



## Neonicotinoid binding, toxicity and expression of nicotinic acetylcholine receptor subunits in the aphid *Acyrtosiphon pisum*.

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Résumé en anglais

Neonicotinoid insecticides act on nicotinic acetylcholine receptor and are particularly effective against sucking pests. They are widely used in crops protection to fight against aphids, which cause severe damage. In the present study we evaluated the susceptibility of the pea aphid *Acyrtosiphon pisum* to the commonly used neonicotinoid insecticides imidacloprid (IMI), thiamethoxam (TMX) and clothianidin (CLT). Binding studies on aphid membrane preparations revealed the existence of high and low-affinity binding sites for [3H]-IMI (Kd of  $0.16 \pm 0.04$  nM and  $41.7 \pm 5.9$  nM) and for the nicotinic antagonist [125I]- $\alpha$ -bungarotoxin (Kd of  $0.008 \pm 0.002$  nM and  $1.135 \pm 0.213$  nM). Competitive binding experiments demonstrated that TMX displayed a higher affinity than IMI for [125I]- $\alpha$ -bungarotoxin binding sites while CLT affinity was similar for both [125I]- $\alpha$ -bungarotoxin and [3H]-IMI binding sites. Interestingly, toxicological studies revealed that at 48 h, IMI (LC50 = 0.038  $\mu$ g/ml) and TMX (LC50 = 0.034  $\mu$ g/ml) were more toxic than CLT (LC50 = 0.118  $\mu$ g/ml). The effect of TMX could be associated to its metabolite CLT as demonstrated by HPLC/MS analysis. In addition, we found that aphid larvae treated either with IMI, TMX or CLT showed a strong variation of nAChR subunit expression. Using semi-quantitative PCR experiments, we detected for all insecticides an increase of Apisum $\alpha$ 10 and Apisum $\beta$ 1 expressions levels, whereas Apisum $\beta$ 2 expression decreased. Moreover, some other receptor subunits seemed to be differently regulated according to the insecticide used. Finally, we also demonstrated that nAChR subunit expression differed during pea aphid development. Altogether these results highlight species specificity that should be taken into account in pest management strategies.

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## Liens

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