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Cover Page Footnote

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A REVIEW ON VALIDATION OF ENZYME LINKED IMMUNOSORBENT ASSAY (ELISA) TECHNIQUES FOR DETECTION AND QUANTIFICATION OF DIFFERENT CONTAMINANT IN AQUATIC ENVIRONMENT

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ABSTRACT

In world, contaminants and microbes are considered as serious problem in house water supplies as well as in aquatic environment for life. As a result, different countries are working to set level of these contaminants and microbes in domestic supply of water and in aquatic environment. In addition to these scientists are also working on reliable sampling and then their suitable analysis method for accuracy. It is also very important to improve and give justification for analytical techniques used in laboratories, which enable us to monitor level of various contaminants in aquatic environment. In this review, we have focused on quantitative value of limit detection and satisfactory accuracy level for different water contaminants detected by Enzyme Linked Immunosorbent Assay (ELISA). The correlation (r) value between detection limit and various ELISA techniques is 0.16 and their regression (R^2) value is 0.027. While, r value for antibody and contaminants in relation to limit detection is 0.081 and -0.046 and their R^2 value is 0.15 and 0.015. Similarly, analysis was also performed to check relationship of satisfactory accuracy level with techniques, antibodies and contaminants and their values for r is -0.032, -0.032 and 0.064. Value of R for satisfactory accuracy level in relation to techniques, antibodies and contaminants is 0.72, 0.072 and 0.150. Obtained results allow us to check and to set the level of contaminants in house supply water samples. We should work to set standard level of these contaminants for drinking and house supplier water and then published according to WHO requirements for healthy life.

Keywords: Contaminants, microbes, immunesorbent assay, aquatic environment.

INTRODUCTION

Different contaminants relate to water are creating severe health problem in exposure to human. The severe health problems include chronic toxicity, mutagenic and carcinogenic effects. Some toxic chemicals create toxicity at very low level so its need to introduce accurate and reliable analytical techniques for determination. In past literature different techniques are discussed for detection and quantification of contaminants. In most of studies ELISA techniques are highlighted as technique for contaminants detection. Enzyme linked immunosorbent assays (ELISA) is bio-technique which is used to detect antigens and antibodies (Zeng et al., 2019). ELISA Kit is made up of plastic called polystyrene. In laboratory we should have reagents which shows specific performance antibody against specific antigen (Sherwood & Hayhurst, 2013) According to ISO, 5725 definitions of accuracy comprise the precision and trueness. On the base of accuracy profile and acceptability limit concept accuracy can be assessed in global way (Masson et al., 2018).

Various chemicals and different technique impact significantly as well as non-significantly on the limit of detection and accuracy of ELISA. Bisphenol A (BPA) is an organic compound belongs to group diphenyl methane derivatives and bisphenol. One of the studies in China has shown that discharge of domestic sewage and industrial waste water are source of BPA. More attention is needed to know the risk of BPA in environment (Zou et al., 2014) Increase in use of BPA in plastic industry has raised the level of BPA from ngL-1 to ug-L. Contaminants relates to BPA are termed as endocrine disruptors due to their negative impacts on endocrine system of different organisms. To remove high level of BPA in river, drinking and waste water, oxidation of BPA by use of ozone has been tested and proved effective to degrade BPA. But, as a result significance lack of information has noticed on synthesis of byproduct and their toxicity (Umar et al., 2013).

Sulfamethazine contaminants are increasing in drinking water, surface water, ground water and waste water due to excretion of antibiotics from body via feces and urine without transformation. The concentration of sulfamethazine in freshwater ranges from 0.1 μgL^{-1} to 12 μgL^{-1}, and for waste water ranges from 10 μgL^{-1} to 231 μgL^{-1}. It was noted that *S obliquus* has less negative impacts at ≤ concentration of 0.5mgL-1 while as concentration increases from 1 to 6 mgL^{-1} then these are badly and severely impacted. Still there is need to investigate the gene and enzymes responsible for metabolism in aquatic microorganisms during biodegradation of emerging contaminants (Xiong et al., 2019). Malchite green (MG) and crystal violet can be analyzed in aquatic environment by using temperature controlled ionic liquid dispersive liquid micro extraction (TC-IL-DLLME). This technique can be applied in other water areas to determine contaminants cause by MG and crystal violet (Zhang et al., 2012). Level of chloram-phenicol (CAP) and ractopamine (RAC) can be determined by time resolve floroimmunoassay TRIFA. TRIFA method was first reported in 2011, for simultaneous identification medicinal residue related various chemical classes (Zhang et al., 2011). TRIFA has higher selectivity and sensitivity in comparison to ELISA, having low cost and same sensitivity with LC-MS/MS.TRIFA has no need to pretreatment of sample except filtration of water sample. TRIFA has high potential in screening of complete sulfonamide (SAs) by using antibody in samples of environmental (Zhang et al., 2010).

