**A sensitive and specific monoclonal antibody based enzyme-linked immunosorbent assay for the rapid detection of pretilachlor in environment**

Linwei Zhang1†,, Xiaoyang Yin1†, Hongfei Yang1, Hao Wen1, Shiyun Han1, Xiaoming Pan1, Huaming Li1, Liangni Qin1, Jiaxu Xiao1, Long Li1, Yu Si1, Jixiang Liang1, Dapeng Peng1, 2, 3, 4\*

National Reference Laboratory of Veterinary Drug Residues (HZAU), Huazhong Agricultural University, Wuhan, Hubei, China.

Fig. S1 PR-SC mass spectrometry (A: [M+Na]+ and B: [M+H]+)



Table S Titer and inhibition rate of serum PY and PR antibody in mice

|  |  |  |
| --- | --- | --- |
| Mice No. | Serum dilution factor（1：X） | Inhibition rate (B/B0)a |
| PRA1 | 4000 | 32% |
| PRA2 | 4000 | 31% |
| PRA3 | 2000 | 48% |
| PRA4 | 4000 | 58% |

a The above inhibition rates were calculated at a concentration of 250 μg/L PR.

Table S2 PR cell antibody titers and inhibition rates

|  |  |  |
| --- | --- | --- |
| Cell Line No. | Antibody titer（1：X） | Inhibition rate a |
| PR-114 | 80 | 42% |
| PR-112 | 40 | 45% |
| PR-121 | 80 | 53% |
| PR-142 | 80 | 53% |

a The above inhibition rates were calculated at a concentration of 50 μg/L PR.

Table S3 11 PR/114 antibody array titration

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Antibody concentration（μg/mL） | Coated antigen concentration（μg/mL） | | | | | |
| 1 | 0.5 | 0.25 | 0.125 | 0.0625 | 0.03125 |
| 2 | 3.132 | 3.091 | 2.888 | 2.642 | 2.302 | 1.887 |
| 1 | 2.969 | 2.684 | 2.459 | 2.212 | 1.905 | 1.487 |
| 0.5 | 2.386 | 2.294 | 1.966 | 1.851 | 1.000 | 0.963 |
| 0.25 | 1.714 | 1.561 | 1.422 | 1.241 | 1.011 | 0.695 |
| 0.125 | 1.150 | 1.097 | 1.086 | 0.944 | 0.838 | 0.666 |
| 0.0625 | 0.863 | 0.881 | 0.806 | 0.788 | 0.701 | 0.599 |

Table S4 Screening for PR/114 antibody and PR-SC-OVA concentrations

|  |  |  |
| --- | --- | --- |
| Coated antigen concentration（μg/mL） | Antibody concentration（μg/mL） | IC50（μg/L） |
| 0.25 | 0.5 | 47.06 |
| 0.125 | 0.5 | 35.41 |
| 0.0625 | 1 | 31.47 |

Table S5 CV of PR/114 antibody ic-ELISA standard curve

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| PR(µg/L) | measured value (C±SD, µg/L) | Coefficient of Variation (%) (CV%, n=5) | measured value  (C±SD, µg/L) | Coefficient of Variation (%) (CV%, n=25) |
| 6.25 | 5.92±0.78 | 13.1 | 6.18±0.26 | 4.2 |
| 6.03±0.82 | 13.6 |
| 6.28±0.97 | 15.4 |
| 6.02±0.78 | 12.9 |
| 6.65±0.74 | 11.1 |
| 12.50 | 13.52±2.07 | 15.3 | 13.35±1.04 | 7.7 |
| 12.02±1.87 | 15.5 |
| 12.34±1.94 | 15.7 |
| 14.07±1.06 | 7.5 |
| 14.79±1.84 | 12.4 |
| 25.00 | 27.52±2.04 | 7.4 | 27.34±0.50 | 1.8 |
| 27.98±1.94 | 6.9 |
| 27.40±1.60 | 5.8 |
| 27.34±2.01 | 7.3 |
| 26.45±1.01 | 3.8 |
| 50.00 | 56.21±4.25 | 7.5 | 52.69±3.13 | 5.9 |
| 52.05±2.22 | 4.2 |
| 56.39±4.39 | 7.7 |
| 50.01±5.16 | 10.3 |
| 48.78±1.07 | 2.1 |
| 100.00 | 89.94±6.12 | 6.8 | 87.85±1.21 | 1.3 |
| 87.33±5.42 | 6.2 |
| 88.38±6.38 | 7.2 |
| 86.56±2.93 | 3.3 |
| 87.01±5.57 | 6.4 |



