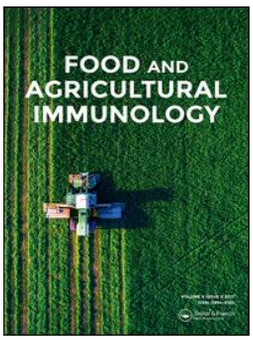

Food and Agricultural Immunology

ISSN: 0954-0105 (Print) 1465-3443 (Online) Journal homepage: https://www.tandfonline.com/loi/cfai20

Development of immunoassays for multi-residue detection of small molecule compounds

Xueyan Cui, Maojun Jin, Pengfei Du, Ge Chen, Chan Zhang, Yudan Zhang, Yong Shao & Jing Wang

To cite this article: Xueyan Cui, Maojun Jin, Pengfei Du, Ge Chen, Chan Zhang, Yudan Zhang, Yong Shao & Jing Wang (2018) Development of immunoassays for multi-residue detection of small molecule compounds, Food and Agricultural Immunology, 29:1, 638-652, DOI: 10.1080/09540105.2018.1428284

To link to this article: https://doi.org/10.1080/09540105.2018.1428284

Published online: 25 Mar 2018.

FOOD AND AGRICULTURAL IMMUNOLOGY
2018, VOL. 29, NO. 1, 638–652
https://doi.org/10.1080/09540105.2018.1428284

Taylor & Francis
Taylor & Francis Group

REVIEW

Development of immunoassays for multi-residue detection of small molecule compounds

Xueyan Cui, Maojun Jin, Pengfei Du, Ge Chen, Chan Zhang, Yudan Zhang, Yong Shao and Jing Wang

Key Laboratory for Agro-Products Quality and Food Safety, Institute of Quality Standards & Testing Technology for Agro-Products, Chinese Academy of Agricultural Sciences, Beijing, People's Republic of China

ABSTRACT

The small molecule compounds like pesticide, veterinary drug, bio-toxin, and heavy metal are widely found in animals, plants, soil, etc. Excessive compounds residues will have a bad influence on human health and the environment. Thus, it is extremely urgent that can detect the small molecules simultaneously. At present, many researches of simultaneous detection for small molecules using the method of an immunoassays have been reported thanks to its advantages of fast speed, simple operation, and high specificity. The small molecules have only one antigenic determinant, so the competitive immunoassay is the main method for small molecule compounds detection. In this paper, our main job is to describe the development of immunoassay for multi-residue detection of small molecule compounds and introduce three ways to complete the analysis of multi-residue immunoassay of small molecule compounds. We also summarize deficiencies and make an expectation of the immunoassays.

ARTICLE HISTORY
Received 12 October 2017
Accepted 10 January 2018

KEYWORDS
Small molecule compounds; multi-residue; immunoassays

1. Introduction

Small molecule compounds include pesticides, veterinary drugs, bio-toxins, and environmental pollutants of small molecules (PCBs, dioxins), which represent a threat to human health and contaminate water, air, soil, and agricultural products (Malarkodi, Rajeshkumar, & Annadurai, 2017). Therefore, developing methods to detect small molecule compounds is of great value. A series of detection techniques for small molecule compounds have been developed, such as high-efficiency liquid chromatography (Beale, Kaserzon, Porter, Roddick, & Carpenter, 2010; Rejczak & Tuzimski, 2017; Xiong et al., 2016), gas chromatography detection (GC) (Farina, Abdullah, Bibi, & Khalik, 2017; Naksen et al., 2016; Qin, Qiao, Wang, & Zhao, 2010), gas chromatography-mass spectrophotometry detection (GC-MS) (Huo et al., 2016; Lee et al., 2017; Ozcan & Balkan, 2017), and liquid chromatography-mass spectrometry detection (LC-MS) (Grund, Marvin, & Rochat, 2016; Kmellár, Pareja, Ferrer, Fodor, & Fernández-Alba, 2011; Valese et al.,

CONTACT Maojun Jin katonking@163.com Key Laboratory for Agro-Products Quality and Food Safety, Institute of Quality Standards & Testing Technology for Agro-Products, Chinese Academy of Agricultural Sciences, Beijing 100081, People's Republic of China

2017). However, these methods are limited by the expensive apparatuses required, long time needed for analysis, and requirement of professional staff, limiting the feasibility of their use in undeveloped areas. The immunoassay involving the specific binding of an antigen to an antibody has the advantages of simplicity, low cost, and high sensitivity (Shankaran, Gobi, & Miura, 2007; Yin, Yin, & Xu, 2008). An antigen contains immunogenicity and reactogenicity. However, small molecule compounds which only have reactogenicity are regarded as haptens (Kim, Cho, Lee, & Lee, 2003; Mitchell, 2010; Yu, Yu, Undersander, & Chu, 1999). Moreover, small molecules have low molecular weights and one antigenic determinant (Kobayashi & Oyama, 2011; Spinks, 2000). For this reason, small molecules can only combine with one antibody and cannot combine with two or more antibodies simultaneously. Therefore, competitive immunoassays are the main method for detecting small molecule compounds (Deng, Li, & He, 2016; Ha, Chung, & Bae, 2016; Zhang, Lai, & Yang, 2013). In addition, non-competitive immunoassays are also being used to detect small molecule compounds (Dong et al., 2014; Islam et al., 2011; Kobayashi et al., 2003; Li, Jeon, Suh, & Kim, 2011). Thus, there is great value in studying the detection of small molecules using immunoassays.

Detecting different types of small molecule compounds in the same sample requires a repeated operation, which is a time-consuming and energy-demanding process. However, a method that can detect multiple types of pesticides (veterinary drugs, bio-toxins, heavy metals, etc.) or different types of small molecules at the same time will result in an easy, fast, and inexpensive process (Chen et al., 2017; Jiang & Fan, 2012; Li et al., 2017). Hence, research on multi-residue detection methods is currently of great significance. Due to the long growth cycle of plants and to protect against various diseases and insects, farmers will spray a variety of pesticides (Tadevosyan, Tadevosyan, Kelly, Gibbs, & Rautiainen, 2013). Thus, the cultivation of plant-type agricultural products requires the detection of multiple pesticides simultaneously. There are three ways to analyse multi-residue immunoassays for small molecule compounds. The first is to design a universal hapten for the multi-residue analysis of small molecules (Liang, Liu, Liu, Yu, & Fan, 2008). The second is to form multiple antigenic determinants by connecting multiple haptens on the carrier protein or prepare multiple specific antibodies to perform the multi-residue analysis of small molecules (Mukunzi et al., 2016; Qiao et al., 2017). The third is to fluorescently label the antigen or antibody to achieve multiple residue detection (Chen, Wen, et al., 2014; Tang, Zhang, Cheng, & Lu, 2008). The similar structures are required in a first way among the analytes and individual targets cannot be distinguished, the second way can solve the problem but they are limited to detect a few kinds of analytes (Le et al., 2013; Liang, Xie, Wang, Gui, & Zhu, 2013; Yaneva, Ivanov, Todorov, & Godjevargova, 2017).

