

## **Development of an enantioselective method by liquid chromatography to monitor 3,4-methylenedioxypropylvalerone in culture media from ecotoxicity assays**

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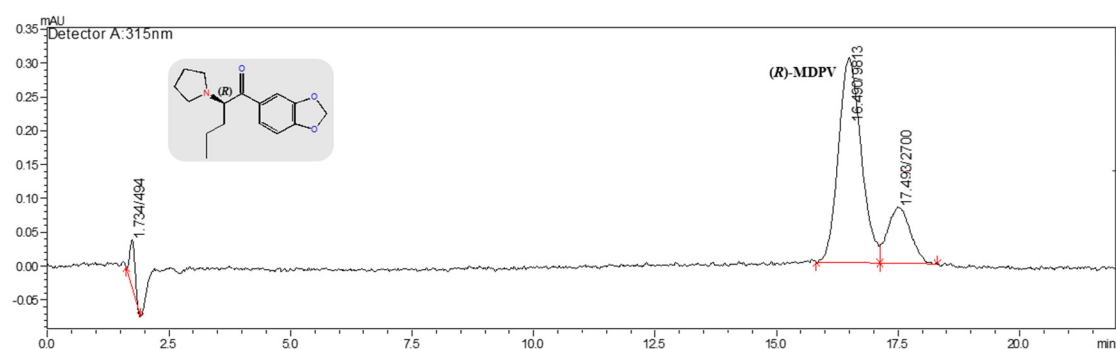
**Figure S1a.** Chromatogram of the enantioseparation of  $1.00 \mu\text{g mL}^{-1}$  (*R*)-MDPV (in UPW) in the analytical Lux<sup>®</sup> 3  $\mu\text{m}$  - Cellulose-2 ( $150 \times 2.0 \text{ mm I.D.}$ ) column by LC-UV/Vis under reversed elution mode in isocratic mode. LC conditions: mobile phase, 20 mM  $\text{NH}_4\text{OAc}$  in UPW (pH 8.5) and ACN (70:30, v/v); flow rate of  $0.3 \text{ mL min}^{-1}$ ; injection volume of  $5 \mu\text{L}$ ;  $\lambda_{\text{max}}$  of 315 nm; column oven temperature set at  $25^\circ\text{C}$ .

**Figure S1b.** Chromatogram of the enantioseparation of  $1.00 \mu\text{g mL}^{-1}$  (*S*)-MDPV (in UPW) in the analytical Lux<sup>®</sup> 3  $\mu\text{m}$  - Cellulose-2 ( $150 \times 2.0 \text{ mm I.D.}$ ) column by LC-UV/Vis under reversed elution mode in isocratic mode. LC conditions: mobile phase, 20 mM  $\text{NH}_4\text{OAc}$  in UPW (pH 8.5) and ACN (70:30, v/v); flow rate of  $0.3 \text{ mL min}^{-1}$ ; injection volume of  $5 \mu\text{L}$ ;  $\lambda_{\text{max}}$  of 315 nm; column oven temperature set at  $25^\circ\text{C}$ .

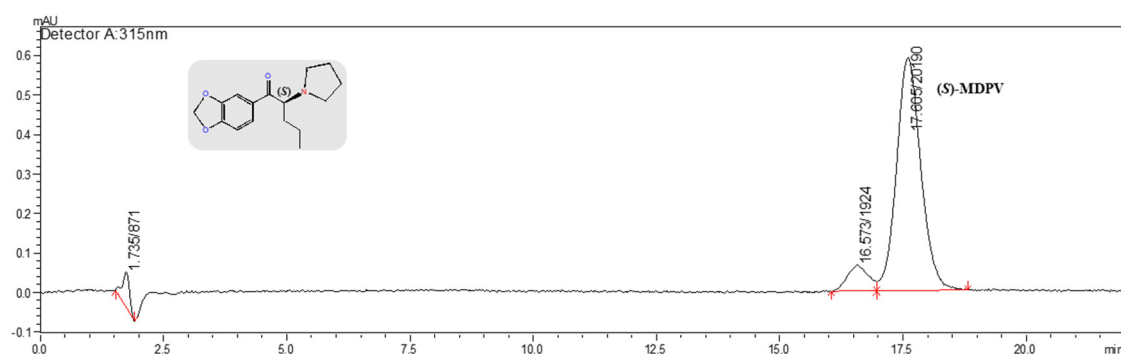
**Figure S2.** Chromatograms of the separation of  $.10 \mu\text{g mL}^{-1}$  (*S*)-MDPV (in UPW at different pH) in the analytical Lux<sup>®</sup> 3  $\mu\text{m}$  - Cellulose-2 ( $150 \times 2.0 \text{ mm I.D.}$ ) column by LC-UV/Vis under reversed elution mode in isocratic mode. LC conditions: mobile phase, 20 mM  $\text{NH}_4\text{OAc}$  and ACN (70:30, v/v, pH 8.5); flow rate of  $0.3 \text{ mL min}^{-1}$ ; injection volume of  $5 \mu\text{L}$ ;  $\lambda_{\text{max}}$  of 315 nm; column oven temperature set at  $35^\circ\text{C}$ . (a) time zero; (b) 24 h after.