Presence of ractopamine hydrochloride in water has significant concern due to ecological impact and toxicity to human. This ractopamine is used as drug to increase yield of meat in animals. This drug is excreted as waste material in animal's urine and then introduce into environment which impacts organisms like fish. Concentration from 0.124 μgL^{-1} to 30 was found in waste water released from shed while $1.3x10^{-5}$ μgL^{-1} to $5.4x10^{-4}$ μgL^{-1} watershed (Sachett et al., 2018). Analysis of serological assay for different disease requires understanding of clinical indicator. No evidence was found that ELISA has lower or higher accuracy than immunoblast. Authentic estimate about specificity and sensitivity of test needs cross-sectional studies. Future researches have needed more targeted standardized value of test (Leeflang et al., 2016). Analytical quantity of results obtained from adding amount in the previous amount of sample is called recovery or accuracy. If results are close to 100 percent recovery then it is more authentic (Cox et al., 2014).

It is estimated that, globally 50 million kg of pentachlorophenol is produced annually. Residue in which pentachlorophenol has been identified are rain, air, snow, drinking water, surface water, ground water fish, blood and milk. Incorporated of these chemical in body interfere the synthesis of high energy phosphate compound which are important for cellular respiration. Illness and deaths in domestic animals have also seen in exposure to pentachlorophenol (Petrlik & Bell, 2017). Some drugs cannot enter mammalian cell but nano particle can enter in mammalian cell at tumor location due to small size. Very low or very less amount or value of drug or pollutant detect by any specific method is limit of detection LOD. Any method which has minimum or low very of LOD and LOQ is consider as more

authentic method (Lange et al., 2012). Limit of detection is most important parameter in modern analytical techniques. Detail information's what effect the limit of detection can help scientists to analyze various bio-analytical technique properly. It helps to design more accurate strategies to get best analytical performance. In past studies, TBBPA was found in abiotic matrices, human and wildlife. TBBPA has ability to absorb and distributed rapidly in tissue and then excreted in feces as primary source of pollution. It has been detected in human milk. TBBPA was found to cause acute toxicity, neurotoxicity, immunotoxicity, endocrine disruptor activity, nephron activity and hepatotoxicity in animals (Weber et al., 2013). The important aspect in ELISA is result therefore experimental conditions are needed to standardize. To fulfill the quantitative result requirement, the best way is to take triplicate mean of standard data and then blank control sample value is compared with deducted value. Standard curve along with line of best fit also helps us to determine sample concentration. Graphical analysis by software and machine base solution helps us to understand about more sophisticated value. Almost in all countries different water contaminants are considered as serious problem in water house hold supply. So, all countries of world should work to set standard level of these water pollutants for domestic supply. It is very important to improve the analytical techniques to monitor the concentration of contaminants in water. From past papers we can get unlimited availability of data to check and compare the validity of different analytical methods for identification of chemicals. The objective of literature was to check the dependency of accuracy and limit detection on the chemical, (Semenova et al., 2012) technique and nature of antibody.

DATA COLLECTION

This study includes systematic review and meta-analysis to assess qualitative and quantitative impacts of contaminants in relation to house supply water and aquatic in environment. Literatures were included if they if they have investigated factors that affect LOD and satisfactory level of ELISA. We have searched relevant research articles in Google scholar and NCBI data base. All qualitative and quantitative properties are listed in below Table 1.