The immunoassay is an analytical method for detecting various substances using antigen–antibody specific binding reactions (Chen, Xu, et al., 2014; Hage, 1999). Immunoassays can be divided into unlabelled immunoassays and labelled immunoassays. Unlabelled immunoassays include immunodiffusion and immunoelectrophoresis. Labelled immunoassays include the radioimmunoassay (RIA), fluorescence immunoassay (FIA), chemiluminescence immunoassay (CLIA), enzyme-linked immunoassay (ELISA), and bio-barcode immunoassay. No matter the single or multiple detections for the small molecules, the most commonly used immunoassay method is ELISA (Briggs, Tapper, Sprosen, Mace, & Finch, 2017; Pavón, González, Martín, & García, 2012).

2. Immunoassays for the detection of small molecule compounds

Immunoassays are based on the specific antigen–antibody binding. The enzyme or luminescent substance is labelled with an antibody or antigen to detect the test compound in a sample. Immunoassays can be either non-competitive (Acharya & Dhar, 2008) or competitive (Hu et al., 2013). Competitive immunoassays are used to detect small molecule compounds with only one antigenic determinant, whereas non-competitive immunoassays are generally used to detect macromolecular compounds such as proteins and polysaccharides with multiple antigenic epitopes (Du et al., 2018; Rao et al., 2016).

2.1. Competitive immunoassays for the detection of small molecule compounds

Competitive immunoassay means the analyte and the hapten-carrier protein conjugations can react with the antibody simultaneous. Besides many reports on competitive immunoassay have been reported (Carter, Triplett, Striemer, & Miller, 2016; Yu et al., 2018). The RIA was first proposed by the American chemists Yalow and Berson in 1958 as the labelled immunoassay (Wang, 2008). The basic principle of the competitive immunoassay is to label the antigen with a radioisotope, and then, both the small molecule compound to be detected and the antigen will react with the antibody (Berson & Yalow, 2006). Thus, the amount of the small molecule to be tested is negatively correlated with the amount of the antigen. The disadvantages of the RIA include poor repeatability and a high rate of non-specific binding (Yan, 2013).

In the FIA, the antigen or antibody or hapten is labelled with a fluorescent substance, next the fluorescent molecules enter an excited state after irradiation with a light source. When the molecules in the excited state return to the ground state, the fluorescent substance emits light, making it possible to calculate the content of the analyte based on the fluorescence intensity. However, a disadvantage of the method is the large amount of background interference (Feng, Shan, Hammock, & Kennedy, 2003; van Genderen et al., 2010; Wu, 2008).

The basic principle of the CLIA is similar to that of the FIA. The antigen or antibody is labelled with a luminescent substance, and a luminescent substrate is added after the reaction ends (Zhao, Sun, & Chu, 2009). Thus, the content of the analyte can be calculated based on the fluorescence intensity (Wang, Wu, Zong, Xu, & Ju, 2012). The difference between the CLIA and FIA is that the CLIA does not require excitation with a light source, whereas the FIA does.

The bio-barcode assay immunoassay was proposed in 2003 by Mirkin et al. (Nam, Thaxton, & Mirkin, 2003). In this method, a conjugate of hapten and a carrier protein is coated on the surface of magnetic nanoparticles or used to coat 96-well plates, and not only the conjugate but also the small molecule compound to be detected can react with the antibody. The method is simple to perform and can be easily popularized and applied (Du et al., 2016; Yang, Zhuang, Chen, Ping, & Bu, 2015).

2.2. Non-competitive immunoassays for the detection of small molecule compounds

The pattern of the non-competitive immunoassay for small molecule compounds is different from the conventional pattern of the "double antibody sandwich," that is, "antibody–

antigen–antibody" (Barnard, Karsiliyan, & Kohen, 1991; Kobayashi & Goto, 2001). Compared with the competitive immunoassay, the non-competitive immunoassay has a higher sensitivity, wider linear range, and higher selectivity (Chen, He, & Xu, 2014; Hua et al., 2015). Because of continuous research efforts and the progress made in science and technology, non-competitive immunoassays can also be used for the detection of small molecule compounds. There have been many studies on non-competitive immunoassay for the detection of small molecules (Akter et al., 2016; Dalgleish & Kennedy, 1988; Liu, Anfossi, Shen, Li, & Wang, 2017). This type of assay has several modes: anti-idiotype antibody (Hu et al., 2017), biotin-avidin-based sandwich immunoassay, anti-metatype antibody, and open sandwich immunoassay (OSIA) (Wang et al., 2017).

In the biotin-avidin-based sandwich immunoassay, the small molecule to be tested is acylated, and then, the small molecule to be tested can be combined with the labelled antibody and solid-phase avidin (Ishikawa, Tanaka, & Hashida, 1990). This mode has a high sensitivity, but it involves a complicated procedure.

The anti-metatype antibody can recognize the complex of the analyte and the antibody but cannot recognize the antibody or analyte alone. Therefore, a shortcoming of this mode is that it is difficult to identify antibodies that recognize antigen–antibody complexes. Hence, a new method called the phage anti-immune complex assay was established (González-Techera, Vanrell, Last, Hammock, & González-Sapienza, 2007), which has a high specificity, high sensitivity, and short screening cycle (González-Techera et al., 2015; Kim et al., 2010).

The reaction of the OSIA occurs in the variable regions; that is, the V_H and V_L regions can bind the small molecule to be detected (Hara, Dong, & Ueda, 2013; Ueda, 2002). Because only one antibody is used, there is increasingly more research on this method. Recently, the open sandwich based on the ELISA, immuno-field effect transistor (Sakata, Ihara, Makino, Miyahara, & Ueda, 2009), and FIA (Chung, Makino, Ohmuro-Matsuyama, & Ueda, 2017) have been reported. However, not all small molecules can bind the V_H and V_L regions; thus, some antibodies will not be able to combine with the analyte, so the OSIA has been limited in practical applications.

3. Approaches to multi-residue immunoassays for small molecule compounds

Specific antibodies are produced when the mice or rabbits or some other mammals were immunized with the corresponding specific small molecule substances-ovalbumin conjugates, which have the same structure with analytes (Tochi et al., 2016; Yaneva, Ivanov, & Godjevargova, 2017). To be specific, different individual conjugates as immunogens were injected into different mammals, which can produce the monoclonal antibodies. Therefore, the monoclonal antibody produced by a mouse can recognize the individual target which was injected into the mouse rather than recognizing other targets. In contrast, the polyclonal antibodies can recognize the family of the target compounds, as some structures among them are same (Bai et al., 2017; Ryan, Jones, Mitchell, & Mett, 2001; Wang, Zhang, Zhang, & Shen, 2011).

3.1. Preparation of a broad-specificity hapten or broad-specificity antibody

Analytes can be detected by preparing a broad-specificity hapten if these substances have the same molecular structure. Meng developed an assay for detecting organic phosphorus pesticides, including chlorpyrifos, triazophos, and phoxim, by preparing a broad-specificity hapten or broad-specificity antibody. These three pesticides all have a diethyl general structure. Thus, diethyl phosphonic acid as a broad-specificity hapten can detect these three pesticides simultaneously. Jiao developed an assay for detecting 10 analytes including 6 penicillins (amoxicillin, penicillin G, ampicillin, penicillin V, carbenicillin, and sulbenicillin) and four tetracyclines (tetracycline, oxytetracycline, doxycycline, and chlortetracycline) by preparing a broad-specificity antibody. The 10 analytes were coupled to bovine serum albumin in turn and were injected into the rabbit, which could produce the broad-specificity antibody. The broad-specificity antibodies can recognize the 10 analytes simultaneously (Jiao, Liu, Zhang, Zhao, & Wang, 2012). However, although this method can detect multiple residues of small molecules, they have to be the same type of small molecule. Moreover, the total content of the same types of small molecule can be detected rather than the concentration of each specific substance.

