any point within a specified time period along the water distribution system. Some contamination events may not be detected because of inaccurate sensor placements and low detection sensitivity [258,259]. Ever since the development of water monitoring technologies, detecting the presence of contaminants in the water supply has become an extremely complex task. Arrays of sensor platforms were implemented to identify unique contaminations according to the sensor capabilities [260]. A study conducted by Ostfeld et al. [261] evaluated the performances of different sensor placements by experimenting with 126 sensor node stations and 168 pipes, which were subjected to a simulation period of 48–96 h per step. Inaccurate analysis might occur as a result of long transmission delays and slow response times in capturing data using sensors [262–264]. In addition, sensor placement approach that requires major in-pipe water distribution system alterations could increase cost [265]. Although sensor placements are among the most analyzed research area, obtaining a 'perfect sensor' when any concentration of contaminants is in contact with the sensor leads to an immediate response, which is considered to be a complete uncertainty.

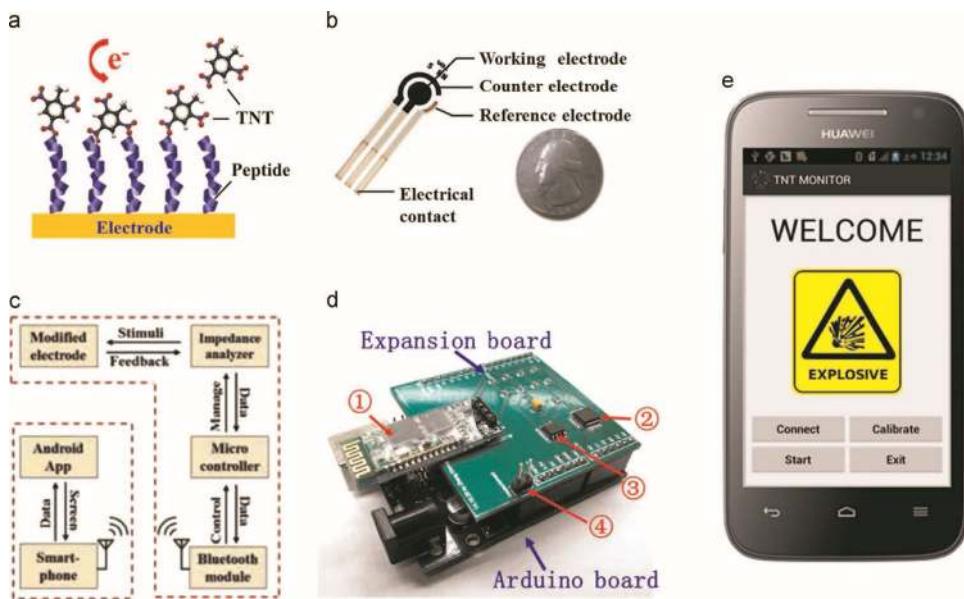


Fig. 15. Smartphone-based impedance monitoring system principle and design for TNT detection, (a) Binding of biorecognition elements (peptides) and TNT analytes on the surface of the electrodes, (b) Schematic of screen-printed electrodes, containing working electrode, counter electrode and reference electrode, (c) Basic diagram of hand-held smartphone-based system, (d) Impedance monitoring device consist of expansion and arduino board and (e) Welcome window of the App in smartphone for TNT measurements (Zhang et al. [308]).

3.2. Microfluidic sensors

Micro-scale technologies have been previously used for detection of non-biological contaminants such as pesticides [266,267], phosphate [268], Hg in water [269], ammonium ion [270] and As ions [271]. In addition, this technology has shown tremendous LOD improvements of biological contaminants. Previous work has successfully identified *E. coli* O157 and *Salmonella typhimurium* using microfluidic reactor with volume of 29–84 nL [272]. Another example of *E. coli* identification that established by Schwartz and Bercovici [273] involved the integration of high concentration labelled antimicrobial peptides (AMPs) within microfluidic channel, aiming to achieve limit of detection as low as 105 cfu/mL and yield 4 bacteria in 2 min. The fabrication of micro-scale sensors can be found in the work of Jiang et al. [274] in which the customized sensors were employed to determine the bacteria concentration in drinking water. Having to adopt the principle of electrical impedance spectroscopy method, the design of a low cost and sensitive bacteria sensor was successfully developed (Fig. 13), aiming to achieve pre-concentration microfluidic-based with LOD of 10 bacterial cells per mL. As shown, wireless system integration is based on Bluetooth receiver via Android cellphone (HTC ONE X), a microcontroller and impedance converter network analyser (AD 5933). The fabrication of microfluidic sensors however varies depending on field of applications.

A miniature microfluidic using long-period fibre grating (LPEG) was designed by Wang [275] and used to measure chloride ion concentration in water samples. The fabricated unit was found to achieve excellent correlation ($R^2 = 0.975$) with light intensity transmitted at 1550 nm. The result showed that the miniature microfluidic could detect chloride ion at concentration of 5.0×10^{-6} mW/mg/L within 2400 mg/L limit of detection. The design and actual setup of LPFG-based microfluidic chip is shown in Fig. 14. Optical-based microfluidic platforms were also found useful to measure various types of chemical and biomolecules at different concentrations [276–278].

Digital microfluidic (DMF) enables the precise control of droplets dispensations on a microliter (10^{-6} L) to picoliter (10^{-12} L) scale for liquid volumes of the fabricated micro-device. Recently,

the studies based on nucleic acid amplification and detection assays using DMF technology could be found in several work [279–281]. The implementation of chip-based nucleic acid assays have led to a significant increase in microfluidic sensor production for the purification and extraction of nucleic acid samples. According to Kaler et al. [282], DMF method is beneficial for proteomics and nucleic acid-based bio-diagnostics application via liquid handling technology, allowing execution of pre-treatments and analysis process on a single device.

A droplet-based sensor embedded on an electro wetting-on-dielectric (EWOD) microfluidics system was also developed by Zengerle et al. [283] by integrating SU-8 polymer micro resonator layered on top of an EWOD plate system. This system only required a single droplet of less than 100 nL of a liquid sample to trigger the sensing process. This trial was the first demonstration of a EWOD-based micro resonator-sensing system with full droplet movement capability. Because of its low power consumption coupled with extremely small sample volume and small data sets, microfluidic sensing platforms are chosen for portable point-of-care (POC) diagnostic devices. In contrast to the low potential of the DMF system for large deployments of chemical and biological micro-reactor applications, an intelligent digital microfluidic system with fuzzy-enhanced feedback for multi-droplet manipulation has been developed by Gao et al. [284]. This pilot DMF prototype aimed to (i) improve complicated image signal processing by using the ability to profile different droplet hydrodynamics, (ii) preserve up to 21% of the charging time using fuzzy-enhanced controllability to enhance the DMF chip's lifetime and (iii) employ automation of multi-droplet routing countermeasure decisions in real-time. The DMF module was made of the following three operation layers: a chip holder, control electronics and a field-programmable gate array (FPGA) board. Volume growth of droplets enabled the execution of sensing module responses that were assembled between two adjacent electrodes. Samples of DI water, phosphate buffered saline (PBS) and 1% bovine serum albumin (BSA) in PBS were injected via a syringe pump into a 4 × 11 mm hole with a constant flow rate of 5 μL/min. The analysis was then carried out using a 0.1 mol/L concentration of Na₂CO₃, PBS, CaCl₂ and FeCl₃.