RESULTS AND DISCUSSION

Analysis of results has shown that different analytical techniques, different chemicals, and nature of antibody (monoclonal and polyclonal) has significant as well as non-significant impact in relation to LOD and satisfactory accuracy level relates to ELISA as shown in table 2. Different Techniques and chemical impact on LOD of ELISA significantly having p value of 0.002 and 0.05, while antibody impacts non-significantly on LOD of ELISA having p value 0.169. In this review it was detected that 2.7 % percent impact on limit detection is due to variations in technique and positive correlation among themselves. While 15 % impacts on limit of detection was due to nature of antibody and has also positive correlation. Value of correlation and regression of different parameters effect on LOD of ELISA have shown in table 2. At 0.85 μgL^{-1} of ractopamine treatment concentration exploration behavior of zebra fish has seen, while at 8 μgL^{-1}. Degradation in exploratory and locomatory activities of zebra fish was detected. At concentration of 0.2μgL^{-1}, inducement in lipid per oxidation occur which leads to increase total thiol contents in brain of zebra fish.

Different factors effects on limit of detection and sensors of bimolecular assay. Same techniques may have different value for different chemicals. Binding affinity between antigen and antibody has contributed in range value of limit detection. Antigen and antibody affinity has wide range of variations 10^5 to 10^{12} M^{-1}. It is necessary to improve the sensitivity of instrument to achieve better analysis of sensitivity. To improve the affinity of binding for

bimolecular, it is important to address the issue related to development of sensors (Poghossian & Schöning, 2014). Concentration of pentacholorophenol is also detected in past studies for sensitive aquatic species. The concentration which impacts badly on reproduction, growth and survival is 8μgL^{-1} to 80 μgL^{-1} for algae and higher plants, 3 μgL^{-1} to 100 μgL^{-1} for invertebrates and less than 1 to 68μgL^{-1} for fishes. For little laboratory mammals and for domestic livestock LD50 ranges from 27 to 300mg/kg (Petrlik & Bell, 2017).

Table 1: Qualitative and Quantitative Properties of Contaminants in Water

Chemicals	Technique	Detection Limit	Satisfactory Accuracy Level	References
TBBPA-MHEE	Competitive Chemiluminescent Imaging Immunoassay	1.85	109.80	(Zeng et al., 2020)
TBBPA-DHEE	Competitive Chemiluminescent Imaging Immunoassay	6.34	122.40	(Zeng et al., 2020)
Sulfamethazine	Floro immune assay	9.8	127.0	(Zhang et al., 2010)
TBBPA	Competitive electrochemical immunesensor	0.1	120.0	(Gyimah et al., 2021)
Bisphenol A	Indirect competitive ELISA	9.934	-	(Zheng et al., 2011)
Sulfadiazine	Floro immune assay	5.4	126.9	(Zhang et al., 2010)
E coli	Sandwich method of immunoassay	3.0	107.0	(Li et al., 2020)
DBP	Indirectly Competitive immunoassay	7.0	120.4	(Liang et al., 2017)
Sulfamethoxazole	Floro immune assay	6.1	126.5	(Zhang et al., 2010)
Ractopamine	Direct competitive immunoassay	0.1	118.5	(Shen et al., 2007)
Bisphenol A	Liquid Chromatography tandem mass spectromerty	9.934	-	(Wu et al., 2017)
Trace Pollutant	Competitive electrochemical immunesensor	0.0655	118.39	(Zhang et al., 2010)
Pentachlorophenol	Indirect competitive ELISA	10.0	110.9	(Sun et al., 2016)
Leucomalachite Green	Direct ELISA	0.1	76.0	(Singh et al., 2011)

Table 2: Impact of different parameters on Limit of Detection and Satisfactory Accuracy Level of ELISA by Meta-analysis

Limit of Detection							
Parameters	**Sum of Squares**	**df**	**Mean Square**	**F**	**Sig.**	**R^2**	**R**
Techniques	4420.237	10	442.024	33.197	.002	.027	.164
Antibodies	30.783	1	30.783	2.142	.169	.151	.081
Contaminants	4441.552	15	296.103	18.506	.05	.015	-.046
Satisfactory Accuracy							
Parameters	**Sum of Squares**	**df**	**Mean Square**	**F**	**Sig.**	**Sum of Squares**	**R2**	**R**
Techniques	2201.477	7	314.497	53.775	.004	2201.477	.072	-.032
Contaminants	2215.683	10	221.568	12.730	.215	2215.683	.150	.064
Antibody	1145.402	1	1145.402	7.430	.034	1145.402	.072	-.032

