Electronic Supplementary Material (ESI) for Analytical Methods.

Supporting information

Quantitative and rapid detection of spinosad and spinetoram by gold nanoparticle-based immunostrip

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Captions:

Fig. S1 The LC-MS and ^{1}H NMR spectrum of hapten SPI-HS. (a) LC-MS, (b) ^{1}H NMR.

Fig. S2 MALDI-TOF MS spectra of three immunogen conjugates and BSA. BSA (a), SPI-HS-BSA 30 (b), SPI-HS-BSA 60 (c), SPI-HS-BSA 90 (d).

Fig. S3 The TEM image (a) and ultraviolet-visible absorption spectrum (b) of CG and CG-mAb.

Fig. S4 Optimization of basic buffer containing 5% (m/v) different additives (b), the volumes of 0.1M K_2CO_3 (b), and the amounts of mAb (b). The amounts of mAb: A, 12 μg/mL; B, 8 μg/mL. the volumes of 0.1M K_2CO_3: 1, 8 μl/mL; 2, 12 μl/mL; 3, 16 μl/mL. SPI=20 ng/mL; Et-SPI=20 ng/mL.

Fig. S5 The test pictures of LF-ICS for detection of SPI (a, c, e, and g) and Et-SPI (b, d, f, and h) in PBS (a and b), rice (c and d), tea (e and f), and onion (g and h).

Fig. S6 The specificity of the LF-ICS with spinosad (2), spinosyn A (3), spinosyn D (4), spinetoram (5), spinetoram J (6), and spinetoram L (7) at 10 ng/mL. 1= 0 ng/mL in PBS.

Fig. S7 The effect of dilution ratio (/n) on the sensitivity of the LF-ICS. (a) rice; (b) tea; (c) onion. N, negative sample; S, spiked sample.

Tab. S1 Antisera evaluation of mice from different antigens.

Tab. S2 Cross-reaction results of the mAbs.

Tab. S3 The T/C values and inhibition rates (IR) of the strips shown in Figure S5. N, negative sample; S, spiked sample.

Tab. S4 The summary of the references about the methods for determination of SPI and Et-SPI.