3.2. Preparation of multiple antigenic determinants or preparation of a variety of specific antibodies

If the compounds to be detected do not have the same molecular structure, then the method of preparing a universal hapten cannot be used. Instead, the substances are detected by preparing multiple antigenic determinants or preparing a variety of specific antibodies. Chen et al. developed a lateral flow immunoassay (LFIA) for detecting aflatoxin B_1, maize ketone, and ochratoxin A in maize, rice, and peanut. In this assay, specific antibodies for all three compounds were prepared, and then, the three antibodies reacted with their respective antigens (Chen, Chen, Han, Zhou, et al., 2016). Guo developed a multiplex bead-based competitive immunoassay based on suspension array technology for the simultaneous detection of the pesticides triazophos, carbofuran, and chlorpyrifos. In this paper, the three hapten–BSA conjugates were coupled with the beads. The three pesticide standards and beads with hapten–BSA conjugates can competitively react with monoclonal antibodies; goat anti-mouse IgG secondary antibodies labelled with horseradish peroxidase were added and were measured subsequently (Guo, Tian, Liang, Zhu, & Gui, 2013).

3.3. Multiple residue detection by fluorescently labelling the antigen or antibody

On the one hand, different fluorescent materials can be used, the different fluorescent materials are labelled with the corresponding antibodies or antigens, and the corresponding antibodies or antigens then undergo an antibody–antigen immune response with the antigens or antibodies. Qualitative and quantitative analyses are carried out using the characteristics and intensity of the fluorescence signals. This method is characterized by a fast speed and high specificity. On the other hand, the same fluorescent materials can be used, these same fluorescent materials are labelled with the corresponding antibodies or antigens, and the corresponding antibodies or antigens are used to coat the wells of

96-well plates. The fluorescence signal intensity in the wells is detected with a microplate reader. The fluorescent materials that can be used in this approach include organic dye molecules and quantum dots. Ye, Li, Zuo, and Li (2008) developed an assay for detecting sulfamerazine, streptomycin, and tylosin simultaneously using the organic dye molecule Cy5. The basic principle is that the antigen carrier protein and hapten were conjugated, the complex was then used to coat 96-well plates, and fluorescently labelled antibodies were added to the wells. Peng et al. (2009) developed an assay for detecting dexamethasone, gentamicin, clonazepam, medroxyprogesterone acetate, and ceftiofur simultaneously based on multicolour quantum dot probes. In this assay, the coating antigen carrier protein and hapten were conjugated and used to coat 96-well plates, and then, quantum dot-labelled antibodies were added to the wells. The maximum emission wavelengths of the five quantum dots used are 520, 545, 570, 590, and 635 nm. After the reaction is completed, the fluorescence intensity of each well is measured.

4. The application of immunoassays in multi-residue detection

4.1. Multi-residue detection of pesticides

Limited amounts of pesticide residues are permitted in agricultural products, but pesticide residues frequently exceeded the maximum residue limit. The problem affects the quality and safety of agricultural products and has captured wide public attention.

Niusha (2016) presented an ELISA and a CLIA for the determination of three pesticide (fenpropathrin, decamethrin, λ-cyhalothrin) residues and compared the results of the two methods. Pesticide residues were detected by preparing a broad-specificity hapten in this assay. The three pesticides have an α-cyano pyrethroid structure, so the common moiety of α-cyano pyrethroids without one side chain of the cyclopropane ring was used as the broad-specificity hapten. The conjugation of the broad-specificity hapten and coating carrier protein was regarded as the immunogen, and it could produce specific antibodies. The IC_{50} values of fenpropathrin, decamethrin, and λ-cyhalothrin using this ELISA were 2.6, 8.2, and 31.1 ng/mL, respectively.

4.2. Multi-residue detection of veterinary drugs

Chen developed an assay for detecting β-lactams, tetracyclines, quinolones, and sulphonamides in milk using a near-infrared fluorescence-based multiplex LFIA. Multi-residues of the four veterinary drugs were detected by preparing four specific antibodies in this assay. The limits of detection of the four veterinary drugs were 8, 2, 4, and 8 ng/mL. The detection ranges of the four veterinary drugs were 0.26–3.56, 0.04–0.98, 0.08–2.0, and 0.1–3.98 ng/mL, and the linear correlation coefficients were greater than 0.97. The recovery rate of spiked samples ranged from 93.7% to 108.2%, and the coefficient of variation was less than 16.3% (Chen, Chen, Han, Liu, et al., 2016). Wang (2013) developed an assay for detecting streptomycin, tetracycline, and penicillin G in milk simultaneously using an FIA with quantum dots. Multi-residues of the three veterinary drugs were detected by preparing three specific antibodies in this assay. The linger ranges of the

three veterinary drugs were 0.01–25, 0.01–25, and 0.01–10 ng/mL. The limit of detection (LOD) of the three veterinary drugs was less than 0.005 ng/mL.

4.3. Multi-residue detection of bio-toxins

Zhu (2016) presented a time-resolved FIA for measuring zearalenone, aflatoxin B_1, and chlorothalonil in corn. Multi-residues of the three bio-toxins were detected by preparing three specific antibodies, and the detection limits were 0.043 μg/kg for zearalenone, 0.011 μg/kg for aflatoxin B_1, and 0.084 μg/kg for chlorothalonil. Guo et al. (2014) presented an ELISA for the determination of deoxynivalenol (DON), 3-acetyl-DON, and 15-acetyl-DON. The IC_{50} values of the three bio-toxins were 22, 15, and 34 ng/mL, respectively.

4.4. Multi-residue detection of persistent organic pollutants

Yang (2014) developed an assay for the detection of polychlorinated biphenyls, including PCB12, PCB37, PCB77, and Aroclor1248, using a bio-barcode direct competitive immunoassay and bio-barcode indirect competitive immunoassay. Multi-residues of the four environmental pollutants were detected by preparing four specific antibodies in this assay. The LOD values of the four compounds by the bio-barcode direct competitive immunoassay were 2.63, 4.35, 1.72, and 10.20 pg/L. However, the limit detection values of the four compounds by the bio-barcode indirect competitive immunoassay were 4.36, 4.64, 9.12, and 2.55 pg/L.

4.5. Multi-residue detection of illegal additives

Shan, Xi, Sun, Zhang, and Wang (2012) developed an ELISA for the detection of Sudan red-1, Sudan red-2, Sudan red-3, Sudan red-4, para red, and Sudan red-G in eggs. Multi-residues of these six environmental pollutants were detected by a preparing broad-specificity hapten. The broad-specificity hapten had a structure in which 4-amino-3-methylbenzoic acid was attached to o-toluidine and then bound to beta-naphthol, which detected the six small molecule compounds simultaneously. The detection limits of the six small molecule compounds ranged from 0.08 to 0.2 ng/g. Some immunoassay methods for detecting pesticides, veterinary drugs, bio-toxins, environmental pollutants, and illegal additives are listed in Table 1.