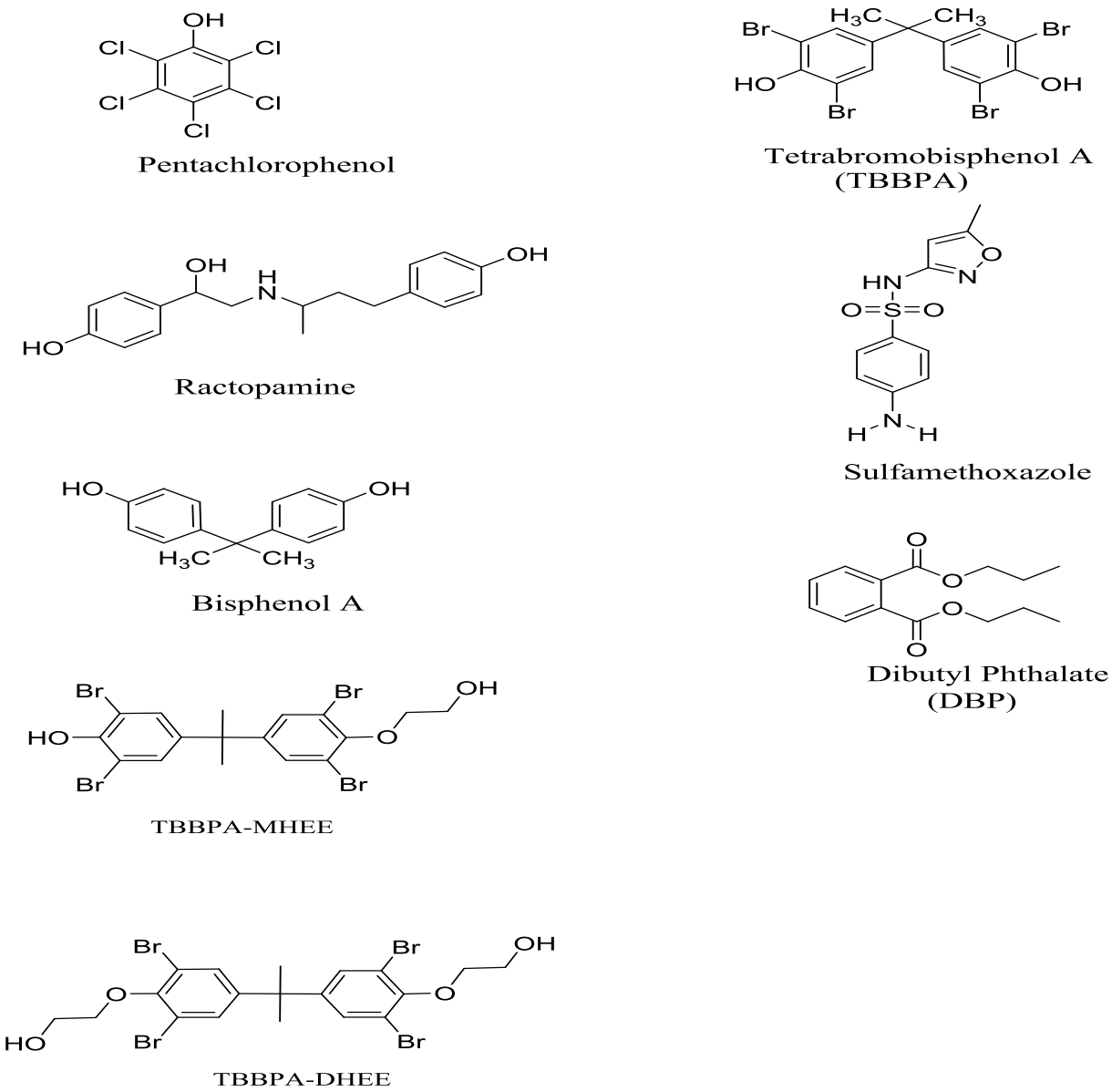

Figure 1: Structure of contaminants relates to water

Satisfactory accuracy level is also affected significantly as well as non-significantly by different technique, contaminants and antibody. P value for technique and antibody in relation to accuracy level is 0.004 and 0.034 which is significant, while p value for chemical 0.215 which non-significant. Regression and correlation value for different parameters impact on satisfactory accuracy level have shown in table 2. It was noted that when cell expose to high concentration of TBBPA then it leads to cause in serious injury in mitochondria and dilated smooth endoplasmic reticulum. TBBPA also induces reactive oxygen species and promote the formation of inflammatory factors like IL-8 and TNF alpha II-6 (Yu et al., 2019).