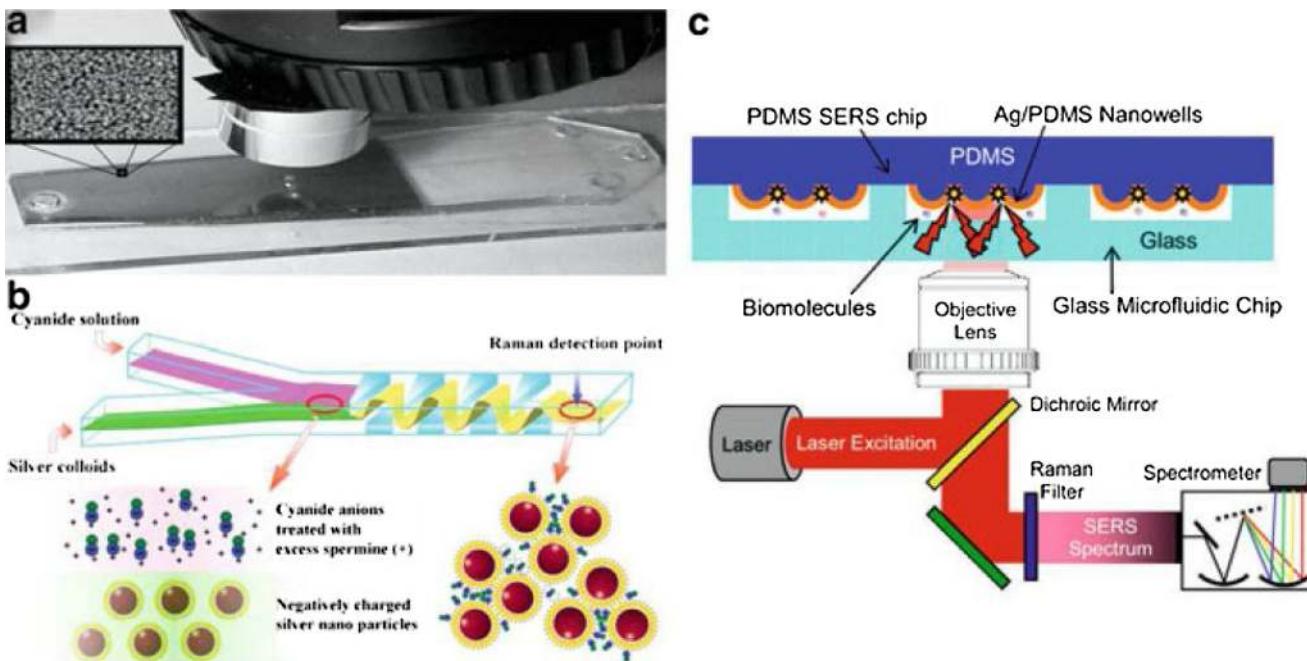


Fig. 16. (a) Depiction of integrated microfluidic SERS device under LabRam Raman spectrometer. The inset shows an SEM image of the silver-PDMS nanocomposite at approximately 90 K magnification, (b) Schematic illustration of alligator teeth-shaped microfluidic channel. The confluent streams of silver colloids and trace analytes are effectively mixed in the channel through the triangular structures and (c) Schematic diagram of the integrated microfluidic chip and the biomolecular Raman imaging system. (Ashok and Dholaki [295]).

Attempts have been made to use an identical approach to detect *E. coli* in drinking water [285], nutritional biomarkers [286], prostate specific antigen (PSA) [287] and *A. acidoterrestris* lysates in milk, juice and water [288]. The abilities of the microfluidic analytical platform to detect water contaminant at a very low concentration and minimum reduction of particle size could promote the usage to distinguish nanoparticles characterization in water samples. A single microfluidic channel has the ability to detect nanoparticles as small as 220 nm [289]. The proposed method utilized microfluidic resistive pulse sensor and was integrated with a submicron sensing gate and two detecting channels using differential amplifier. Detection of CdS electrochemical quantum dots nanoparticles in water sample was able to be detected using integration of hybrid polydimethylsiloxane-polycarbonate microfluidic chip with screen printed electrodes [290]. Under optimized condition, the fabricated microfluidic chip was able to detect CdS QDs with concentration of 50–8000 ng/mL, whilst having LOD of 0.0009 μ A/(ng/mL). For more details regarding the detection and quantification of inorganic nanomaterial using microfluidic chip, one can refer to the relevant review article [291].

There is a huge potential associated with microfluidic sensor fabrication and the introduction of a new cost-effective measure is forecasted to increase in response to commercialization demands [292]. Previously, the issue on chemical contaminant analysis using microfluidic platform was raised due to lack of ability to conduct concurrent analysis [293]. However, many studies have developed a method combining microfluidic and microarrays technologies, enabling multiplex detection of contaminants [294–296]. The pre-treatment of samples however is vital when utilizing chip-based microfluidic sensors. It must also be noted that this additional step may cause the overall process and operation system more complex [297–301].

3.3. Spectroscopic techniques

In principle, spectroscopic technique employs a light electromagnetic radiation source to interact with matter, and requires

a specific probe (depends on the features of a sample) to analyze chemical or biological components. The spectra obtained from different spectroscopic techniques provide an understanding of the properties associated with light electromagnetic radiation and its interaction with matter. Nowadays, there are many types of spectroscopic techniques available for utilization. These include impedance sensing, light emission, vibrational, Raman and surface-enhanced Raman spectroscopy.

3.3.1. Impedance sensing approach

Electrical impedance spectroscopy (EIS) and dielectric impedance spectroscopy (DIS) are the types of impedance sensing technique. Both spectroscopies have been productively used for the bio-detection of targets, such as bacteria and biomarkers. However, EIS is most likely the preferable method for bio-sensing detection applications [302]. It is correlated with microfluidic sensing systems, which implies the integration of electrodes (a working electrode, reference electrode, and counter electrode) that can be either conventional or screen-printed electrodes. Multi-layers of screen-printed electrodes are implemented on flat substrate surfaces. EIS has been widely used in fields, such as medicine, water quality analysis and environmental engineering [303–307].