5. Conclusions

The main methods for the detection of multiple residues of small molecule compounds include RIA, FIA, CLIA, ELISA, and bio-barcode assay immunoassay. These immunological techniques have their own advantages and disadvantages. Immunoassay methods are used to detect a specific substance instead of an unknown substance. However, commercial monoclonal antibodies are expensive, and some small molecule antibodies are difficult to prepare.

Compared with GC-MS and LC-MS methods, immunoassay methods feature simpler sample processing steps, faster processing speeds, and lower costs. With the development

Table 1. Examples of multi-residue immunoassays for the detection of small molecules.

Group	Compounds	Method	Sensitivity (μg/kg)	References
Pesticide	Parathion, methyl parathion, fenitrothion	CLIA	1.24–5.57[a]	Zou et al. (2017)
	Organothiophosphate pesticides, organophosphate pesticides	ELISA	13–135[b]	Li, Zhao, Zhao, and Yang (2015)
	Chlorpyrifos fenthion	ELISA	0.20–0.50[b]	Navarro et al. (2013)
Veterinary drug	Clindamycin, lincomycin	ELISA	1.8, 6.8[b]	He et al. (2017)
	Penicillin G, amoxicillin, ampicillin, penicillin V, cloxacillin, oxacillin, cloxacillin, carbenicillin, dicloxacillin, methicillin, nafcillin	ELISA	0.7–9.3[b]	Jiao (2013)
	Streptomycin, tetracycline, penicillin G	FIA	5×10^{-3}[b]	Song et al. (2015)
Bio-toxin	Deoxynivalenol, zearalenone, T2/HT2	LFIA	80–1000[b]	Foubert, Beloglazova, and De Saeger (2017)
	Fumonisin B_1, fumonisin B_2	FIA	157.4, 290.6[b]	Li, Mi, et al. (2015)
Persistent organic pollutants	Permethrin, Aroclor1248, Aroclor1254	ELISA	1.5–14.3[c]	Bronshtein, Chuang, Van Emon, and Altstein (2012)
Illegal additive	Malachite green, brilliant green, crystal violet	ELISA	1.34–1.98[a]	Shen et al. (2011)
	Sudan red-1, Sudan red-2, Sudan red-3, Sudan red-4, Sudan red-G	ELISA	0.2–0.6[b]	Chang et al. (2011)

[a]IC_{50};
[b]LOD;
[c]I_{20}.

of immunological techniques, the sensitivity and application range of these assays continue to increase.

6. Prospects

Immunoassays have attracted attention because they allow fast analysis in the field. There are three methods for multiple residue detection of small molecule compounds: preparation of a broad-specificity hapten or broad-specificity antibody, preparation of multiple antigenic determinants or preparation of a variety of specific antibodies, and multiple residue detection by fluorescently labelling the antigen or antibody. Two of these methods can be combined to expand the detection range. Because of the hazards associated with these compounds, researching and developing methods to detect a variety of small molecule compounds simultaneously and quickly has become a popular area of study.

With the development of science and technology, there have been several new immunoassays, such as aptamer technology. Aptamer technology replaced the traditional pattern in which an antibody binds specifically to the target substance. Furthermore, with of the development of nanotechnology, nano-materials such as nano-gold, quantum dots, and carbon materials are being used in immune technologies to amplify signals and improve sensitivity. The multiple detection of small molecules using immunoassays has good development prospects with the development of gene engineering and cell engineering techniques.

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

The authors gratefully thank the projects of National Key Research Program of China (2016YFD0401101); National Natural Science Foundation (31671938); and Innovation team "Residual detection and behavior research of agricultural chemical pollutants" for the financial support.

References

Acharya, D., & Dhar, T. K. (2008). A novel broad-specific noncompetitive immunoassay and its application in the determination of total aflatoxins. *Analytica Chimica Acta*, *630*(1), 82–90. doi:10.1016/j.aca.2008.09.063

Akter, S., Vehniäinen, M., Spoof, L., Nybom, S., Meriluoto, J., & Lamminmäki, U. (2016). Broad-spectrum noncompetitive immunocomplex immunoassay for cyanobacterial peptide hepatotoxins (microcystins and nodularins). *Analytical Chemistry*, *88*, 10080–10087. doi:10.1021/acs.analchem.6b02470

Bai, Y., Hu, J. Y., Liu, S. Z., Zhang, W. Y., Zhang, J., He, J., … Wang, Z. H. (2017). Production of antibodies and development of an enzyme-linked immunosorbent assay for 17β-estradiol in milk. *Food and Agricultural Immunology*, *28*(6), 1519–1529. doi:10.1080/09540105.2017.1350833

Barnard, G., Karsiliyan, H., & Kohen, F. (1991). Idiometric assay, the third way: A noncompetitive immunoassay for small molecules. *American Journal of Obstetrics and Gynecology*, *165*, 1997–2000. doi:10.1016/S0002-9378(11)90565-1

Beale, D. J., Kaserzon, S. L., Porter, N. A., Roddick, F. A., & Carpenter, P. D. (2010). Detection of s-triazine pesticides in natural waters by modified large-volume direct injection HPLC. *Talanta*, *82*(2), 668–674. doi:10.1016/j.talanta.2010.05.030

Berson, S. A., & Yalow, R. S. (2006). General principles of radioimmunoassay. *Clinica Chimica Acta*, *369*(2), 125–143. doi:10.1016/j.cca.2006.05.002

Briggs, L., Tapper, B., Sprosen, J., Mace, W., & Finch, S. (2017). Development of an enzyme-linked immunosorbent assay for the detection of lolines in pastures. *Food and Agricultural Immunology*, *28*(6), 1058–1070. doi:10.1080/09540105.2017.1326466

Bronshtein, A., Chuang, J. C., Van Emon, J. M., & Altstein, M. (2012). Development of a multianalyte enzyme-linked immunosorbent assay for permethrin and aroclors and its implementation for analysis of soil/sediment and house dust extracts. *Journal of Agricultural and Food Chemistry*, *60*(17), 4235–4242. doi:10.1021/jf300043g

Carter, J. A., Triplett, E., Striemer, C. C., & Miller, B. L. (2016). A label-free, multiplex competitive assay for small molecule pollutants. *Biosensors and Bioelectronics*, *77*, 1–6. doi:10.1016/j.bios.2015.08.064

Chang, X. C., Hu, X. Z., Li, Y. Q., Shang, Y. J., Liu, Y. Z., Feng, G., & Wang, J. P. (2011). Multi-determination of Para red and Sudan dyes in egg by a broad specific antibody based enzyme linked immunosorbent assay. *Food Control*, *22*(1), 1770–1775. doi:10.1016/j.foodcont.2011.04.014

Chen, Y. Q., Chen, Q., Han, M. M., Liu, J. Y., Zhao, P., He, L. D., … Zhang, L. Y. (2016). Near-infrared fluorescence-based multiplex lateral flow immunoassay for the simultaneous detection of four antibiotic residue families in milk. *Biosensors and Bioelectronics*, *79*, 430–434. doi:10.1016/j.bios.2015.12.062

Chen, Y. Q., Chen, Q., Han, M. M., Zhou, J. Y., Gong, L., Niu, Y. M., … Zhang, L. Y. (2016). Development and optimization of a multiplex lateral flow immunoassay for the simultaneous determination of three mycotoxins in corn, rice and peanut. *Food Chemistry*, *213*, 478–484. doi:10.1016/j.foodchem.2016.06.116

Chen, B., He, Q. H., & Xu, Y. (2014). Latest progress on noncompetitive immunoassays for small molecules. *Food Science*, *35*(15), 310–313.

