

Insect biomass quality and safety: basic concepts, recent issues, and future challenges

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Outline









Challenges and future prospective



News 3 Jul 2017 6151 views 4 comments

July 1: A milestone for insect protein



As of July 1 2017, insect protein will be allowed to be used in aquafeed. The insect sector is thrilled that this milestone to start using insect protein is now official.

The EU Regulation 2017/893 now permits the use of insect proteins as fish feed, derived from the following insect species:

- Black Soldier Fly (Hermetia illucens) and Common Housefly (Musca domestica)
- Yellow Mealworm (Tenebrio molitor) and Lesser Mealworm (Alphitobius diaperinus)
- House cricket (Acheta domesticus), Banded cricket (Gryllodes sigillatus) and Field Cricket (Gryllus assimilis).

The use of insect protein for other livestock species is not allowed yet.

In the wake of this legislative reform, IPIFF President Antoine Hubert reacted: "We are particularly pleased with the opening of this legislation, which constitutes a major milestone towards the development of the European insect production sector".

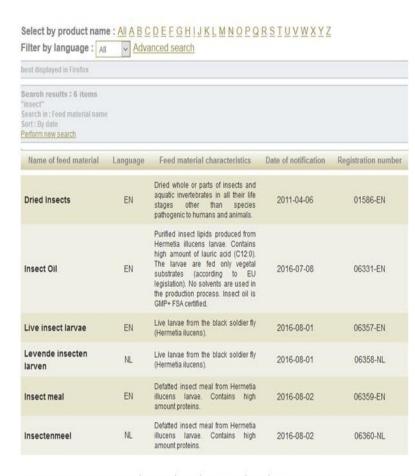
"We also trust that this legislation will bring new opportunities for the European aquaculture sector, as insects should represent a promising source of proteins for farmed fish in the near future: as being a natural component of the diets of carnivorous fish, whilst combining high proteins levels - between 55% and 75% - and ex-





Insects as feed

- Reg.(EU) No 2017/1017 which amended Reg. 68/2013 on the EU Catalogue of feed materials introduces revised descriptions explicitly referring to processed animal proteins and fats from insects (see 9.4.1 'processed animal protein' & 9.2.1 'animal fat', whose descriptions now refer to invertebrates).
- Processed animal protein more limitations
- Animal fat less limitations



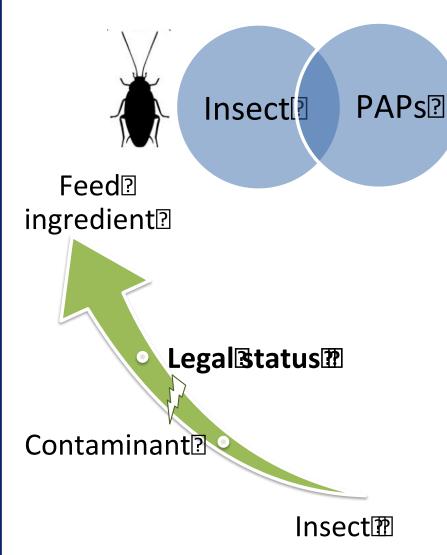


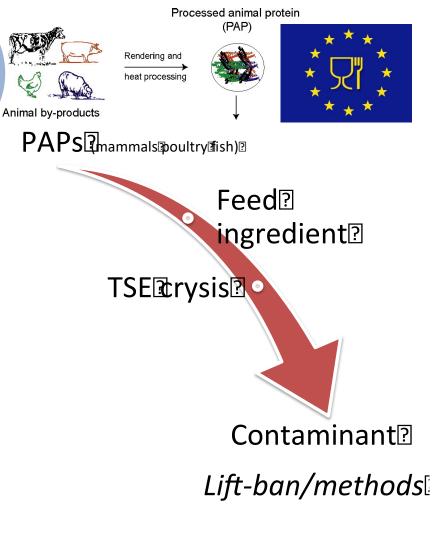


Insects as feed

- the EU 'feed ban rules' contained in the so called 'TSE Regulation' (i.e. Reg. 999/2001) so far prohibited the use of PAP to be used in feed for farmed animals, including for fish.
- Reg. 2017/893 partially uplifts the feed ban rules regarding the use of <u>insect processed animal proteins</u> (PAPs) for aquaculture animals.
 - the text introduces a specific section for insects & insect products (Annex IV, section F of Regulation 999/2001)



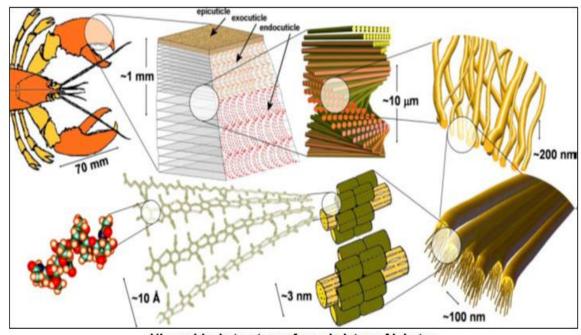








- Several similarities
 with Marine
 arthropods (Shrimp,
 krill, contained in
 fish meal)
- Authorized only for farmed fish



Hierarchical structure of exoskeleton of lobster

Exoskeleton features!