Previously, the development of impedance screen-printed electrodes has been explored by Zhang et al. [308] to monitor 2,4,6-trinitrotoluene (TNT) in water using a bio-sensing platform (Fig. 15). The integrated system developed with an alternative current (AC) impedance of approximately 20 kHz consisted of an AD5933 impedance analyzer chip, an Arduino microcontroller and a smartphone-based platform. The detection limit concentration is as low as 10^{-6} M TNT-specific impedance properties. Initially, the TNT was purposely attached to the peptide that was bonded to the electrode surface. This was to prevent electron transmission and allow electrode interface impedance. Signals were then transmitted to a smartphone app on real-time basis. The detection of TNT steadily increased at low frequency ranging between 10 and

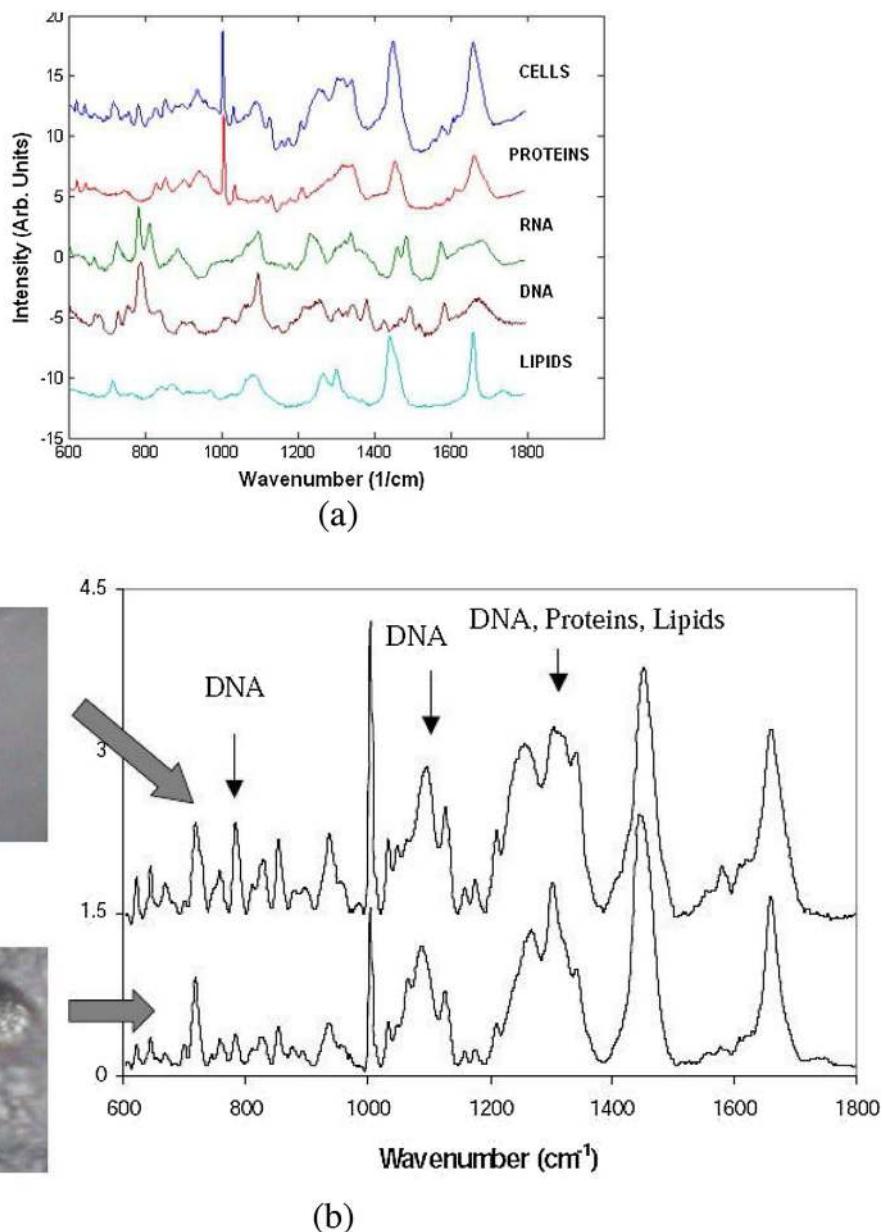


Fig. 17. (a) Typical Raman spectra of a living cell and the main biopolymers components found in cells and (b) Comparison between Raman spectra of living and dead cells (loan [347]).

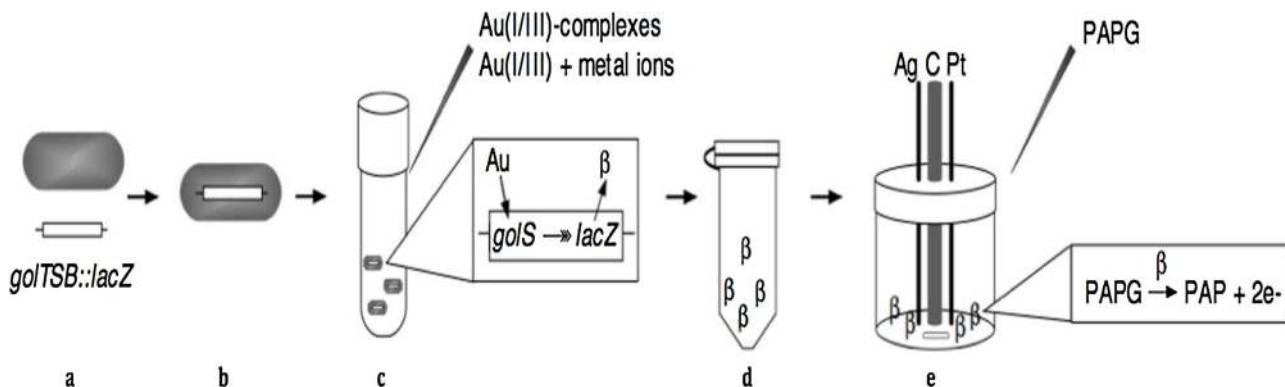


Fig. 18. (a) The *golTSB* regulon regulated by Au ions in *Salmonella enterica* serovar *typhimurium*. A synthetic *golTSB* regulon was made by fusing a promoter-less *lacZ* reporter gene downstream of the *golB* open reading frame as a transcriptional fusion, (b) *golTSB::lacZ* transcriptional fusion was introduced as a single copy into the chromosome of *E. coli*, (c) A single clone was taken for testing and incubated overnight used to inoculate new media, then metals were added for incubation process (16 h), (d) Cells were permeabilized for access to the β -galactosidase produced by *lacZ* gene in the presence of Au and (e) The permeabilized cells were transferred to the electrochemical cell (Zammit et al. [365]).

30 kHz. However, the optimum frequency-dependent impedance measurement for TNT detection was reported to be 20 kHz [308].

On the other hand, Ghaffari et al. [309] developed a low-cost wireless multi-sensor to detect nitrate fertilizer in water sample using DIS platform. The measurements were monitored and controlled via a wireless network system. The DIS platform was developed by assembling commercial microelectronics components that included a multiplexer, a dielectric spectroscopy analyzer, a digital signal controller and a ZigBee transceiver. Similar to this approach, another impedance-based microelectronic sensor, known as a Real-Time Cell Electronic Sensor (RT-CES) was also developed by Xing et al. [310] for dynamic monitoring of cytotoxicants in the water supply. Measurements of cells, including the cell number, viability, morphology and adherence, were carried out using an electrical impedance-based sensor. The system consisted of a circle-on-line microelectrode array that was specially designed to cover almost 80% of the bottom of the sensor area. These micro-electrode arrays were then assembled on a glass slide separating 16-layered wells with 9 mm spacing between each one. The ability of the sensor was to select wells for measurement automatically and to conduct testing continuously.

