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Feeding experiments with black soldier fly larvae and yellow mealworm – the effect of fasting on animal DNA and pesticide contents

Advisory report from DCA – Danish Centre for Food and Agriculture

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Data sheet

Title:	Feeding experiments with black soldier fly larvae and yellow mealworm – investigating the transfer of pesticides from substrate to larvae
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Reviewer:	Senior researcher Martin Tang Sørensen, Department of Animal and Veterinary Sciences, AU.
Quality assurance, DCA:	Special consultant Stine Cecilie Mangaard Sarraf, DCA Center unit
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External contributions:	Yes. Technical University of Denmark (DTU) and the Danish Veterinary and Food Administration (DVFA) have performed the chemical analyzes.
Comments to the answer:	The report presents results which at the delivery have not been under external peer review, nor are published elsewhere. In case of subsequent publishing in journals with external peer review, changes may appear.
This report presents a Danish summary of an unsubmitted manuscript to facilitate later publication in a peer reviewed journal. The manuscript (Appendix to this paper) can be distributed as a hard copy upon request.	
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Preface

Using insects for food and feed is associated with several challenges regarding food and feed safety. Therefore, it has become a priority to revise current legislation to facilitate this new industry without risking health consequences for consumers, e.g., if used as feed for production animals or for human consumption. In 2019, the Danish Ministry of Food, Agriculture and Fisheries (MFVM) requested Aarhus University (AU), University of Copenhagen (KU), and the Technical University of Denmark (DTU) to frame the research needed to implement a sustainable industrial production of insects in Denmark. Current legislation prohibits the feeding of insects with several biomasses which would otherwise support a green transition in food and feed production. It was indicated that more knowledge on former foodstuffs and household waste as insect feed is needed before being able to expand the range of usable biomasses, including biomasses potentially containing meat and residues of pesticides, with a goal to improve sustainability in feed production without introducing risks to food and feed safety. Research-based knowledge may support decision making and facilitate progress in European Union (EU) legislations. Here, we report results on the effect of introducing a fasting period before insects are collected for processing. Similar experiments were performed on larvae of the black soldier fly (*Hermetia illucens*), and on yellow mealworm (larvae of the beetle *Tenebrio molitor*), which are the two most important insect species commercially produced in Denmark. Meat or pesticides were added to insect substrates, and the contents of animal DNA or pesticide residues in the insect larvae after different lengths of fasting before collection were analysed. The experiments were performed at AU Viborg. Samples were then shipped to DTU where pesticide contents were determined, and to the Danish Veterinary and Food Administration (DVFM) where DNA analyses were performed.

Dansk sammendrag

Genanvendelse af madaffald fra køkkener til insektproduktion som foderemne er under den nuværende lovgivning ikke tilladt, da fødevaresikkerheden i forbindelse hermed ikke er grundigt undersøgt. Lovgivningen om brug af køkken- og madaffald til foder i insektproduktion skal løbende opdateres efterhånden som risikofaktorer bliver afdækket. Opdateringen vil afhænge af, om skadelige eller potentielt skadelige stoffer overføres til insekterne og dermed videregives til husdyr, når insekterne bruges i foder. Vi har i foregående undersøgelser vist, at udvalgte pesticider ophobes i insekter (larver af sorte soldaterfluer (*Hermetia illucens*) og melorm (*Tenebrio molitor*)), når de findes i deres vækstsubstrat. Vi har derimod fået forskellige resultater med hensyn til insekternes indhold af DNA fra kød. Dette kunne skyldes, om vi inkluderede en fasteperiode eller ej inden indsamling, eller det kunne skyldes forskelle imellem de to undersøgte arter.

I denne rapport har vi undersøgt, om en fasteperiode efter vækstfasen kan eliminere indholdet af DNA fra kød og af pesticidrester hos larver af sorte soldaterfluer og melorm, efter larverne er udviklet på substrater tilsat kød eller pesticider. Larverne fik først en vækstperiode, hvor enten kød (ko, gris og kylling) eller pesticider (boscalid, etofenprox og fluopyram) var tilsat vækstsubstratet for hver af de to arter. Vi gav derefter larverne 0, 12, 24, 48 eller 72 timers faste, hvorefter de blev indsamlet og analyseret for indhold af DNA fra de tre typer kød eller for indhold af de tre pesticider. Tre gentagelser blev lavet per substrat og fasteperiode for hver insektart. Vækst og efterfølgende faste foregik i et klimakontrolleret rum ved 26 - 28 °C og 45 - 55 % relativ luftfugtighed.

Undersøgelserne viste store forskelle i indholdet af DNA og pesticider imellem de to insektarter, og i effekten af faste på indholdet. DNA fra kød blev ikke genfundet i soldaterfluelarverne, ikke engang når de blev indsamlet direkte fra substratet uden faste. Modsat blev DNA fra kød genfundet hos alle grupper af melorm, også dem der havde fastet i 72 timer. De tre pesticider blev genfundet i alle grupper af soldaterfluerne og melormene, men blev generelt kraftigt reduceret indenfor de første 12 timers faste. Dog var reduktionen meget hurtigere hos soldaterfluelarver end hos melorm. Et ofenprox (insekticid) blev hos begge arter reduceret mere gradvist end boscalid (fungicid) og fluopyram (fungicid og nematocid).

Larvernes overlevelse var ikke påvirket af pesticider hverken hos soldaterfluer eller melorm. Til gengæld var væksten hos melorm reduceret på medie med kød, mens kød nærmere påvirkede soldaterfluerne vækst positivt end negativt, dog ikke med signifikant forskel. Vi fandt ingen negativ effekt af pesticider på væksten hos de to insektarter.

Det kan konkluderes, at soldaterfluelarver kan produceres med kød i substratet, uden at DNA fra kød findes i larverne, selv uden en fasteperiode. Til gengæld eliminerer 72 timers faste ikke DNA fra kød hos melorm. Pesticider bliver kraftigt reduceret under faste hos begge arter, dog hurtigst hos soldaterfluelarver. Ingen af pesticiderne blev fuldstændigt elimineret indenfor 72 timer, med lavest reduktion af etofenprox.