Chen, X. J., Li, Z. Z., Guo, J. Y., Li, D. M., Gao, H. L., Wang, Y., & Xu, C. L. (2017). Simultaneous screening for marbofloxacin and ofloxacin residues in animal derived foods using an indirect

competitive immunoassay. *Food and Agricultural Immunology*, *28*(3), 489–499. doi:10.1080/09540105.2017.1297780

Chen, M., Wen, K., Tao, X. Q., Ding, S. Y., Xie, J., Yu, X. Z., … Jiang, H. Y. (2014). A novel multiplexed fluorescence polarisation immunoassay based on a recombinant bi-specific single-chain diabody for simultaneous detection of fluoroquinolones and sulfonamides in milk. *Food Additives &Contaminants: Part A*, *31*(12), 1959–1967. doi:10.1080/19440049.2014.976279

Chen, X. J., Xu, L. G., Ma, W., Liu, L. Q., Kuang, H., Wang, L. B., & Xu, C. L. (2014). General immunoassay for pyrethroids based on a monoclonal antibody. *Food and Agricultural Immunology*, *25*(3), 341–349. doi:10.1080/09540105.2013.794328

Chung, C. I., Makino, R., Ohmuro-Matsuyama, Y., & Ueda, H. (2017). Development of a fluorescent protein-antibody Förster resonance energy transfer probe for the detection and imaging of osteocalcin. *Journal of Bioscience and Bioengineering*, *123*(2), 272–276. doi:10.1016/j.jbiosc.2016.09.003

Dalgleish, A. G., & Kennedy, R. C. (1988). Anti-idiotypic antibodies as immunogens: Idiotype-based vaccines. *Vaccine*, *6*(3), 215–220. doi:10.1016/0264-410X(88)90213-7

Deng, X. L., Li, P., & He, W. H. (2016). Recent advances in immunoassays for small molecules. *Food Science*, *37*(11), 277–282.

Dong, J. X., Xu, C., Wang, H., Xiao, Z. L., Gee, S. J., Li, Z. F., … Hammock, B. D. (2014). Enhanced sensitive immunoassay: Noncompetitive phage anti-immune complex assay for the determination of malachite green and leucomalachite green. *Journal of Agricultural and Food Chemistry*, *62*(34), 8752–8758. doi:10.1021/jf5019824

Du, P. F., Jin, M. J., Chen, G., Zhang, C., Cui, X. Y., Zhang, Y. D. … Wang, J. (2016). Research and progress of bio-barcode assay in the detection of agricultural product. *Analytical Instrumentation*, *S1*, 59–63.

Du, P. F., Jin, M. J., Chen, G., Zhang, C., Cui, X. Y., Zhang, Y. D., … Wang, J. (2018). Highly sensitive detection of triazophos pesticide using a novel bio-bar-code amplification competitive immunoassay in a micro well plate-based platform. *Sensors and Actuators B: Chemical*, *256*, 457–464. doi:10.1016/j.snb.2017.10.075

Farina, Y., Abdullah, M. P., Bibi, N., & Khalik, W. M. A. W. M. (2017). Determination of pesticide residues in leafy vegetables at parts per billion levels by a chemometric study using GC-ECD in Cameron Highlands, Malaysia. *Food Chemistry*, *224*, 55–61. doi:10.1016/j.foodchem.2016.11.113

Feng, J., Shan, G. M., Hammock, B. D., & Kennedy, I. M. (2003). Fluorescence quenching competitive immunoassay in micro droplets. *Biosensors and Bioelectronics*, *18*(8), 1055–1063. doi:10.1016/S0956-5663(02)00218-X

Foubert, A., Beloglazova, N. V., & De Saeger, S. (2017). Comparative study of colloidal gold and quantum dots as labels for multiplex screening tests for multi-mycotoxin detection. *Analytica Chimica Acta*, *955*, 48–57. doi:10.1016/j.aca.2016.11.042

González-Techera, A., Vanrell, L., Last, J. A., Hammock, B. D., & González-Sapienza, G. (2007). Phage anti-immune complex assay: General strategy for noncompetitive immunodetection of small molecules. *Analytical Chemistry*, *79*(20), 7799–7806. doi:10.1021/ac071323h

González-Techera, A., Zon, M. A., Molina, P. G., Fernández, H., González-Sapienza, G., & Arévalo, F. J. (2015). Development of a highly sensitive noncompetitive electrochemical immunosensor for the detection of atrazine by phage anti-immunocomplex assay. *Biosensors and Bioelectronics*, *64*, 650–656. doi:10.1016/j.bios.2014.09.046

Grund, B., Marvin, L., & Rochat, B. (2016). Quantitative performance of a quadrupole-orbitrap-MS in targeted LC–MS determinations of small molecules. *Journal of Pharmaceutical and Biomedical Analysis*, *124*, 48–56. doi:10.1016/j.jpba.2016.02.025

Guo, Y., Sanders, M., Galvita, A., Heyerick, A., Deforce, D., Bracke, M., … De Saeger, S. (2014). Heterologous screening of hybridomas for the development of broad-specific monoclonal antibodies against deoxynivalenol and its analogues. *World Mycotoxin Journal*, *7*(3), 257–265. doi:10.3920/WMJ2013.1668

Guo, Y. R., Tian, J., Liang, C. Z., Zhu, G. N., & Gui, W. J. (2013). Multiplex bead-array competitive immunoassay for simultaneous detection of three pesticides in vegetables. *Microchimica Acta*, *180*(5–6), 387–395. doi:10.1007/s00604-013-0944-4

Ha, M. S., Chung, M. S., & Bae, D. H. (2016). Surface modification techniques and competitive immunoassay to detect residual ciprofloxacin in foods. *Food and Agricultural Immunology*, *27* (6), 886–896. doi:10.1080/09540105.2016.1202212

Hage, D. S. (1999). Immunoassays. *Analytical Chemistry*, *71*(12), 294–304. doi:10.1021/a1999901+

Hara, Y., Dong, J., & Ueda, H. (2013). Open-sandwich immunoassay for sensitive and broad-range detection of a shellfish toxin gonyautoxin. *Analytica Chimica Acta*, *793*(5), 107–113. doi:10.1016/j.aca.2013.07.024

He, J., Wu, N., Luo, P. J., Guo, P., Qu, J. W., Zhang, S. Y., … Jiang, W. X. (2017). Development of a heterologous enzyme-linked immunosorbent assay for the detection of clindamycin and lincomycin residues in edible animal tissues. *Meat Science*, *125*, 137–142. doi:10.1016/j.meatsci.2016.11.024