3.3.2. Light emission/luminescence spectroscopy

The principle underlying light emission or luminescence spectroscopy is the high-energy level absorbance of molecular matter that emits energy as light. The excitation of a high-temperature energy source induces light emitted from matter, which is known as an optical emission. Classifications of light emission spectroscopy include absorbance, reflectance, chemoluminescence, luminescence and light scattering signal and optical-based spectroscopy [311]. Of the reported luminescence platforms, several characteristics have been associated with light emission spectroscopy. For instance, reagent-mediated activity in the form of a light emission medium was employed as an auxiliary reagent to detect and identify contaminants [312]. These emerging techniques have been extensively discussed as a potential tool to monitor water quality *in-situ* [313].

Several studies have acknowledged the potential use of fluorescence spectroscopy in detecting dissolved organic matter (DOM), which can be used as an alternative for standard water quality parameters [314–316]. In addition, the fluorescence spectroscopy approach has also been reported useful to quantify the structural composition of DOM in water samples collected from eight different urban rivers [317]. In this work, the quality of river samples was analyzed using multivariable analysis by segregating the structural composition of DOM that was collected from the west side of Shenyang City, China River. River pollution is increasing at an alarming rate because of high concentration of phosphorus and ammonia nitrogen, released from industrial and domestic sewage [317]. Measurements of ammonia nitrogen ($\text{NH}_4\text{-N}$), nitrate nitrogen ($\text{NO}_3\text{-N}$) and DOC were carried out via the fluorescence-based water quality analyzer known as the YSI 600 multi-probe. Such analyses however required respective reagent to perform.

As the characteristics of lakes and rivers are influenced by climate changes, on-going industrial wastewater release and anthropogenic activities, *in situ* continuous detection and quantification of river water quality are thus urgently needed. A portable chromophoric dissolved organic matter fluorescence sensor (FDOM) is available in the market and can be used to monitor the conditions of different water environments such as wetlands, watersheds and tidal marshes. FDOM can be considered for the detection of DOC concentrations as well as other biogeochemical compositions [318–320]. Niu et al. [321] developed potential applications for real-time water monitoring using FDOM, by taking 218 water samples from Lake Taihu, China as target samples. The CDOM concentration of the lake water was determined using an *in-situ*

CDOM fluorescence tool invented by TROIS GmbH, Germany, with emission wavelengths of 370 and 460 nm.

Apart from the fluorescence-sensing platform, a great deal of the current methods is based on the liquid-based microelectrode glow discharges developed by Wilson and Gianchandani [322]. To improve the detection limit, the liquid electrode spectral emission chip (Led-SpEC) was used to trace Na and Pb concentrations in water samples with detection limit of <10 mg/L and 5 mg/L, respectively. Similar to the glowing detection techniques, a patented submersible spectrofluorometer for the real-time sensing of water quality was developed by Puiu et al. [323]. The submersible spectrofluorometer was designed to extend the fluorescence measurements into an LED excitation spectrum interval between 200 nm and 1100 nm, enabling the instrument to detect chlorophyll-a concentrations as low as 0.2 $\mu\text{g}/\text{L}$. A comparison of the temperature, turbidity, water Raman scattering and fluorescence emissions parameters was also conducted to validate the design.

Similarly, measurements of the DOM concentrations in water were also extracted by using a dipping-based deposition method [324]. A water sensor for global oxygen detection was invented, and it is generally known as anodized-aluminium pressure-sensitive paint (AA-PSP). AA-PSP was anodized in sulphuric acid followed by the dipping of the anodized aluminium coated model into a luminophore solution. It was placed at the bottom of a water tank, and a xenon lamp was excited through a 400 nm band-pass filter. Fourteen-bit CCD cameras were used to capture luminescent images through an optical fibre located 90 mm from the AA-PSP. The work found that AA-PSP was capable of detecting oxygen with a sensitivity of 4.0%/mg/L at a temperature of -2.8°C .

Light emission due to reagent-mediated activations, such as glucose, benzalkonium chloride and chromium (VI), has also been introduced over the years [325]. This approach is similar to the one used by Sharma et al. [326] to evaluate As and *E. coli* contamination in the India River via bioluminescent bio-reporter. For more details about the light emission spectroscopy via an optical fibre sensor platform, one can refer to the work conducted by Ibanez et al. [327] and Chong et al. [328].

In relation to water quality monitoring, the detection of *E. coli* and *B. subtilis* in drinking water by employing quartz tubes as optical light guidance tools with UVC-light emitting diodes (LEDs) has been developed by Gross et al. [329]. The experimental setup consisted of two containers, a soda-lime AR glass and a 100 cm quartz glass, filled with 9 mL of *E. coli* and *B. subtilis*. These samples were compared in relation to their disinfection rates with and without the total reflection of UVC radiation for time interval of 10, 40 and 90 s. The determination of *E. coli* was also discussed in the work of Miyajima et al. [330] in which a fibre-optic fluoroimmunoassay system was employed to monitor the fluorescence dynamics. In addition to the optical spectroscopy approach, a multi-wavelength based optical density sensor for monitoring microalgae growth in real-time has also been established by Jia et al. [331]. In comparison with the previous studies, the sensing system constituted a laser diode module as a light source, photodiodes, a controller circuit, a flow cell and temperature controller housing for the sensor platform. The detection of the microalgae concentration was identified via optical density measurements at wavelength of 650, 685 and 780 nm.

Although emission, optical and luminescence methods are among the most commonly used water contaminant detection technologies, there are associated with several drawbacks. One notable drawback is the sensitivity of light emission spectroscopy to changing temperatures. Because of this, it requires expert guidance during monitoring process [332]. Furthermore, as the

techniques are sensitive to illumination, extensive care must be taken when they are in-use.

3.3.3. IR, MIR and NIR

Several studies have been conducted to identify chemical/biological compositions, monitor reaction progress and study hydrogen bonding using Infrared (IR) spectra measurements based on three different spectra, i.e., the near-infrared (NIR), mid-infrared (MIR) and far-infrared (FIR). The IR spectroscopy is a noninvasive technique, known as a 'green' analytical method due to its reagent-free approach [333]. Instead of analysing measurements based on a singular mean or maximum value, compact data sets provided by employing vibrational spectroscopy can be easily interpreted, resulting in a continuous detection of contaminants and increasing the chance to overlook high contamination [334].

The commercial MIR spectrometer has been expanding for the use in environmental monitoring assessments due to its dense spectral information and high intensity of its spectral peaks [335]. Previously, MIR spectrometer was reported for oil and grease determinations using tetrachloroethylene extraction [335]. A 100 mg/mL oil and grease solution, that contained octanoic acid and isoctane, was used as the target sample. Absorbance measurements were analysed using a PerkinElmer® Spectrum™ 400 FT-IR spectrometer in MIR mode.

A comparison between NIR and MIR reflectance spectroscopy for measuring soil fertility parameters has been reported by Reeves et al. [336] using IR spectra method. The combination of these techniques with chemo-metric analysis led to good calibration for the detection of organic carbon, total nitrogen (TN) and soil texture. However, it appeared that MIR tended to provide better calibration than that of the NIR region. These precise new approaches for the acquisition of soil data correlate to various degrees of MIR abilities. Despite the high cost in comparison with NIR and UV sensors, the advantages of using MIR sensors outweigh its disadvantages to end users.