**Figure S3.** Total ion chromatograms (black) and selected reaction monitoring ( $m/z$  276.00 > 126.15 (pink);  $m/z$  276.00 > 135.05 (blue)) of the MDPV enantiomers in the analytical Daicel<sup>®</sup> 3  $\mu\text{m}$  - CHIRALPAK<sup>®</sup> IF-3 ( $150 \times 2.1 \text{ mm I.D.}$ ) column by LC-MS/MS under reversed elution mode in isocratic mode. LC conditions: 5 mM  $\text{NH}_4\text{HCO}_3$  in UPW (pH 8.8) with ACN (10:90, v/v) as mobile phase; flow rate of  $0.3 \text{ mL min}^{-1}$ ; injection volume of  $5 \mu\text{L}$ ; column oven temperature set at  $30^\circ\text{C}$ . (a) (*R,S*)-MDPV at  $1000 \mu\text{g L}^{-1}$  in UPW (sample matrix after SPE); (b) (*R,S*)-MDPV at  $1000 \mu\text{g L}^{-1}$  in UPW (standard).

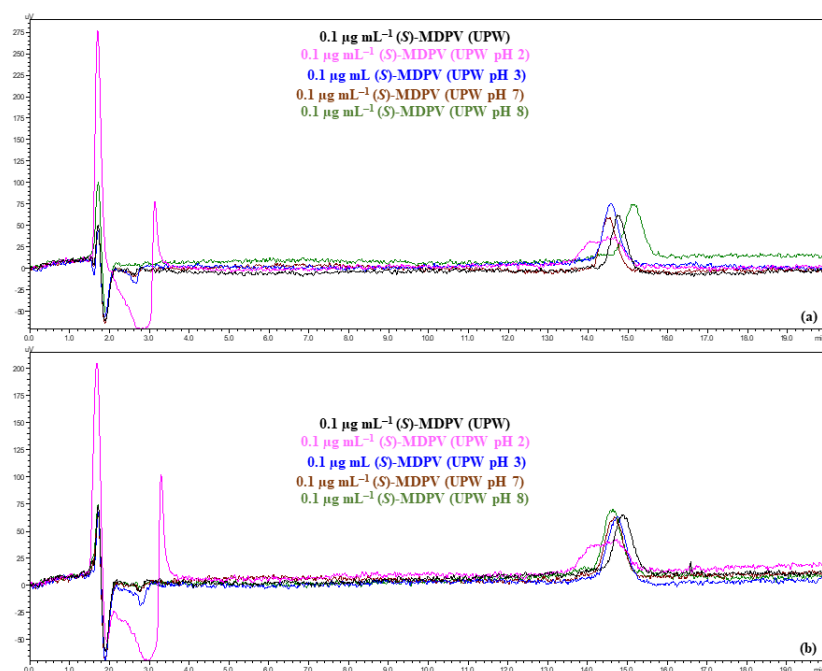
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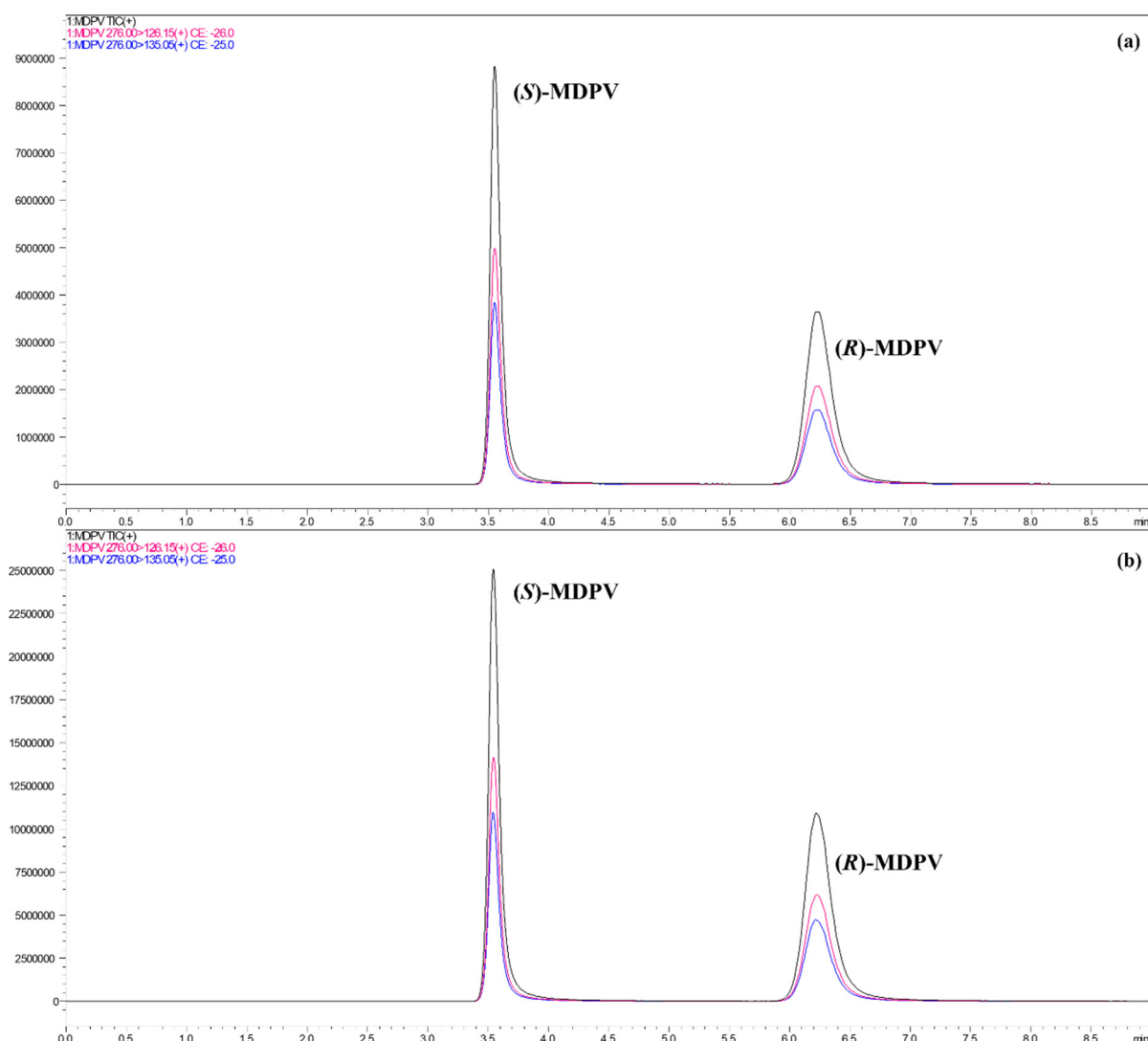
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Sample (Average)	e.r. of ( <i>S</i> )-MDPV	e.r. of ( <i>R</i> )-MDPV
( <i>R,S</i> )-MDPV <sup>(a)</sup>	49.5	50.5