Hu, W. H., Li, X., He, G. L., Zhang, Z. W., Zheng, X. T., Li, P. W., & Li, C. M. (2013). Sensitive competitive immunoassay of multiple mycotoxins with non-fouling antigen microarray. *Biosensors and Bioelectronics*, *50*, 338–344. doi:10.1016/j.bios.2013.06.037

Hu, L., Liu, A., Chen, W., Yang, H., Wang, X., & Chen, F. (2017). A non-toxic enzyme-linked immunosorbent assay for aflatoxin B1 using anti-idiotypic antibodies as substitutes. *Journal of The Science of Food and Agriculture*, *97*, 1640–1645. doi:10.1002/jsfa.7914

Hua, X. D., Zhou, L. L., Feng, L., Ding, Y., Shi, H. Y., Wang, L. M., … Wang, M. H. (2015). Competitive and noncompetitive phage immunoassays for the determination of benzothiostrobin. *Analytica Chimica Acta*, *890*, 150–156. doi:10.1016/j.aca.2015.07.056

Huo, F. F., Tang, H., Wu, X., Chen, D. Z., Zhao, T., Liu, P., & Li, L. (2016). Utilizing a novel sorbent in the solid phase extraction for simultaneous determination of 15 pesticide residues in green tea by GC/MS. *Journal of Chromatography B*, *1023–1024*, 44–54. doi:10.1016/j.jchromb.2016.04.053

Ishikawa, E., Tanaka, K., & Hashida, S. (1990). Novel and sensitive noncompetitive (two-site) immunoassay for haptens with emphasis on peptides. *Clinical Biochemistry*, *23*(5), 445–453. doi:10.1016/0009-9120(90)90238-P

Islam, K. N., Ihara, M., Dong, J. H., Kasagi, N., Mori, T., & Ueda, H. (2011). Direct construction of an open-sandwich enzyme immunoassay for one-step noncompetitive detection of thyroid hormone T4. *Analytical Chemistry*, *83*(3), 1008–1014. doi:10.1021/ac102801r

Jiang, H., & Fan, M. T. (2012). Multi-analyte immunoassay for pesticides: A review. *Analytical Letters*, *45*(11), 1347–1364. doi:10.1080/00032719.2012.675493

Jiao, S. N. (2013). Study on the immunoassay for detection of penicillin drugs in milk (Doctoral dissertation). Retrieved from http://d.wanfangdata.com.cn/Thesis_Y2386874.aspx?gathStatIcon=true

Jiao, S. N., Liu, J., Zhang, Y. F., Zhao, G. X., & Wang, J. P. (2012). Preparation of a bi-hapten antigen and the broad-specific antibody for simultaneous immunoassay of penicillins and tetracyclines in milk. *Food and Agricultural Immunology*, *23*(3), 273–287. doi:10.1080/09540105.2011.624175

Kim, Y. J., Cho, Y. A., Lee, H. S., & Lee, Y. T. (2003). Investigation of the effect of hapten heterology on immunoassay sensitivity and development of an enzyme-linked immunosorbent assay for the organophosphorus insecticide fenthion. *Analytica Chimica Acta*, *494*(1–2), 29–40. doi:10.1016/j.aca.2003.07.003

Kim, H. J., Rossotti, M. A., Ahn, K. C., González-Sapienza, G. G., Gee, S. J., Musker, R., & Hammock, B. D. (2010). Development of a noncompetitive phage anti-immunocomplex assay for brominated diphenyl ether 47. *Analytical Biochemistry*, *401*(1), 38–46. doi:10.1016/j.ab.2010.01.040

Kmellár, B., Pareja, L., Ferrer, C., Fodor, P., & Fernández-Alba, A. R. (2011). Study of the effects of operational parameters on multiresidue pesticide analysis by LC–MS/MS. *Talanta*, *84*(2), 262–273. doi:10.1016/j.talanta.2010.12.006

Kobayashi, N., & Goto, J. (2001). Noncompetitive immunoassays for small molecules with high sensitivity and specificity. *Advances in Clinical Chemistry*, *36*, 139–170. doi:10.1016/S0065-2423(01)36027-4

Kobayashi, N., Kubota, K., Oiwa, H., Goto, J., Niwa, T., & Kobayashi, K. (2003). Idiotype–anti-idiotype-based noncompetitive enzyme-linked immunosorbent assay of ursodeoxycholic acid 7-N-

acetylglucosaminides in human urine with subfemtomole range sensitivity. *Journal of Immunological Methods*, *272*, 1–10. doi:10.1016/S0022-1759(02)00115-1

Kobayashi, N., & Oyama, H. (2011). Antibody engineering toward high-sensitivity high-throughput immunosensing of small molecules. *The Analyst*, *136*, 642–651. doi:10.1039/C0AN00603C

Le, T., Xu, J., He, H. Q., Niu, X. D., Chen, Y., & Jia, Y. Y. (2013). Development and validation of an enzyme-linked immunosorbent assay for rapid detection of multi- residues of five quinoxaline-1,4-dioxides in animal feeds. *Food and Agricultural Immunology*, *24*(4), 457–466. doi:10.1080/09540105.2012.716024

Lee, J., Kim, L., Shin, Y., Lee, J., Lee, J., Kim, E., … Kim, J. H. (2017). Rapid and simultaneous analysis of 360 pesticides in brown rice, spinach, orange, and potato using microbore GC-MS/MS. *Journal of Agricultural and Food Chemistry*, *65*, 3387–3395. doi:10.1021/acs.jafc.7b00576

Li, T. H., Jeon, K. S., Suh, Y. D., & Kim, M. G. (2011). A label-free, direct and noncompetitive FRET immunoassay for ochratoxin A based on intrinsic fluorescence of an antigen and antibody complex. *Chemical Communications*, *47*, 9098–9100. doi:10.1039/c1cc12604k

Li, C. L., Mi, T. J., Conti, G. O., Yu, Q., Wen, K., Shen, J. Z., … Wang, Z. H. (2015). Development of a screening fluorescence polarization immunoassay for the simultaneous detection of fumonisins B1 and B2 in maize. *Journal of Agricultural and Food Chemistry*, *63*(20), 4940–4946. doi:10.1021/acs.jafc.5b01845

Li, Y. F., Sun, Y. M., Beier, R. C., Lei, H. T., Gee, S., Hammock, B. D., … Xu, Z. L. (2017). Immunochemical techniques for multianalyte analysis of chemical residues in food and the environment: A review. *Trends in Analytical Chemistry*, *88*, 25–40. doi:10.1016/j.trac.2016.12.010

Li, Y. L., Zhao, F. C., Zhao, L. Y., & Yang, Z. Y. (2015). Development of a broad-specificity immunoassay for determination of organophosphorus pesticides using dual-generic hapten antigens. *Food Analytical Methods*, *8*(2), 420–427. doi:10.1007/s12161-014-9906-7

Liang, Y., Liu, X. J., Liu, Y., Yu, X. Y., & Fan, M. T. (2008). Synthesis of three haptens for the class-specific immunoassay of O,O-dimethyl organophosphorus pesticides and effect of hapten heterology on immunoassay sensitivity. *Analytica Chimica Acta*, *615*(2), 174–183. doi:10.1016/j.aca.2008.03.050