A simultaneous quantification analysis of xylene, benzene and toluene isomers in water has been successfully performed with detection limits reported to be 20 ng/L, 45 ng/L and 80 ng/L, respectively [337]. This study developed an evanescent field spectroscopy that correlates with ATR crystal or MIR transparent optical fibres that serves as an optical transducer. Approximately 18 min of operational time response was needed to determine the solute concentrations in water samples. In contrast to the MIR measurements, the *in-situ* monitoring of water parameter composition by NIR spectroscopy is increasingly favourable among scientists worldwide. This approach includes the detection of nucleation and polymorphic transformations using different types of sensor arrays [338]. A novel approach to chemo-metric analysis was introduced by applying a composite sensor array (CSA) to obtain better information and to detect various crystallization mechanisms. A comparison between CSA-based techniques using Raman, NIR, FBRM, PVM and thermocouple probes was made to determine the optimal robust detection of matter. Having a clear view of nucleation and polymorphic transformation was found to be very informative in relation to Raman and NIR spectra relative to other probe. However, the ability to detect nucleation, crystal growth and polymorphic transformation by NIR was limited due to presence of water in the system [338].

A novel and simplified optoelectronic system was designed on the basis of an NIR technique at wavelength acquisitions of 630, 690, 750 and 850 nm using a LED as a light source for evaluating fruits and vegetables [339]. Validation was performed on dye solutions resulting in the system's ability to discriminate among the reflectance rate's low limit levels, which were in the range of 2–4%. The tendency to reduce the cost and size of instrument analysis has led to the development of an LED device as a source of

narrow bands that are able to excite NIR radiation. As NIR can be used to measure translucent packaging material, there have been many deployments of NIR instruments in raw material quality control such as development of non-invasive detection of hydrogen peroxide and its concentration in a drinking bottle [340].

3.3.4. Raman in comparison to surface-enhanced Raman spectroscopy

The principle of Raman spectroscopy correlates the excitation of atoms or molecules to a higher energy state using monochromatic light radiation. When an atom at a higher energy level returns to the ground state, energy is dispersed by Rayleigh scattering, which results in the frequency shifting of atoms known as the Raman effect. The basic Raman experimental setup consists of an angle configuration (90° and 180°), a wavelength selector, a filter, a mirror and an excitation source.

In developing advanced technology for analyte detection, an approach using Raman spectroscopy in microfluidics (MRS) was reported in the work of Ashok and Dholakia [295]. The Raman spectroscopy detection was assembled using an on-chip fibre with a polydimethylsiloxane (PDMS)-based microfluidic platform. The setup of a PDSM-based chip via soft lithography as presented in Fig. 16 involved an excitation probe, fluidic channels, a collection probe and an inlet and outlet. Device validation was conducted using a urea solution with a 200 mW laser at a 785 nm excitation wavelength with an acquisition time of 5 s. The minimum detection limit is 140 mM.

An investigation of dissolved sulphate ions (SO_4^{2-}) and methane (CH_4) in pure water was performed using Raman spectroscopy based on two new detection approaches, namely the liquid core optical fibre (LCOF) for SO_4^{2-} and an enrichment process for CH_4 [341]. Both methods employed Raman instrumental measurements, which primarily consisted of 0.3 W laser power, a detachable dichroic mirror and a Raman optical fibre probe. An LCOF-based Raman signal for SO_4^{2-} was captured, and the analysis showed that the intensity was 10 times greater than that of conventional Raman setup. The extraction of CCl_4 for the detection of CH_4 indicated the location of the Raman peak at 2907 cm^{-1} with a methane concentration of less than 1.14 mmol/L.

The use of Raman spectroscopy for in-line water quality monitoring is also found in large-scale deployments due to its superiorities with respect to ease of use, portability, compactness and sensitivity to water environment [342]. Application areas such as environmental analysis of organic and inorganic samples [343], tissue imaging [344] and liquid sample evaluations [345] have employed the Raman scattering spectroscopy technique to a tremendous extent. The non-destructive Raman approach has a fast operation time which enables detection of pesticides not only in the water samples but also food products. Raman spectroscopy is well recognized as a powerful method for the on-site evaluation and determination of chemical-biological molecular compositions [346]. However, one of the major challenges in bio-detection is the ability to instantly diagnose variety of low concentrated toxin contaminants. Hence, a cell-based Raman spectroscopy biosensor was introduced by Iloan [347] to differentiate the biochemical changes that occurred in cells with a large range of toxic agents. The target samples were nucleic acids, proteins, lipids and carbohydrates. An example of Raman spectra results is shown in Fig. 17(a) in which different target samples were found in a living cell. More importantly, the cell-based biosensor Raman spectroscopy was able to quantify and qualify between live and dead cells as shown in Fig. 17(b).

A lower limit of detection is achievable by using the SERS technique in comparison with the standard level of LOD contaminants in water as set by the EPA [342]. Because of the high-intensity signal used in SERS, this technique is among the most useful tools for environmental science, electrochemistry and

analytical chemistry/biology applications. The application of SERS technique is to discriminate antibacterial properties such as tranquilizer (phenothiazine), diclofenac sodium and diclofenac sodium β -cyclodextrin complex and non-natural β -amino acids [348]. In comparison with the Raman spectra signal analysis, the SERS spectra provided tremendous measurement information about the molecular structures and absorbance behaviours. A similar approach used for bacterial classification and discrimination was also reported in the work of Wu et al. [349] in which vancomycin-functionalized silver nanorod array (VAN AgNR) was considered. This analysis was conducted through the isolation of 27 different bacteria from 12 species. Measurements were obtained from an NIR diode laser excitation source at 785 nm, and underwent chemometric analysis using a combination of PCA and HCA approaches. The results showed that the use of VAN AgNR substrates tended to generate more SERS spectra, making the bacteria differentiation more accurately.

Although vibrational spectrometer measurement seems to be a promising technique for water monitoring technology, poor applicability to continuous on-line monitoring because of high water interferences must be addressed [350]. It has a tendency to operate efficiently because of its low light absorbance in water, but spectrum analysis presents a hurdle owing to the overtones and overlapping absorption bands [351]. For these reasons, the spectral bands become weaker.

3.4. Biosensors

Biosensors have been increasingly utilised in recent years in the field of pathogen detections. This kind of sensors has the ability to measure molecular signals by applying specific bio-recognition elements, such as enzymes, whole cells, antibodies and nucleic acids, which are integrated with electrical interfaces via transducer platforms to obtain measurable signals. Because of the high sensitivity and no requirement of sample pre-concentration step, biosensors offer a faster operational time in comparison to other conventional techniques [352]. An environmental pollution can be caused by both human activities and industrial discharge. A biology approach was reported to be useful in detecting heavy metal ions and bacterial compositions in water source [323,353]. Examples of genes that act as bio-receptors and have been previously determined by whole-cell biosensors are genetically modified mutant *Pseudomonas* sp. *Dmpr*, *Pgp* protein-based resistance genes and *mer* resistance genes [354–357].