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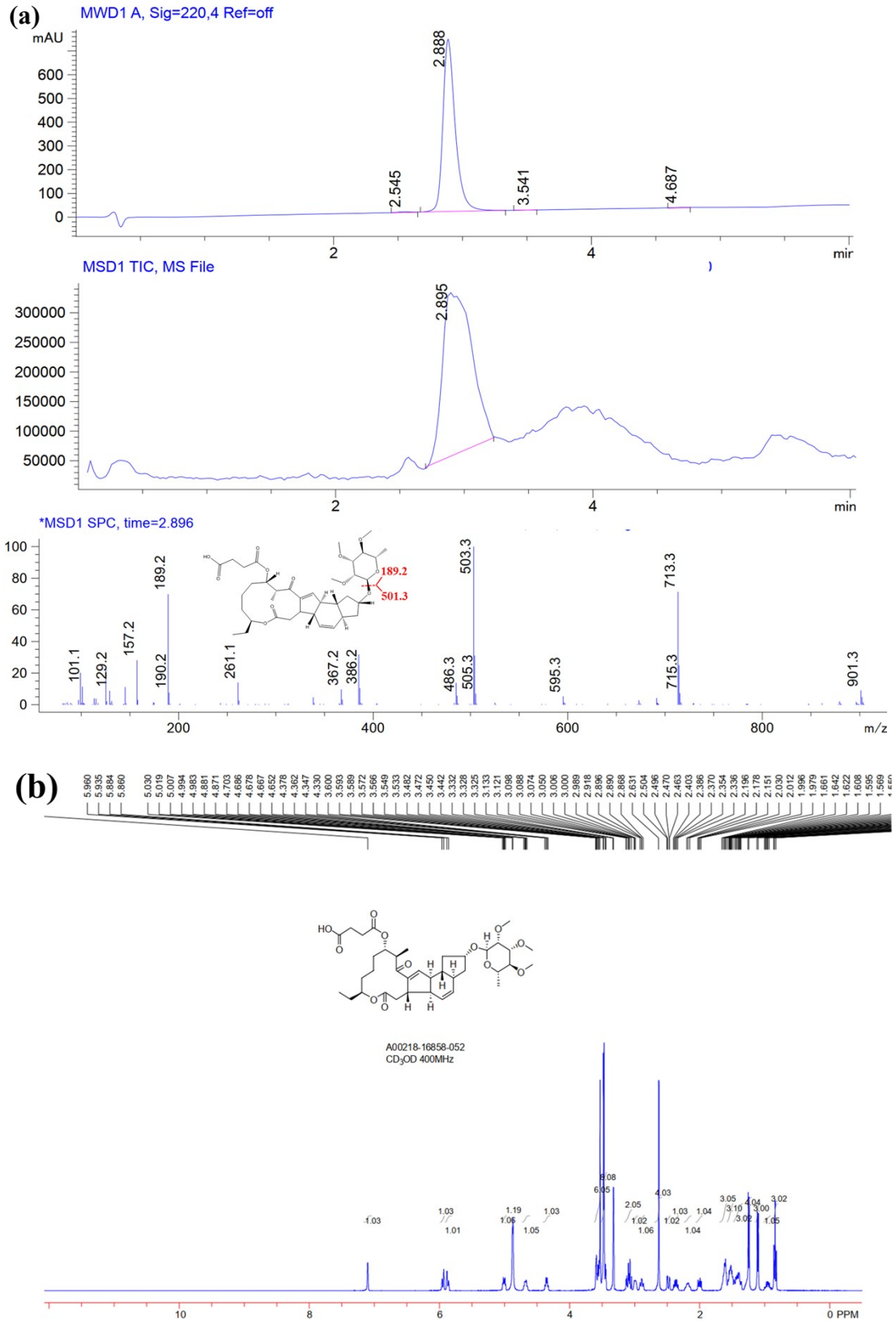
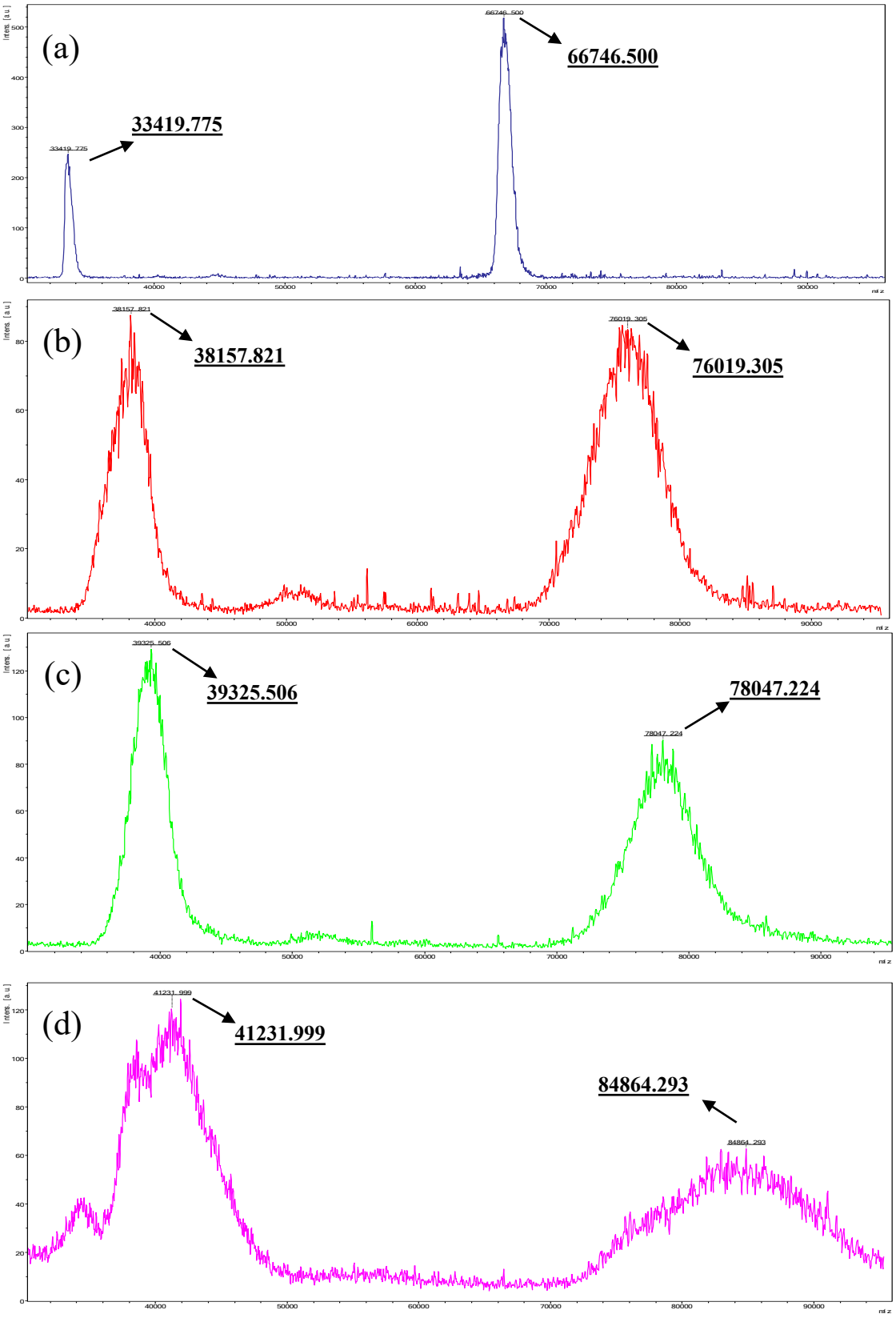