The important aspect in ELISA is results therefore experimental conditions are needed to standardize. To maintain the consistence help us in achieving accuracy. To fulfill the quantitative result requirement the best way is to take triplicate mean of standard data and then blank control sample value is compared with deducted value. Standard curve along with line of best fit also helps us to determine sample concentration. Graphical analysis by software and machine base solution helps us to understand about more sophisticated value

(Boyack & Klavans, 2010). In sample, interface of some other constituents may cause hindrance and antigen detection by antibody. Chelating ions, Proteins and metal ions in bio-sample may effect on antigen and antibody complex which results in false negative. To deal this such type of issue known protein concentration is spiked in sample matrix and standard diluents. Quantification for spike protein is carried out for sample and standard diluents and then compared. Difference in results may conclude that constituents of sample matrix have strong relation with detection process (Zeng et al., 2019).

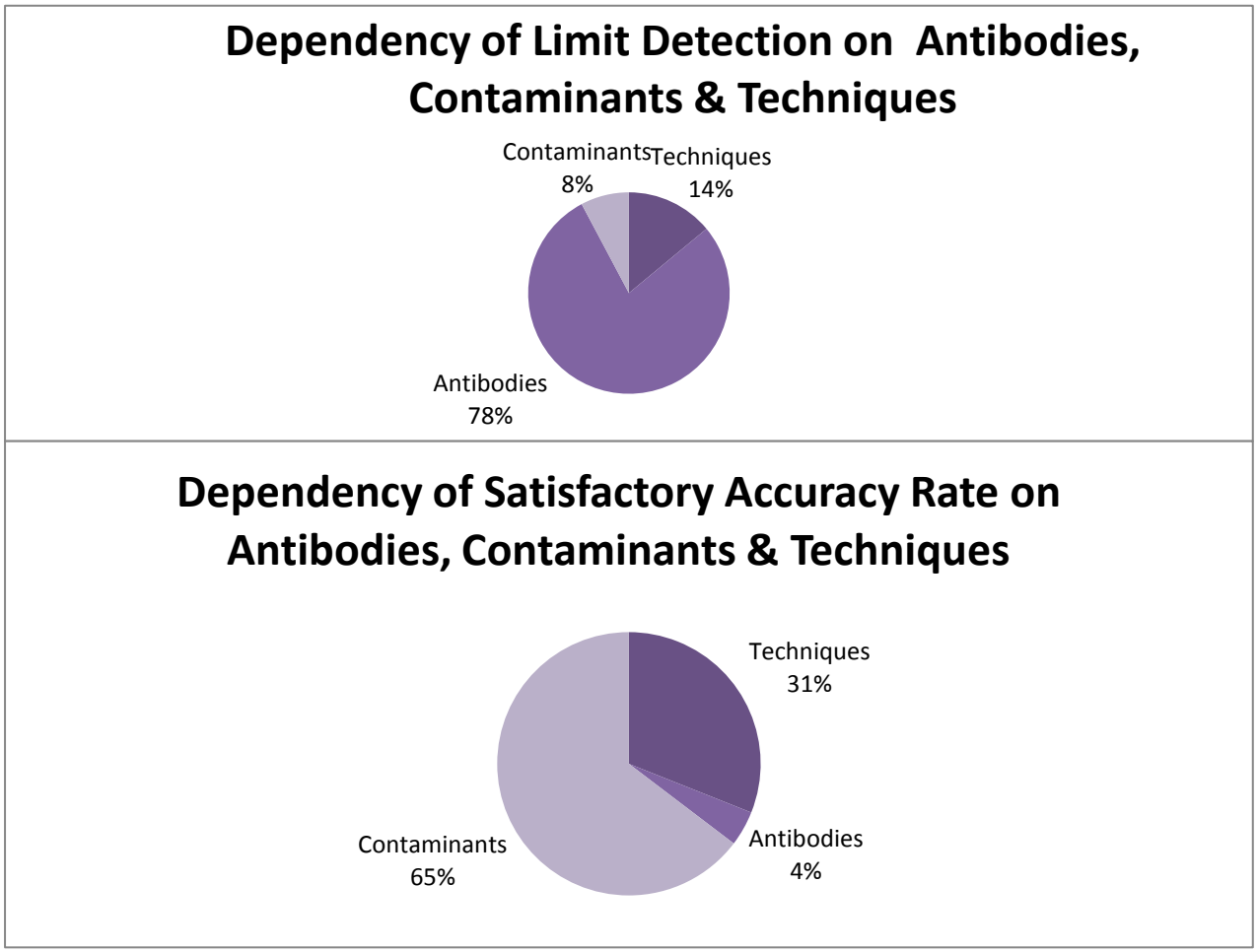

Figure 2: Graphical representation for dependency of limit detection and satisfactory accuracy level on antibodies, contaminants, and techniques

