

Sub-lethal effects at stake: Does the acaricide Coumaphos and fungicide Folpet affect the hypopharyngeal glands size?

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Introduction

The development of hypopharyngeal glands (HPG), producing royal jelly (RJ) to feed brood, is an established marker to assess sub-lethal effects of pesticides where for example the size of the acini can be measured. Its size depends of different natural factors: the age of the bee and the type of task performed. According to the EFSA bee guidance document

(2013)¹ observations of effects on HPGs development are recommended to cover potential effects on brood care. Sublethal effects on the commonly and widely used varroacide Coumaphos and the fungicide Folpet in agriculture are currently unknown. Therefore, we assessed the acini size and head weight of newly emerged bees fed with field realistic and non-realistic concentrations.

Method

Beebread spiked with the treatments below was provided to newly emerged bees in "Liebefeld cages" either in presence or absence of young honeybee larva brood. 50 bees per cage were kept in a climate chamber. Coumaphos was diluted in acetone (solvent control 1), Folpet in chloroform (solvent control 2) and untreated pollen was used as negative control (Table 1). The acini diameters of the HPG and heads weight of the tested bees were measured after 10 days of chronic exposure to the treated pollen patties (Fig. 1).

		Treatment		
		Coumaphos		Folpet
		1mg/g (unreal.)	2mg/kg (real.)	2mg/g (unreal.)
Brood	+	C=3 ; B=50	C=3 ; B=50	C=3 ; B=50
	-	C=3 ; B=50	C=3 ; B=50	C=3 ; B=50

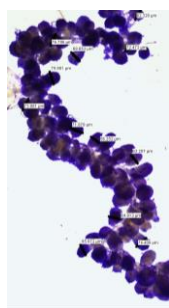


Table 1: Tested treatments C= Nr. cages; B= Nr. bees

Fig. 1: Acini measurements on HPG

Results

First, the acini size is correlated with the heads weight (Pearson Correlation=0.341, p-value=0.000). Moreover, the

acini size is significantly bigger in presence of brood (p-value=0.000) (Fig. 2).

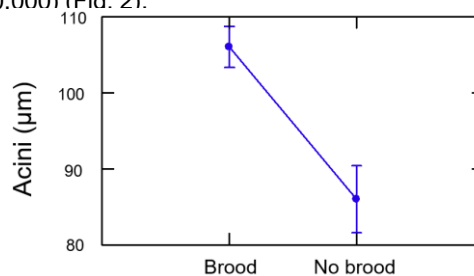


Fig. 2: Least squares means acini size per experiment

The quantity of pollen consumption by the bees was influenced by treatments and treatment concentrations. We observed a repellent effect of Coumaphos, which was considered in our general linear model (Fig. 3). The experiment without brood is harder to interpret, as the glands have not been activated since no brood was present. For the experiment with brood, we observed that Folpet has no effect on the acini size (p-value=0.9046). In contrast, Coumaphos seems to hypertrophy the acini size at 1 mg/g and 2mg/kg when compared to the solvent control 1 (p-value 1mg/g=0.0004; 2mg/kg= 0.0046).

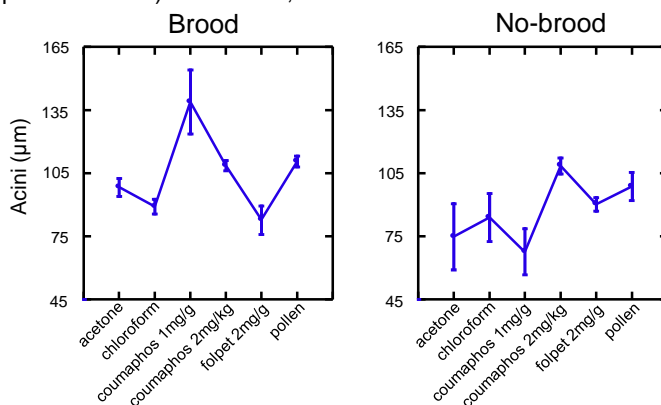


Fig. 3: Least squares means acini size for each treatment with and without brood presence

Conclusions

Our results demonstrate the need of additional research to determine an appropriate method for accurate and comparable results of sub-lethal effects on HPG and its functionality. Brood presence for HPG testing is crucial to cover both maturity

growth and functionality growth (triggered by brood presence and RJ production) of the HPG. Therefore, to assess sublethal effects of pesticides on HPG, brood presence to activate the royal jelly production should be considered and included in the test method (design) under laboratory conditions.