Fig. S2 MALDI-TOF MS spectra of three immunogen conjugates and BSA. BSA (a), SPI-HS-BSA 30 (b), SPI-HS-BSA 60 (c), SPI-HS-BSA 90 (d).



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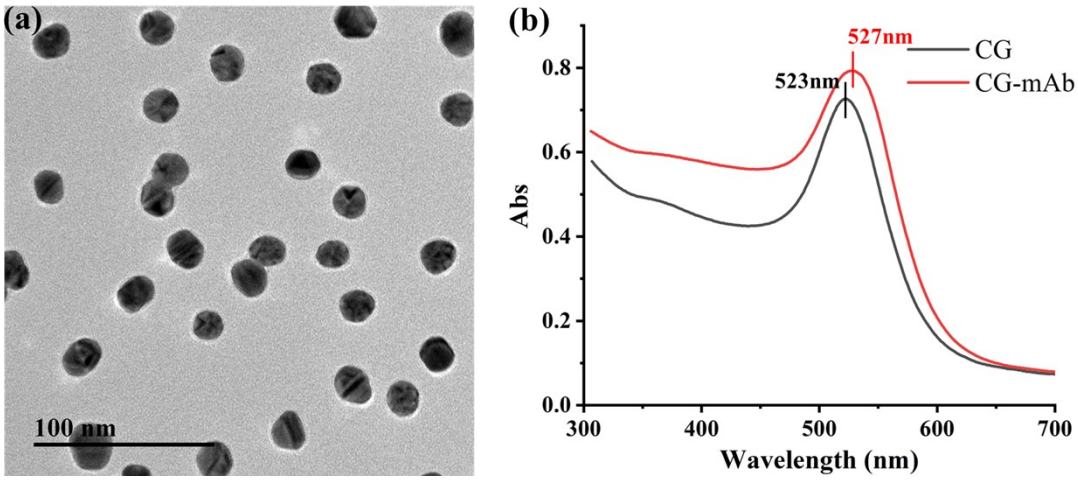