The efficiency of whole-cell biosensors to monitor the presence of water contaminants is dependent on bio-receptors, transducers and immobilization techniques [358]. For instance, study demonstrated by Kuncova et al. [359] using *Pseudomonas putida* strain TVA8 bio reporter was able to detect benzene, toluene, ethyl benzene and xylene in water samples with concentration in the range of 0.5–120 mg/L. Kubisch et al. [360] evaluated the robustness of different cell lines by detecting cytotoxic substances in wastewater using whole-cell biosensors via eukaryotic cell lines. The detection of selected target samples (i.e., NiCl_2 , CuSO_4 , nicotine and acetaminophen) was carried out on the basis of the cellular effects of each substance, which involved the pH, O_2 and the impedance of water. HT-29, canine hepatocytes, HepG2, L6 and NHDF cell lines were used to test the acidification rate of target samples, whereas V79 cells were used to obtain the respiration rate of the target samples. These cell-lines were assembled onto six different channels of a silicon surface biosensor chip. Nevertheless, the study was only aimed to identify the presence of contaminants. Quantifying single substances was not performed.

Nanosilver is known to be the most commonly used engineered nanomaterial for water treatments, thus it is likely that Ag^+ ions will be released to the environments [361]. Previous works have

reported that such nanomaterial can be detected using label-free sensitivity biosensor approach [362,363]. In order to meet the need for reliable and sensitive methodology for the detection of nanoparticles in aqueous samples, a low-cost and portable detection assay was established to determine the reactivity and characterization of selected nanoparticles (Ag , Au , CeO_2 , SiO_2 and VO_2) with particle size ranging between 5 and 400 nm [364]. As a leading approach for detection and exploration of nanoparticles, whole-cell biosensors was developed by integrating *golTSB* genes from *Salmonella enterica* serovar *typhimurium* to induce Au (I/III) complexes as illustrated in Fig. 18 [365]. The quantification of gold nanoparticle complexes with concentration as low as 0.1 μM was able to be identified. The fabricated biosensor was also used to identify other metal ions, including Ag (I), Cu (II), Fe (III), Ni (II), Co (II), Zn and Pb (II). This contradicted a previous study where the response of *golB* genes only increased in response to Au (III)-complexes but not other metal ions [366].

Another genetically engineered gene system found in the literature is yeast/mammalian cell line which was used to study galactosidase and luciferase activity in water sources [367]. However, it must be noted that engineered microbial biosensors do not provide complete quantification, but rather on semi-quantitative analysis [368]. Bioassays based on sulphur-oxidizing bacteria (SOB) reactor have also been used for the toxicity identification of Cr (III) and Cr (IV) in water samples [369]. The results indicated that significant increase in the slope of electrical conductivity could be obtained when SOB DNA was exposed to Cr (III) which may be caused by increment of salt concentration and exposures of unstable reactor conditions. On the other hand, a trace amount of atrazine in ground water supply was able to be identified using an integration of printed circuit board chip nanoporous alumina membrane label-free bioassays with electrochemical impedance spectroscopy [370].

Bioassays adjacent to sophisticated biotechnology instrumentations demonstrate a fast response with a relatively simple method for identification of various water contaminants [371]. An inline water analyzer adjacent to whole-cell biosensor was established to carry out surveillance of water network using reporter gene of bacterial luciferase *lux* operon (*luxCDABE*) driven by *E. coli* promoter P_{lpod} [372]. In accordance with non-biological contaminants, a portable gold screen printed electrodes amperometric biosensor was developed by Salvador et al. [373] for the detection of Irgarol 1051 in water samples. The immunoassays reagents (As87- and 4e-BSA-based) used in this study were also found elsewhere [374,375]. Antibody peroxide (AntiIgG-HRP) was used for the binding reaction between target analytes and 4e-BSA competitor. In the presence of analytes, stable signals were able to achieve within 10 s upon initial acquisition. Separately, Belkhamssa et al. [376] designed a biosensor and used it to detect alkylphenol in water environment. The analytes detection was observed through an immunoreaction of 4-nonylphenol and the accuracy of developed biosensor was validated with enzyme-linked immunosorbent assay (ELISA). The outcomes are very promising with reproducibility of $0.56 \pm 0.08\%$, repeatability of $0.5 \pm 0.2\%$ and LOD for nonylphenol as low as 5 $\mu\text{g/L}$.

Cytotoxic substances present in tap water could also be detected using bioluminescent *E. coli* bio-reporter strain TV1061 via integration of specific heat-shock *grpe* promoter with *luxCDABE* reporter operon [377]. The bioassays and microbial biosensors employed for the toxicity assessment involved *Chlorella* sp., *Chlorella vulgaris*, *Monoraphidium* sp., *Scenedesmus subspicatus* and *Brachionus calyciflorus* sp. [378]. Due to presence of countless toxic cyanobacteria in water, Weller [379] made an attempt to study their existence using biosensors. Cyanobacteria produces algal toxins in fresh water which are hazardous to aquatic ecosystem and human health. In order to reduce the risk of a possible breakdown of toxic cyanobacterial in drinking water, a multi-barrier approach,

Table 3

Parameters of online water quality monitoring.

Category	Water Quality Parameter
Physical	Turbidity, color, conductivity, hardness, temperature
Inorganic	pH, DO level, disinfectants, metals, fluoride, nutrients
Organic	TOC, hydrocarbon, VOCs, pesticides, DBP
Biological	Algae, protozoa, pathogens, BOD
Hydraulics	Flow, pressure

comprising prevention, source control, detection optimization and monitoring was recommended [380].

On the other hand, a corresponding approach using an ammonia-oxidizing bacterium (AOB)-based *nitrosomonas europaea* biosensor has been designed by Zhang et al. [381] to determine *allylthiourea* and *thioacetamide* concentrations in water by measuring the ammonium oxidation rates. The results showed 0.17 µM and 0.46 µM for *allylthiourea* and *thioacetamide*, respectively. Another enzymatic-based (2-phospho-L-ascorbic acid trisodium salt) biosensor made of screen-printed carbon electrodes with modified gold nanoparticles was used to detect the tungsten ions present in tap water, purified laboratory water and bottled drinking water [382]. More information about the use of membrane-based biosensors for pathogen could be found elsewhere [383–386].

Since *E. coli* is the frequently found contaminant in drinking water, a rapid and sensitive assays for bacterial identification is required. Rapid detection of *E. coli* was developed by Hassan et al. [387] using 4-methylumbelliferyl-β-D-glucuronide (MUG) substrates. The quantitative results was obtained due to the yielding of a fluorogenic 4-methylumbelliferone (4-MU) product via substrates hydrolization. Bacterial such as *Klebsiella*, *Salmonella*, *Enterobacter* and *Bacillus*, which were used for validating the MUG substrate specificity could result in significant fluorescence signals.

3.5. Wireless sensor network and remote sensing applications

Online monitoring is usually defined as a real-time measurements for sampling and analysis, providing larger data frequency in comparison to the conventional sample-based method. Online monitoring is more flexible and can be conducted in remote locations with faster response. The design of an online monitoring instrumentation strongly depends on the desired identification of water parameters. Table 3 summarizes the online water quality monitoring parameters for each category.