Modified from: Raabe et al. | Acta Matarialia 53 (2005) 4281



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	Ruminants	Unweaned ruminants	Non ruminants	Fish	Pets and fur animals
Ruminant PAP (included ruminant blood meal)	NA	NA	NA	NA	А
Non ruminant PAP	NA	NA	NA	А	A
Insect PAP	NA	NA	NA	А	А
Fish meal	NA	A	А	А	Α
Ruminant collagen and gelatine	NA	NA	NA	NA	А
Non ruminant collagen and gelatine	А	А	Α	А	Α
Ruminant blood products	NA	NA	NA	NA	Α
Non ruminant blood products	NA	NA	А	Α	Α
Ruminant hydrolyzed proteins	NA	NA	NA	NA	А
Non ruminant hydrolyzed proteins	А	А	А	А	Α
Ruminant hydrolyzed proteins from hides and skins	Α	А	А	А	Α
Di and tricalcium phosphate of animal origin	NA	NA	NA	NA	А
Milk and milk products	А	А	А	А	Α
Colostrum and derivates	Α	А	А	А	Α
Eggs and egg products	А	A	А	А	А

A = authorised; NA = not authorised



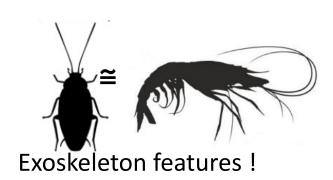
Summary: legal status



- Insect materials are:
 - Processed Animal Proteins (PAP)
 - -Animal Fats

From invertebrates

- Several similarities with Marine arthropods (Shrimp, krill, contained in fish meal)
- Authorized only for farmed fish





Outline



- Legal status
- Efficiency/Quality



- substrate
- -Time
- Insect metamorphosis/harvest life stage
- Technological quality