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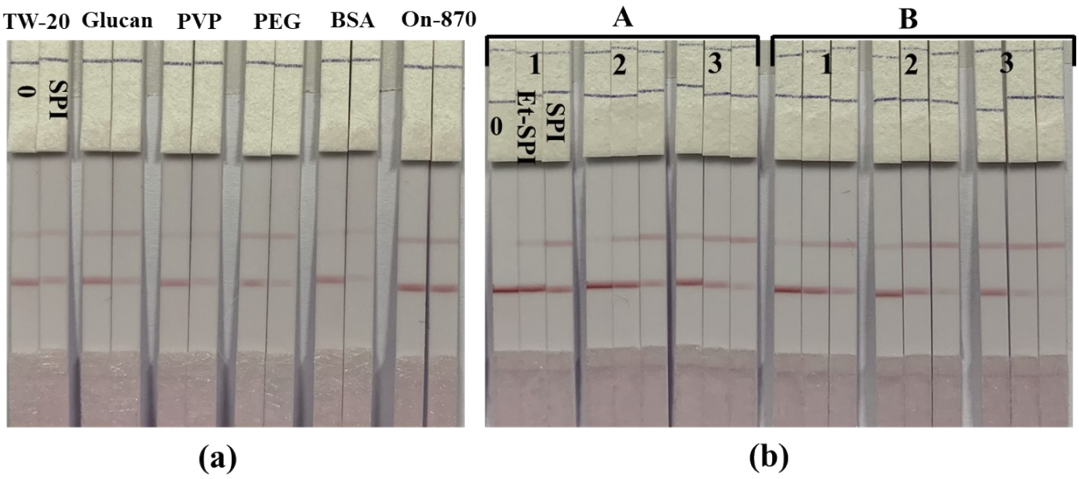

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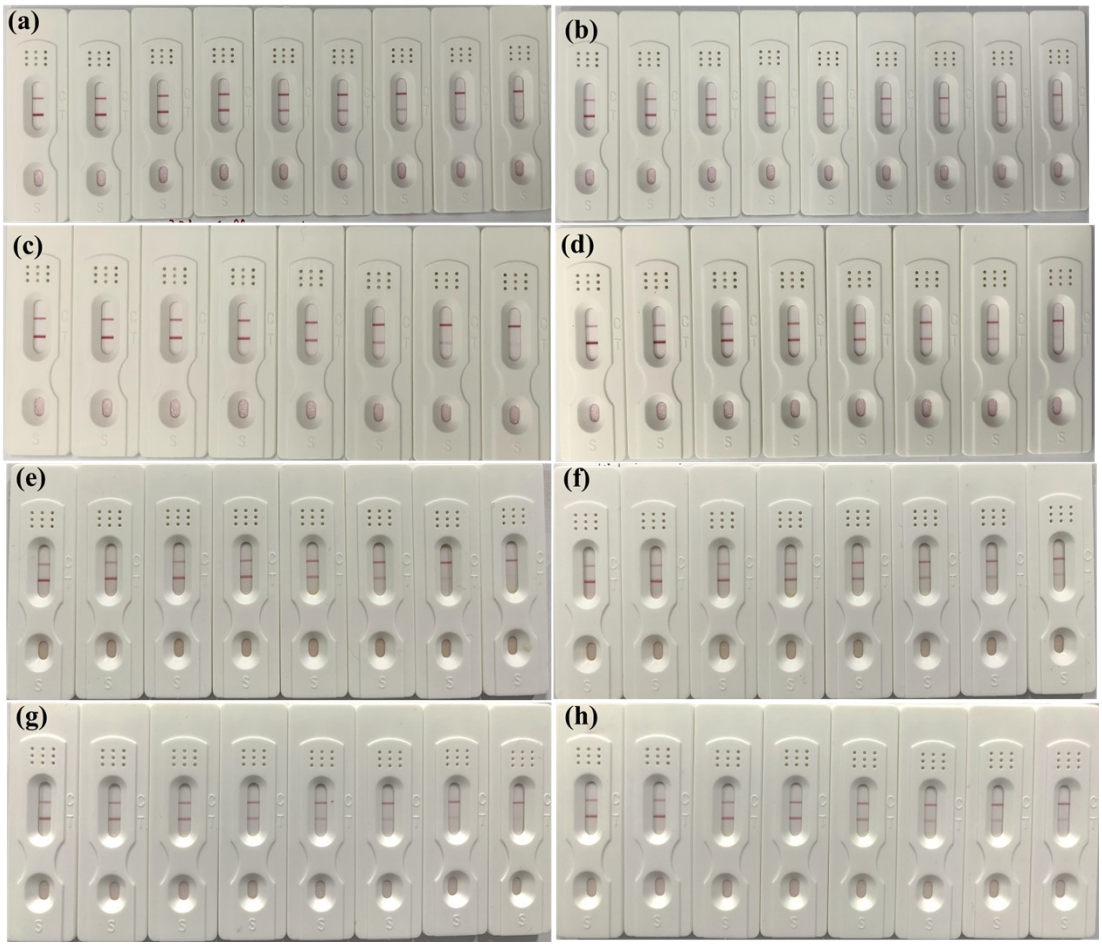