Constructing an online monitoring detection system using wireless sensor network (WSN) could offer several advantages such as simultaneous data measurements, higher detection accuracy and sensitivity, sufficient data sets and easy monitoring assessments. Furthermore, WSN which requires low power consumption results in lower operating cost [388]. There have been several applications of WSN in water monitoring [389,390]. For instance, a WSN-based online monitoring system that consisted of data monitoring nodes, base station and monitoring centre was developed for the water quality assessment on the artificial lake at Hangzhou Dianzi University, China [391]. With this monitoring system, water quality parameters such as pH, dissolved oxygen, EC and temperature could be easily transmitted to a remote monitoring centre for further analyses via GPRS network. The measurement was automatically carried out every h generating sufficient data for monitoring purpose. On the other hand, Wu et al. [392] designed a self-powered mobile sensor for real-time contaminant detection in water distribution pipelines aiming to detect pH level, water hardness (Ca^{2+} , Mg^{2+} and HCO_3^{-}) and disinfectant-related ions (NH_4^+ and Cl^-). The mobile sensor operated within a 2.76 inch diameter of spherical-shaped shell consisting of potentiometric

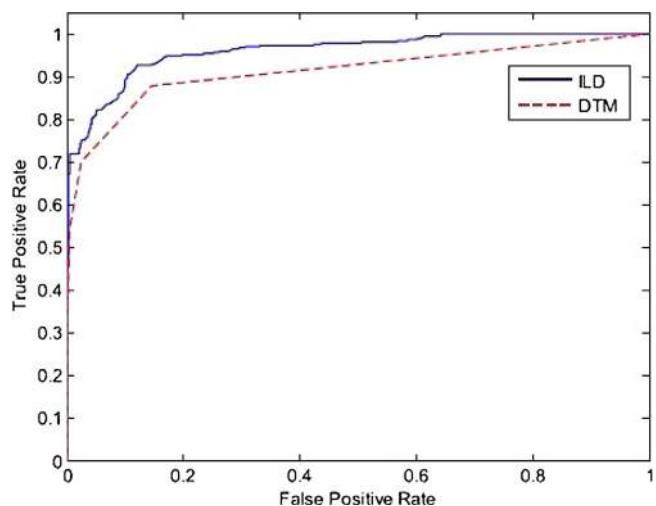


Fig. 19. ROC curve comparing the performance of the two methods (ILD and DTM) on low type events (Housh and Ostfeld [406]).

electrochemical-based multi-analyte biochip, microfluidics, electronics controller and energy harvesting system for power supply.

Furthermore, a low cost, miniaturized and sensitive micro-electronic wireless nitrate sensor network was established for quantification of nitrate concentration in water environments [393]. The conceptual design of sensor network consisted of sensor interface (input and output parameter interface), a low-power processor and wireless communication named 'Imote2'. To obtain wireless communication, electrochemical potentiostat was needed to be miniaturized and portable. The results showed that the microsensor was able to detect nitrate concentration in water samples with LOD between 25 and 83 ng/L. However, the implementation of such sensor network on field is still at the early stage of development.

Nitrate concentration in water samples was also identified using a similar approach based on a dielectric impedance sensor (DIS) node on ZigBee mesh communication [309]. The system was constructed to perform a continuous detection within a frequency between 5 and 100 kHz under 250 mW. According to the author, this was the first development of wireless platform via AD5933 touchscreen device and chemical sensor. The detection of water-borne disease-causing bacteria in water sources were carried out by Kim and Myung [394] using an enzyme substrate assay method. The colorimetric properties were monitored via Wi-Fi connection through a web-based user interface.

The use of WSN often involves multiple sensors to improve system stability and fault tolerance [395]. However, managing continuous long-range communication networks is a challenging task due to constant requirements in power supply. Energy harvesting system has been recommended to manage wireless-based sensor power supply, however, most energy harvesting systems rely on solar cells [396]. Several self-powering mobile sensors were found to be used in water distribution pipelines [397,398]. They are operated via rotational miniaturized motor, hydraulic energy and thermal energy in water-air-temperature gradient and kinetic energy in water pressure.

4. Algorithmic model-based event detection

Generally, there are two methods used to conduct detection algorithm. Initially, the model-based event detection method involves a signal-to-noise principles using laboratory and sensor test-loop evaluation. Indication of contamination events is derived from the chemical changes in background water quality signals

Table 4

Comparison between water contamination detection methods.

Methods	Advantages	Disadvantages	References
Discontinuous (sample-based) Analysis	- Accurate contaminants detection - Better quantitative data measurements	- Time-consuming (48–96 h) - Lack of sensitivity for low concentration - Limited detection of contaminants	[88,89], [137–143], [163], [167], [217], [244,245], [429,430]
Sensor Placements Approach	- High sensitivity event detection - Multiple water quality parameter measurements	- Relatively high cost - Small data sets - Complex design analysis and optimization - High inaccuracy rate - Transmission time delay - High maintenance	[249,250], [258–265]
Microfluidics Sensors	- Label-free detection method - Portable and affordable - Minimum sample volume - Fast operating responses - High detection sensitivity	- Requirement of specialized setup - Requirement of sample pre-treatments - Manual sample collection	[279–281], [292–301]
EIS and DIS Spectroscopy	- Label-free detection tool - Simplicity and cost-effective	- Inadequate detection sensitivity - Non-continuous detection - Requirement of expertise guidance - Sensitive to illuminations	[303–308]
Light Emission / Luminescence	- Easy detection of BOD/DOM - Simple separation steps - Compact, non-destructive	- Expensive for large-scale deployments - Sample pre-processing required - Limited water quality parameter detections - Sensitive to surroundings temperature	[313–317], [325–328], [332], [431,432]
IR, MIR, NIR	- Simplicity and cost-effective - Rapid detection speeds - Portable and in-situ measurements	- High water interference - Existence of overtone and overlapping bands - Poor signal intensity	[333–337], [339], [433]
Raman and SERS	- Multiple detections - No sample preparation - Strong light absorption in water - Reagent- and waste-free - Portable and compact - High detection sensitivity	- Poor signal intensity - Complicated installation - Lack of reproducibility substrates - Highly expensive - Existence of overtone and overlapping absorption band	[294–300], [341,351], [433]
Biosensors	- High sensitivity to biological contaminants - Suitable for in-situ monitoring - Minimum sample preparations - Portable and miniaturization - Fast response time	- Laws limitation of modified organism genetically - Sensitive to environments - Lack of system stability - Limited transducer life expand - Risk of bio-receptor leakages	[372–377], [380–388], [391], [400–407], [434–438]
Event Detection Model-based	- High true positive alarm rate - Low false alarm detections - Fast response time	- Complicated calibration process - Highly dependable on predictions and estimations - Computationally intensive	[338], [353], [410,411], [417,418]

which are responsive to integration of event detection technique [399,400]. However, it must be pointed out that the variation of the background water quality in the experimented systems would not be exactly the same as the variation of actual WDS [401]. Meanwhile, the second method used for event detection is based on signal processing and data-driven technique. Many studies have focused on the development of data-driven estimation model detection algorithm such as statistical, pattern-based recognition, machine learning approach, and image processing to detect contaminants based on real-time water quality measurements [402–405]. Contamination event detection in WDS has become a challenging research topic, in accordance with improved water system analysis.