Liang, X., Xie, R., Wang, C. M., Gui, W. J., & Zhu, G. N. (2013). Development of a broad-selective immunoassay for multi-residue determination of type II pyrethroids in West Lake water. *Food and Agricultural Immunology*, *24*(1), 59–78. doi:10.1080/09540105.2011.641169

Liu, A. P., Anfossi, L., Shen, L., Li, C., & Wang, X. H. (2017). Non-competitive immunoassay for low-molecular-weight contaminant detection in food, feed and agricultural products: A mini-review. *Trends in Food Science & Technology*, *71*, 181–187. doi:10.1016/j.tifs.2017.11.014

Malarkodi, C., Rajeshkumar, S., & Annadurai, G. (2017). Detection of environmentally hazardous pesticide in fruit and vegetable samples using gold nanoparticles. *Food Control*, *80*, 11–18. doi:10.1016/j.foodcont.2017.04.023

Mitchell, J. (2010). Small molecule immunosensing using surface plasmon resonance. *Sensors*, *10*, 7323–7346. doi:10.3390/s100807323

Mukunzi, D., Tochi, B. N., Isanga, J., Liu, L. Q., Kuang, H., & Xu, C. L. (2016). Development of an immunochromatographic assay for hexestrol and diethylstilbestrol residues in milk. *Food and Agricultural Immunology*, *27*(6), 855–869. doi:10.1080/09540105.2016.1183601

Naksen, W., Prapamontol, T., Mangklabruks, A., Chantara, S., Thavornyutikarn, P., Robson, M. G., … Panuwete, P. (2016). A single method for detecting 11 organophosphate pesticides in human plasma and breastmilk using GC-FPD. *Journal of Chromatography B*, *1025*, 92–104. doi:10.1016/j.jchromb.2016.04.045

Nam, J. M., Thaxton, C. S., & Mirkin, C. A. (2003). Nanoparticle-based bio–bar codes for the ultrasensitive detection of proteins. *Science*, *301*, 1884–1886. doi:10.1126/science.1088755

Navarro, P., Perez, A. J., Gabaldon, J. A., Nunez-Delicado, E., Puchades, R., Maquieira, A., & Morais, S. (2013). Detection of chemical residues in tangerine juices by a duplex immunoassay. *Talanta*, *116*, 33–38. doi:10.1016/j.talanta.2013.04.062

Niusha, T. (2016). Chemiluminescent enzyme immunoassay for rapid detection of three α-cyano pyrethroid residues in agricultural products (Doctoral dissertation). Retrieved from http://d.g.wanfangdata.com.cn/Thesis_Y3029699.aspx

Ozcan, C., & Balkan, S. (2017). Multi-residue determination of organochlorine pesticides in vegetables in Kirklareli, Turkey by gas chromatography-mass spectrometry. *Journal of Analytical Chemistry*, *72*(7), 761–769. doi:10.1134/S1061934817070036

Pavón, MÁ, González, I., Martín, R., & García, T. (2012). Competitive direct ELISA based on a monoclonal antibody for detection of ochratoxin A in dried fig samples. *Food and Agricultural Immunology*, *23*(1), 83–91. doi:10.1080/09540105.2011.604769

Peng, C. F., Li, Z. K., Zhu, Y. Y., Chen, W., Yuan, Y., Liu, L. Q., … Xu, C. L. (2009). Simultaneous and sensitive determination of multiplex chemical residues based on multicolor quantum dot probes. *Biosensors and Bioelectronics*, *24*(12), 3657–3662. doi:10.1016/j.bios.2009.05.031

Qiao, B., Li, Y. S., Meng, X. Y., Sun, Y., Hu, P., Lu, S. Y., … Zhou, Y. (2017). Development of an indirect competitive ELISA for the detection of acenaphthene and pyrene. *Food and Agricultural Immunology*, *28*(5), 789–800. doi:10.1080/09540105.2017.1313201

Qin, S., Qiao, X. W., Wang, X., & Zhao, L. J. (2010). Determination of 4 dithiocarbamate residues in 22 matrices by gas chromatography. *Chinese Journal of Chromatography*, *28*(12), 1162–1167.

Rao, M. F., Wu, W. J., Xu, C., Mao, X. X., Xu, Z. L., Wang, H., … Sun, Y. M. (2016). Progress in noncompetitive detection of small molecules by open sandwich immunoassay. *Food Science*, *37*(07), 219–226.

Rejczak, T., & Tuzimski, T. (2017). QuEChERS-based extraction with dispersive solid phase extraction clean-up using PSA and ZrO2-based sorbents for determination of pesticides in bovine milk samples by HPLC-DAD. *Food Chemistry*, *217*, 225–233. doi:10.1016/j.foodchem.2016.08.095

Ryan, G. B., Jones, W. T., Mitchell, R. E., & Mett, V. (2001). Polyclonal antibody production against chito-oligosaccharides. *Food and Agricultural Immunology*, *13*(2), 127–130. doi:10.1080/09540100120055600

Sakata, T., Ihara, M., Makino, I., Miyahara, Y., & Ueda, H. (2009). Open sandwich-based immunotransistor for label-free and noncompetitive detection of low molecular weight antigen. *Analytical Chemistry*, *81*, 7532–7537. doi:10.1021/ac900457m

Shan, W. C., Xi, J. Z., Sun, J., Zhang, Y. J., & Wang, J. P. (2012). Production of the monoclonal antibody against Sudan 4 for multi-immunoassay of Sudan dyes in egg. *Food Control*, *27*(1), 146–152. doi:10.1016/j.foodcont.2012.03.017

Shankaran, D. R., Gobi, K. V., & Miura, N. (2007). Recent advancements in surface plasmon resonance immunosensors for detection of small molecules of biomedical, food and environmental interest. *Sensors and Actuators B: Chemical*, *121*(1), 158–177. doi:10.1016/j.snb.2006.09.014

Shen, Y. D., Deng, X. F., Xu, Z. L., Wang, Y., Lei, H. T., Wang, H., … Sun, Y. M. (2011). Stimultaneous determination of malachite green, brilliant green and crystal violet in grass carp tissues by a broad-specificity indirect competitive enzyme-linked immunosorbent assay. *Analytica Chimica Acta*, *707*(1), 148–154. doi:10.1016/j.aca.2011.09.006

Song, E. Q., Yu, M. Q., Wang, Y. Y., Hu, W. H., Cheng, D., Swihart, M. T., & Song, Y. (2015). Multicolor quantum dot-based fluorescence immunoassay array for simultaneous visual detection of multiple antibiotic residues in milk. *Biosensors and Bioelectronics*, *72*, 320–325. doi:10.1016/j.bios.2015.05.018

Spinks, C. A. (2000). Broad-specificity immunoassay of low molecular weight food contaminants: New paths to Utopia!. *Trends in Food Sciences & Technology*, *11*(6), 210–217. doi:10.1016/S0924-2244(01)00009-7

Tadevosyan, A., Tadevosyan, N., Kelly, K., Gibbs, S. G., & Rautiainen, R. H. (2013). Pesticide use practices in rural Armenia. *Journal of Agromedicine*, *18*(4), 326–333. doi:10.1080/1059924X.2013.826118