CONCLUSION

In conclusion, this review focused on the quantitative value of limit detection and satisfactory accuracy level for water contaminants using ELISA. The analysis revealed weak correlations between detection limit and ELISA techniques, as well as antibodies and contaminants in relation to limit detection. Additionally, no strong relationships were found between satisfactory accuracy level and techniques, antibodies, or contaminants. These findings emphasize the need for further research and the development of more reliable analytical techniques to effectively monitor water contaminants and establish appropriate standards for drinking and household water supplies in accordance with WHO guidelines.

AUTHOR CONTRIBUTION

All authors have equally contributed in this work.

CONFLICTS OF INTEREST

The authors declare that they have no conflict of interest.

REFERENCES

Boyack KW, Klavans R (2010). Co-citation analysis, bibliographic coupling, and direct citation: Which citation approach represents the research front most accurately? J. Ameri. Soci. Inform. Sci. Tech., 61(12): 2389-2404.

Cox J, Hein MY, Luber CA, Paron I, Nagaraj N, Mann M (2014). Accurate proteome-wide label-free quantification by delayed normalization and maximal peptide ratio extraction, termed MaxLFQ. Mole. Cell. Proteo., 13(9): 2513–2526.

Gyimah E, Xu H, Dong X, Qiu X, Zhang Z, Bu Y, Akoto O (2021). Developmental neurotoxicity of low concentrations of bisphenol A and S exposure in zebrafish. Chemosp., 262: 128045.

Lange FT, Scheurer M, Brauch, HJ (2012). Artificial sweeteners-a recently recognized class of emerging environmental contaminants: a review. Anal. Bioanaly. Chem., 403(9): 2503-2518.

Leeflang MMG, Ang, CW, Berkhout J, Bijlmer HA, Van Bortel W, Brandenburg AH, Van Burgel ND, Van Dam AP, Dessau RB, Fingerle V (2016). The diagnostic accuracy of serological tests for Lyme borreliosis in Europe: a systematic review and meta-analysis. BMC Infect. Dise., 16(1): 1-17.

Li X, Dong S, Arul P, Liu H, Liu L, Wang H, Zhang Q, Gyimah E, Yakubu S, Zhang Z (2020). A novel and facile immunosensor based on a barometer: Application for rapid analysis of Escherichia coli in waters. Talanta, 214: 120859.

Liang YR, Zhang ZM, Liu ZJ, Wang K, Wu XY, Zeng K, Meng H, Zhang Z (2017). A highly sensitive signal-amplified gold nanoparticle-based electrochemical immunosensor for dibutyl phthalate detection. Biosen. Bioelectr., 91: 199-202.

Masson P, Dalix T, Daugey G, Soulé P. (2018). Comparison of wavelength-dispersive X-ray fluorescence spectrometry and inductively coupled plasma optical emission spectrometry for the elementary determination in plants through the accuracy profile method. X-Ray Spect., 47(4): 287-293.

Petrlik J, Bell L (2017). Toxic ash poisons our food chain. International POPs Elimination Network (IPEN).

Poghossian A, Schöning MJ (2014). Label-free sensing of biomolecules with field-effect devices for clinical applications. Electroanal., 26(6): 1197–1213.

Sachett A, Bevilaqua F, Chitolina R, Garbinato C, Gasparetto H, Dal Magro J, Conterato GM, Siebel AM (2018). Ractopamine hydrochloride induces behavioral alterations and oxidative status imbalance in zebrafish. J. Toxicol. Environ. Heal., Part A, 81(7): 194-201.

Semenova VA, Schiffer J, Steward-Clark E, Soroka S, Schmidt DS, Brawner MM, Lyde F, Thompson R, Brown N, Foster L. (2012). Validation and long term performance characteristics of a quantitative enzyme linked immunosorbent assay (ELISA) for human anti-PA IgG. J. Immunol. Meth., 376(1-2): 97-107.