At present, a wide variety of event detection approaches, including statistical, machine learning and optimization methods have been used. However, challenges in utilizing this methodology are the merging of single alarms that could be triggered by each quality indicator and the false detection alarms [406]. Because of this reason, an event detection model-based approach known as integrated logit detection (ILD) was proposed, which is an extended statis-

tically based fusing process of dynamic threshold method (DTM) [406]. These two event detection models generate algorithmic evaluations to explore the most effective training phase performance for identifying contaminants using Receiver Operating Characteristic (ROC) curve, which represents the trade-off between false and true positive for probability threshold as shown in Fig. 19. The ROC curve demonstrated that the higher true positive rate of ILD than that of rate of DTM. The observation resulted in high probability threshold of 0.9 and low probability threshold of 0.5 for ILD and DTM, respectively.

A similar methodology was used to study the effectiveness of two different event detection models of multivariate classification techniques. Commonly used sequence analysis of classifying events are an un-supervised minimum volume ellipsoid (MVE) and a supervised support vector machine (SVM) [53]. In terms of formulation and framework unity, the MVE model reduces the complexity of the algorithmic analysis in predicting contamination events relative to the SVM model. In addition, the Gaussian distribution data that were used for the MVE model approach could contribute high accuracy of 17% in separating modular boundaries.

On the other hand, anomaly-based water contamination detection methods that include Artificial Intelligence (AI), have converged over the last decade. The classification of water quality measurements into anomalous categories often employs an artificial neural network (ANN) and a support vector machine (SVM) [407–409].

In order to improve accuracy in data event modelling, combined method that included integration of data analysis from all sensors, hydraulic model networks and single spatial warning systems was introduced [44]. Several studies have made targeted improvements in event detection decision-making by extending a single-sensor event detection model to a spatial multiple sensor with an on-line approach. It has been previously reported that the conventional water quality sensors integrated with a real-time method based on the Mahalanobis distance approach was highly dependent on feature vector of each contaminant [55].

Another type of model-based event detection approach is by using Monte-Carlo simulation. Such approach has been used to detect chlorine at various sensing locations along water distribution system [410,411]. The event detection model has a wide variety of optional algorithms based on the quality parameter analysis and their accuracies in determining different contaminants. Execution on the basis of multi-sensor fusion can also be achieved by deploying an extended Dempster-Shafer method [56]. Research on the prediction of future water quality parameters in the absence of automated on-line water quality sensors using an autoregressive model was also investigated to compare the performance of various event detection models. Several studies on chlorine concentration measurements in water have been reported using a Radial-Basis Function network [56,411]. Nonetheless, there are contradictory views on its efficiencies.

A web-based tool LOAD ESTimator (LOADEST) reported in the work of Park et al. [412,413] was developed to estimate the pollutant load by integrating stream-flow watershed data measurements and water quality data as the model inputs via server web access. A model-based event detection using a fuzzy comprehensive genetic algorithm was introduced by Wen et al. [57] to measure the toxicity of seawater samples with high levels of spatial variation, oil contamination, silicate and heavy metals (Zn and Pb). In short, assumptions of ideal and realistic sensor placements are essential for high accuracy in event detections [414]. The genetic algorithm has become the preferable method used by many researchers for water system design optimization techniques [415,416]. Although there are challenges associated with a wide variety of optimization problems, each water quality factor can be weighted carefully using additional logic simulation methods. Efforts to deploy the contamination event detection and surrogate approach has been made as alternative ways to overcome drawbacks regarding conventional laboratory-based analysis and the SPA method. A variety of techniques for water quality event detection have been well-developed, including the statistical, heuristic, machine learning and optimization methods used to analyze contaminant changes and the possibility of contamination [417]. Unlike other available methods, developing a model-based detection scheme involves intensive computational algorithm [406]. In addition, the requirements for calibrations and fabrications would further increase the complexity of the overall system [411]. Predictions and assumptions are the primary variables when utilizing an event detection model-based approach. The contamination evaluation process is complex due to high variability in environmental conditions [418].

5. Future recommendations

Current global challenges caused by climate changes, urbanization and industrialization have prompted the need of safe, clean and

readily treatable water resources. The production of high-quality water is becoming more challenging because of alignments in the detection limit concentration that correlates with the WHO and EPA water quality parameter standards. Owing to the fact that one in nine people around the world does not have access to clean water supplies, innovative water contamination detection technologies must be able to (1) achieve a fast response early warning detection, (2) improve water treatment efficiency, (3) minimize risk of harmful contaminant exposure, (4) quantify and identify the types of contaminants and (5) continuously detect unwanted contaminants simultaneously.

A comparison on the pros and cons of the state-of-the-art water monitoring technologies is summarized in Table 4. The overall monitoring and detection system must acquire accurate data to minimize statistical methods, increase spatial hybridity (with a combination of quantitative and qualitative measurements), reduce operation time to evaluate the presence of contaminants and reduce the project cost. However, it is rather difficult to continuously monitor real-time water contaminants, especially when they reach the point of end-user.

Although the achievements that have been made in the conventional analytical techniques are remarkable, the use of agents as receptors and transducers to capture contaminants could negatively affect raw data measurements to a certain extent [419]. More research efforts is still needed to develop efficient yet cost effective water quality monitoring systems. In general, the analyses done by the conventional instruments are not only labour intensive [420] but also relatively expensive [421]. These instruments in most of the cases are only capable to yield small data sets [422,423]. Major challenges that limit commercialization are instrumental complexity and large data mining capabilities.

Even though WSN and remote sensing technologies have been adopted in current water monitoring systems, many of them are lack of hybrid analysis and are not user friendly for continuous detection/monitoring [424–426]. Those wireless sensing detection devices are mainly focused on the deployments in water network distribution rather than the point of water consumption, which is end-user water supply. Hence, the opportunities to develop water monitoring tool kits with a graphical user-interface (GUI) that is user-friendly, easy to operate and re-usable are on demand. As micro-scale device is more sensitive to micro-organisms than macro-scale device, it is more ideal to overcome these challenges. Nano-scale sensing device meanwhile has received a great deal of attention in recent years owing to its extremely low detection limits for contaminant concentrations [427,428].

Furthermore, it is highly recommended to incorporate two or more sensing devices, such as hybrid of microfluidic-based or biosensor-based platform with a spectroscopic detection system to enhance detection sensitivity and accuracy. This approach combines potential *in-situ* water monitoring technologies, such as NIR-Raman spectroscopy and NIR-FTIR-SERS techniques, which are particularly suitable for water analysis because of its simplicity, high speed detection response, strong light absorption in water and very low LOD [224]. It is quite certain that this innovative approach could play an important role in meeting the ever-increasing demand for water quality assessment.

In large deployments of an on-line monitoring system, it is crucial to minimize the complexity of the overall system to prevent data transmission interruptions, which might result in data losses. In view of this, highly reliable hand-held devices and novel user-friendly toolkits are crucial during water monitoring process. In assessing water quality using contaminant detection technologies, potential contaminants in potable water should also be considered by conducting intensive risk management, risk assessments

and risk research to minimize the hazardous contaminants that are present in tap and drinking water.

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