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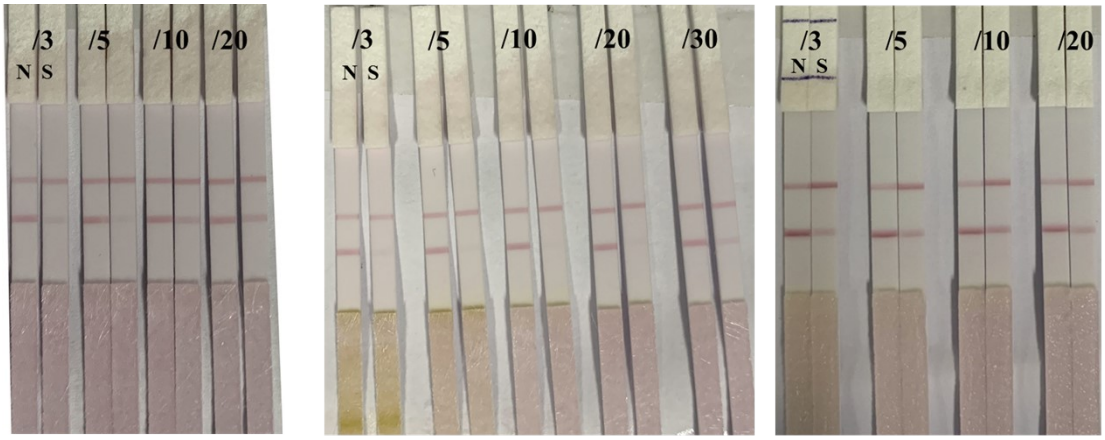
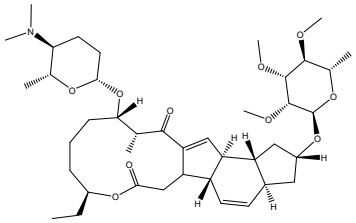
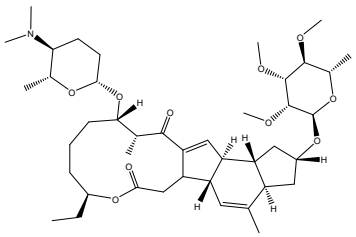
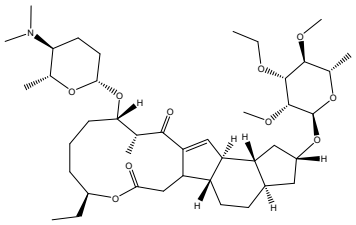
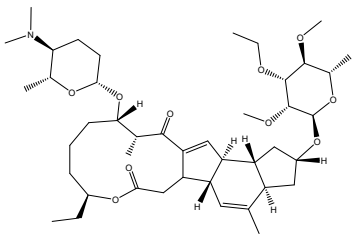


Tab. S1 Antisera evaluation of mice from different antigens.

Antigens	Standards (ng/mL)	Mice				
		1#	**2#**	3#	4#	5#
SPI-HS-BSA 30:1	0	1.894	1.625	1.668	1.412	1.55
	10(SPI)	1.225	0.875	1.022	0.686	1.119
	IR (%)	35%	46%	39%	51%	28%
	10(Et-SPI)	1.477	1.106	1.231	0.952	1.289
	IR (%)	22%	32%	26%	33%	17%
SPI-HS-BSA 60:1	0	1.762	1.698	1.852	1.996	1.663
	10(SPI)	0.941	0.835	0.595	0.863	0.967
	IR (%)	47%	51%	**68%**	57%	42%
	10(Et-SPI)	1.258	1.114	1.033	1.319	1.21
	IR (%)	29%	34%	**44%**	34%	27%
SPI-HS-BSA 90:1	0	1.601	2.104	1.686	1.913	1.779
	10(SPI)	0.648	1.39	0.965	0.891	0.925
	IR (%)	60%	34%	43%	53%	48%
	10(Et-SPI)	0.946	1.617	1.203	1.182	1.19
	IR (%)	41%	23%	29%	38%	33%

Inhibition ratio (IR) = (1-OD_{450}(10 ng/mL)/ OD_{450}(0 ng/mL)) * 100%.