Shen J, Zhang Z, Yao Y, Shi W, Liu Y, Zhang S (2007). Time-resolved fluoroimmunoassay for ractopamine in swine tissue. Analy. Bioanaly. Chem., 387: 1561-1564.

Sherwood LJ, Hayhurst A (2013). Ebolavirus nucleoprotein C-termini potently attract single

domain antibodies enabling monoclonal affinity reagent sandwich assay (MARSA) formulation. PloS One, 8(4): e61232.

Singh G, Koerner T, Gelinas JM, Abbott M, Brady B, Huet AC, Charlier C, Delahaut P, Benrejeb Godefroy S (2011). Design and characterization of a direct ELISA for the detection and quantification of leucomalachite green. Food Addit. and Conta., 28(6): 731-739.

Sun C, Liu L, Song S, Kuang H, Xu C (2016). Development of a highly sensitive ELISA and immunochromatographic strip to detect pentachlorophenol. Food Agri. Immunol., 27(5): 689-699.

Umar M, Roddick F, Fan L, Aziz HA (2013). Application of ozone for the removal of bisphenol A from water and wastewater–a review. Chemosph., 90(8): 2197–2207.

Weber D, Milkovic L, Bennett SJ, Griffiths HR, Zarkovic N, Grune T (2013). Measurement of HNE-protein adducts in human plasma and serum by ELISA-Comparison of two primary antibodies. Red. Biol., 1(1): 226–233.

Wu X, Lu C, Dong X, Zhang Z, Yang M, Xu H (2017). Proteomics analysis of zebrafish brain following chronically exposed to bisphenol A. Toxicol. Environ. Chem., 99(3): 469-481.

Xiong JQ, Kim SJ, Kurade MB, Govindwar S, Abou-Shanab RAI, Kim JR, Roh HS, Khan MA, Jeon BH (2019). Combined effects of sulfamethazine and sulfamethoxazole on a freshwater microalga, Scenedesmus obliquus: toxicity, biodegradation, and metabolic fate. J. Hazard. Mater., 370: 138-146.

Yu Y, Yu Z, Chen H, Han Y, Xiang M, Chen X, Ma R, Wang Z (2019). Tetrabromobisphenol A: disposition, kinetics and toxicity in animals and humans. Environ. Poll., 253, 909-917.

Zeng K, Wei D, Zhang Z, Meng H, Huang Z, Zhang X (2019). Enhanced competitive immunomagnetic beads assay with gold nanoparticles and carbon nanotube-assisted multiple enzyme probes. Sens. Actuat. B: Chemi., 292: 196-202.

Zeng K, Zhang X, Gyimah E, Bu Y, Meng H, Zhang Z (2020). Chemiluminescence imaging immunoassay for simultaneous determination of TBBPA-DHEE and TBBPA-MHEE in aquatic environments. Analy. Bioanaly. Chem., 412: 3673-3681.

Zhang Z, Liu JF, Shao B, Jiang GB (2010). Time-resolved fluoroimmunoassay as an advantageous approach for highly efficient determination of sulfonamides in environmental waters. Environ. Sci. Tech., 44(3): 1030-1035.

Zhang Z, Liu J, Yao Y, Jiang G (2011). A competitive dual-label time-resolved fluoroimmunoassay for the simultaneous determination of chloramphenicol and ractopamine in swine tissue. Chin. Sci. Bull., 56: 1543-1547.

Zhang Z, Zhou K, Bu Y, Shan Z, Liu J, Wu X, Yang L, Chen Z. (2012). Determination of malachite green and crystal violet in environmental water using temperature-controlled ionic liquid dispersive liquid–liquid microextraction coupled with high performance liquid chromatography. Analy. Meth., 4(2): 429-433.

Zheng J, Zhao SQ, Xu XT, Zhang K (2011). Detection of bisphenol A in water samples using ELISA determination method. Water Sci.Tech.: Water Supp., 11(1):55-60.

Zou Y, Zhang Z, Shao X, Chen Y, Wu X, Yang L, Zhu J, Zhang D (2014). Hollow-fiber-supported liquid-phase microextraction using an ionic liquid as the extractant for the pre-concentration of bisphenol A, 17-β-estradiol, estrone and diethylstilbestrol from water samples with HPLC detection. Water Sci. Tech., 69(5): 1028-1035.