Tang, J. S., Zhang, M., Cheng, G. H., & Lu, Y. T. (2008). Development of fluorescence polarization immunoassay for the detection of organophosphorus pesticides parathion and azinphos-methyl. *Journal of Immunoassay and Immunochemistry*, *29*(4), 356–369. doi:10.1080/15321810802329757

Tochi, B. N., Peng, J., Song, S. S., Liu, L. Q., Kuang, H., & Xu, C. L. (2016). Production and application of a monoclonal antibody (mAb) against ofloxacin in milk, chicken and pork. *Food and Agricultural Immunology*, *27*(5), 643–656. doi:10.1080/09540105.2016.1148125

Ueda, H. (2002). Open sandwich immunoassay: A novel immunoassay approach based on the interchain interaction of an antibody variable region. *Journal of Bioscience and Bioengineering*, *94*(6), 614–619. doi:10.1016/S1389-1723(02)80203-1

Valese, A. C., Molognoni, L., de Souza, N. C., Ploencio, L. A. D., Costa, A. C. O., Barreto, F., & Daguer, H. (2017). Development, validation and different approaches for the measurement uncertainty of a multi-class veterinary drugs residues LC–MS method for feeds. *Journal of Chromatography B*, *1053*, 48–59. doi:10.1016/j.jchromb.2017.03.026

van Genderen, F. T., Gorus, F. K., Vermeulen, I., Vekens, E. M., De Pauw, P. E., Pipeleers, D. G., & Van Schravendijk, C. (2010). Development of a multipurpose time-resolved fluorescence immunoassay for rat insulin. *Analytical Biochemistry*, *404*(1), 8–13. doi:10.1016/j.ab.2010.04.024

Wang, S. T. (2008). Immuno-specificity of multi-determinat artificial antigen and gold immunochromatography assay for multi-residue of pesticide (Doctoral dissertation). Retrieved from http://d.g.wanfangdata.com.cn/Thesis_Y1302640.aspx

Wang, Y. Y. (2013). Stimultaneous and rapid detection of multiplex antibiotic residues in milk with multicolor quantum dots based on fluorescence immunoassay (Doctoral dissertation). Retrieved from http://d.g.wanfangdata.com.cn/Thesis_Y2310617.aspx

Wang, Y. H., Li, Y. S., Song, L. T., Zhang, J., Sun, C. F., You, Y. L., … Yang, J. R. (2017). A review of non-competitive immunoassays for small molecule compounds. *Journal of Food Science and Biotechnology*, *36*(02), 113–121.

Wang, C., Wu, J., Zong, C., Xu, J., & Ju, H. X. (2012). Chemiluminescent immunoassay and its applications. *Chinese Journal of Analytical Chemistry*, *40*(01), 3–10.

Wang, Z. H., Zhang, J., Zhang, S. X., & Shen, J. Z. (2011). Heterologous structure of coating antigen on sensitivity of ELISA for sulfamethazine: Evidence from molecular similarity analysis. *Food and Agricultural Immunology*, *22*(2), 115–124. doi:10.1080/09540105.2010.533752

Wu, Z. M. (2008). The preparation of the sensitity ehancement of fluoroimmunoassay (Doctoral dissertation). Retrieved from http://d.g.wanfangdata.com.cn/Thesis_Y1448087.aspx

Xiong, L., Huang, L. L., Shimo, S. P., Li, W. H., Yang, X. L., & Yan, P. (2016). Multi-residue method for the screening of benzimidazole and metabolite residues in the muscle and liver of sheep and cattle using HPLC/PDAD with DVB-NVP-SO_3Na for sample treatment. *Journal Citation Reports*, *79*(19–20), 1373–1380. doi:10.1007/s10337-016-3144-7

Yan, X. (2013). The research on enzyme-linked immunosorbent assay for multi-pesticides residue (Doctoral dissertation). Retrieved from http://d.g.wanfangdata.com.cn/Thesis_Y2528324.aspx

Yaneva, M., Ivanov, Y., & Godjevargova, T. (2017). Preparation of polyclonal antibodies with application for an organophosphorus pesticide immunoassay. *Analytical Letters*, *50*(8), 1307–1324. doi:10.1080/00032719.2016.1221417

Yaneva, M., Ivanov, Y., Todorov, N., & Godjevargova, T. (2017). Magnetic-nanoparticles-based fluorescent immunoassay for individual and simultaneous determination of dichlorvos and paraoxon in milk. *Food and Agricultural Immunology*, *11*, 1–16. doi:10.1080/09540105.2017.1368458

Yang, G. X. (2014). Study on the detection of polychlorinated biphenyls based on immuno-PCR bio-barcode assay (Doctoral dissertation). Retrieved from http://d.g.wanfangdata.com.cn/Thesis_D786332.aspx

Yang, G. X., Zhuang, H. S., Chen, H. Y., Ping, X. Y., & Bu, D. (2015). A gold nanoparticle based immunosorbent bio-barcode assay combined with real-time immuno-PCR for the detection of polychlorinated biphenyls. *Sensors and Actuators B: Chemical*, *214*, 152–158. doi:10.1016/j.snb.2015.02.128

Ye, B. C., Li, S. Y., Zuo, P., & Li, X. H. (2008). Simultaneous detection of sulfamethazine, streptomycin, and tylosin in milk by microplate-array based SMM–FIA. *Food Chemistry*, *106*(2), 797–803. doi:10.1016/j.foodchem.2007.06.006

Yin, X. G., Yin, L. M., & Xu, C. L. (2008). Research development of residue determination of organophosphorus pesticides based on multianalyte immunoassay. *Food Science*, *29*(10), 684–688.

Yu, W. J., Yu, F. Y., Undersander, D. J., & Chu, F. S. (1999). Immunoassays of selected mycotoxins in hay, silage and mixed feed. *Food and Agricultural Immunology*, *11*(4), 307–319. doi:10.1080/09540109999690

Yu, X. Z., Zhang, X. Y., Wang, Z. H., Jiang, H. Y., Lv, Z. Q., Shen, J. Z., … Wen, K. (2018). Universal simultaneous multiplex ELISA of small molecules in milk based on dual luciferases. *Analytica Chimica Acta*, *1001*, 125–133. doi:10.1016/j.aca.2017.11.038

Zhang, S. W., Lai, X. T., & Yang, G. W. (2013). Enzyme-linked fab' fragment based competitive immunoassay for ovalbumin in hot processed food. *Journal of Immunoassay and Immunochemistry*, *34*(4), 393–403. doi:10.1080/15321819.2012.755629

Zhao, L. X., Sun, L., & Chu, X. G. (2009). Chemiluminescence immunoassay. *TrAC Trends in Analytical Chemistry*, *28*(4), 404–415. doi:10.1016/j.trac.2008.12.006

Zhu, J. G. (2016). *Multi-component analysis for mycotoxins and pesticide residues in grain and oil products* (Doctoral dissertation). Retrieved from http://g.wanfangdata.com.cn/details/detail.do?_type=degree&id=D01016271.

Zou, R. B., Liu, Y., Wang, S. J., Zhang, Y., Guo, Y. R., & Zhu, G. N. (2017). Development and evaluation of chemiluminescence enzyme-linked immunoassay for residue detection of three organophosphorus pesticides. *Chinese Journal of Pesticide Science*, *19*(01), 37–45.