( <i>S</i> )-MDPV <sup>(a)</sup>	96.4	3.6
( <i>R</i> )-MDPV <sup>(a)</sup>	15.6	84.4
( <i>R,S</i> )-MDPV <sup>(b)</sup> C1	48.9	51.1
( <i>R,S</i> )-MDPV <sup>(b)</sup> C2	48.2	51.8
( <i>R,S</i> )-MDPV <sup>(b)</sup> C3	48.5	51.5
( <i>R,S</i> )-MDPV <sup>(c)</sup> C1	49.5	50.5
( <i>R,S</i> )-MDPV <sup>(c)</sup> C2	49.2	50.8
( <i>R,S</i> )-MDPV <sup>(c)</sup> C3	49.7	50.3
( <i>S</i> )-MDPV <sup>(b)</sup> C2	75.0	25.0
( <i>S</i> )-MDPV <sup>(b)</sup> C3	81.5	18.5
( <i>S</i> )-MDPV <sup>(c)</sup> C2	75.5	24.5
( <i>S</i> )-MDPV <sup>(c)</sup> C3	80.1	19.9
( <i>R</i> )-MDPV <sup>(b)</sup> C1	28.0	72.0
( <i>R</i> )-MDPV <sup>(b)</sup> C2	25.8	74.2
( <i>R</i> )-MDPV <sup>(b)</sup> C3	30.7	69.3
( <i>R</i> )-MDPV <sup>(c)</sup> C1	31.7	68.3
( <i>R</i> )-MDPV <sup>(c)</sup> C2	30.4	69.6
( <i>R</i> )-MDPV <sup>(c)</sup> C3	23.3	76.7

**(a)** MDPV Standards at 1.00 µg L<sup>-1</sup> (in UPW); **(b)** culture medium samples spiked at 1.00 µg L<sup>-1</sup> (in UPW); **(c)** MDPV samples at 1.00 µg L<sup>-1</sup> (in UPW); **(C1)** Sample Collection 1; **(C2)** Sample Collection 2; **(C3)** Sample Collection 3.