Tab. S2 Cross-reaction results of the mAbs.

Chemicals	Structure	IC_{50} (ng/mL)	CR
Spinosad	A mixture with major component of spinosyn A and minor component of spinosyn D	0.68	100.0%
Spinosyn A		0.61	111.4%
Spinosyn D		0.89	76.4%
Spinetoram	A mixture with major component of spinetoram J and minor component of spinetoram L	1.89	36.0 %
Spinetoram J		1.64	41.5 %
Spinetoram L		1.81	37.6 %

Tab. S3 The T/C values and inhibition rates (IR) of the strips shown in Figure S5. N, negative sample; S, spiked sample.

Samples	Repetition		/n									
			/3		/5		/10		/20		/30	
			T/C	IR (%)	T/C	IR (%)	T/C	IR (%)	T/C	IR (%)	T/C	IR (%)
Rice	1	N	1.35	80.0	1.22	86.9	1.16	54.3	0.98	34.7		
		S	0.27		0.16		0.53		0.64			
	2	N	1.31	75.6	1.15	88.7	1.19	59.7	1.05	41.9		
		S	0.32		0.13		0.48		0.61			
	3	N	1.28	82.0	1.28	91.4	1.12	58.0	0.96	30.2		
		S	0.23		0.11		0.47		0.67			
Tea	1	N	1.42	84.5	1.31	87.8	1.33	92.5	1.25	90.4	1.13	63.7
		S	0.22		0.16		0.1		0.12		0.41	
	2	N	1.4	81.4	1.36	84.6	1.4	93.6	1.31	92.4	1.22	59.8
		S	0.26		0.21		0.09		0.1		0.49	
	3	N	1.35	80.0	1.39	86.3	1.31	90.8	1.29	92.2	1.17	67.5
		S	0.27		0.19		0.12		0.1		0.38	
Onion	1	N	2.31	87.0	2.12	83.0	2.2	81.4	2.08	78.4		
		S	0.3		0.36		0.41		0.45			
	2	N	2.4	86.7	2.15	78.6	2.08	76.4	1.97	78.7		
		S	0.32		0.46		0.49		0.42			
	3	N	2.38	89.5	2.27	81.9	2.04	77.0	2.02	75.7		
		S	0.25		0.41		0.47		0.49			

Tab. S4 The summary of the references about the methods for determination of SPI and Et-SPI.

References	Years	Methods	Samples	Drugs (cross-reaction, %)	[a]LOD (ng/g or ng/mL)	Detection time (min)	recovery values (%)	[b]CV([c]RSD, %)
[1]	2019	ic-Elisa	Milk fruits vegetables	spinosyn A (100) spinosyn B (80.4) spinosyn D (75.7)	0.63	>60	78.1-103.2	3.3-7.3
[2]	1999	Fluorescent Excitation Transfer	water	spinosyn A	0.01	20-30	96-120	
[3]	2000	magnetic particle-based immunoassay (IA) test kit	Water Sediment Crops animal tissues	spinosyns A (100) and D (15) spinosyn B (48), spinosyn K (32) and N-demethylspinosyn D (16)	0.1 (water) 50 (sediment) 10 (crops and animal tissues)		77-112	
[4]	2021	UPLC-MS/MS	tea	Spinetoram J Spinetoram L N-demethyl- Spinetoram J N-formyl- Spinetoram J	20 (Spinetoram J, Spinetoram L N-demethyl-Spinetoram J) 50 (N-formyl-Spinetoram J)	>30	72.4-105.7	0.9-16.1
[5]	2012	HPLC	Tomato	spinetoram	10		88.5-97.5	5.3-12.2
[6]	2015	LC–MS/MS	soil, rice straw, paddy water, husk and brown rice	Spinetoram J Spinetoram L	1 (husk, rice and straw); 0.5 (soil and brown rice); 0.25 (paddy water)		72.7-100.3	1.9-8.1

[a]LOD, low of detection; [b]CV, coefficient variable; [c]RSD, relative standard deviation.

References:

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