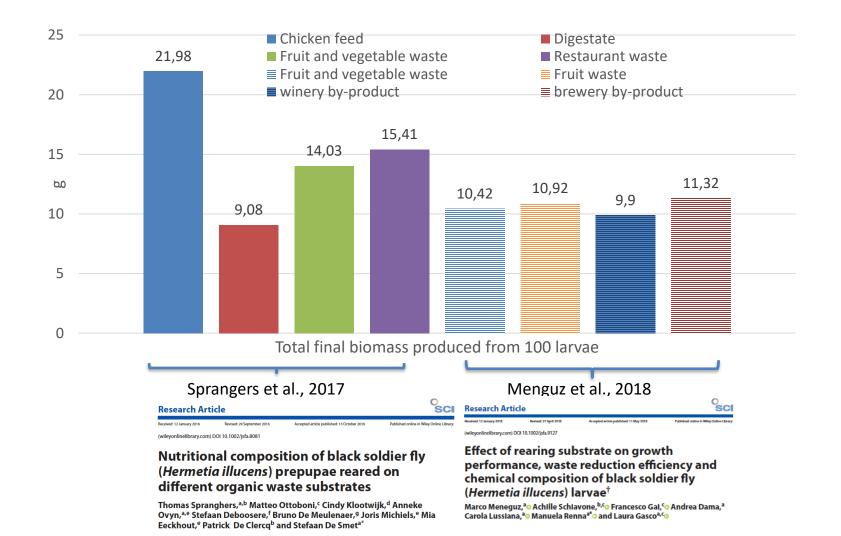
Safety



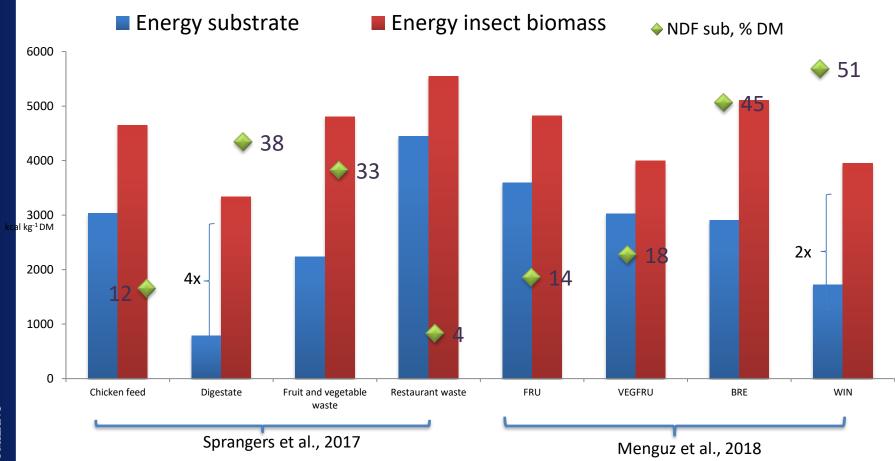
Challenges and future prospective



Efficiency: Total final biomass produced

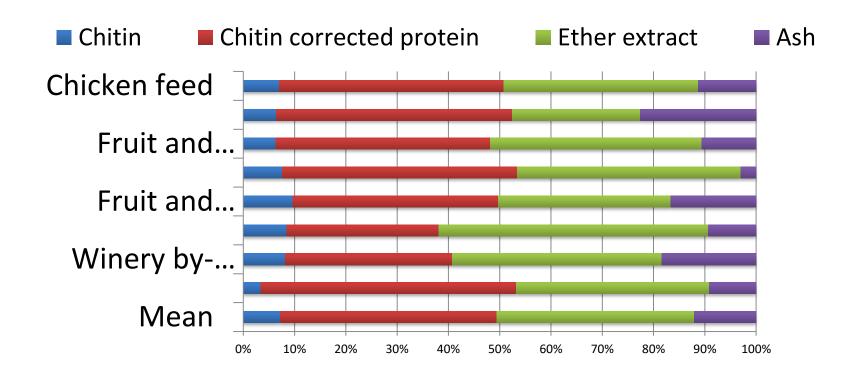


Energy and NDF in substarte vs Energy in biomass



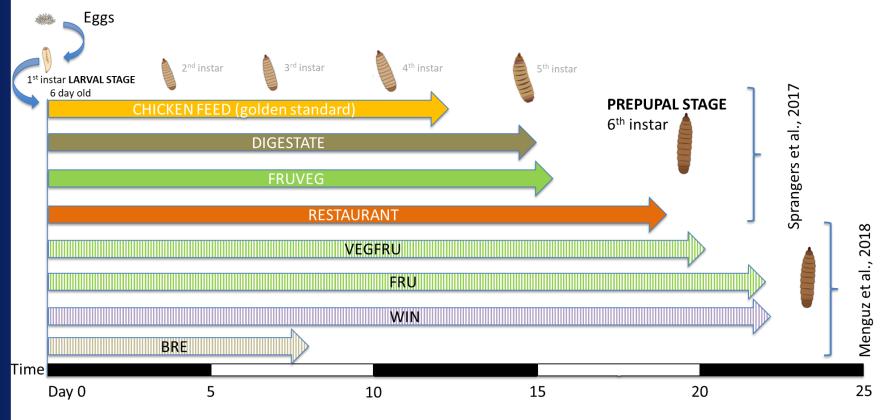


Insect (BSFL) produced on different substrate





Time needed to reach the harvesting stage





Summary: Substrate



Efficiency

- Insect are able to process efficiently substrate high in moisture and fibre.
 - -from 8% DM
 - Opportunity for limited pre-processing
 - Can bioconvert wasteshigh in fibre content (38-55% NDF)

Quality

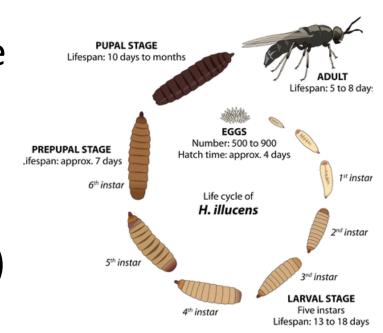
- Protein content and quality is high and comparable for insects reared on different substrates
- Lipid and Ash contents may depend on the substrate
 - Ash in the substrate are higly correlated to ash in harvest insect (r=0.85;P<.001)
 - Ash in the substrate are inv.
 correlated to fat in harvest insect (r=-0.72;P<.005)



Summary:Time



- Time needed to reach harvest phase is variable
 - -1-3 wks, species,substrate dependent
- Micro-livestock features (environment, density,..)





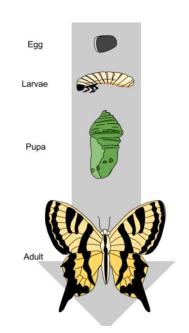
INSECT METHAMORPHOSIS

COMPLETE

88% of al insects

Four Stages

- 1. Egg
- 2. Larva
- 3. Pupa
- 4. Adult

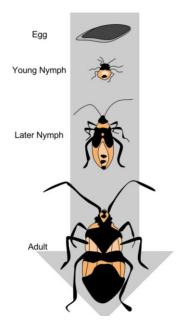


COMPLETE e.g. butterfly, housefly, bees

- 1. black soldier fly,
- 2. house fly,
- 3. yellow mealworm,
- 4. lesser mealworm,

INCOMPLETE 12% of all insects

- Three Stages
 - 1. Egg
 - 2. Nymph
 - 3. Adult



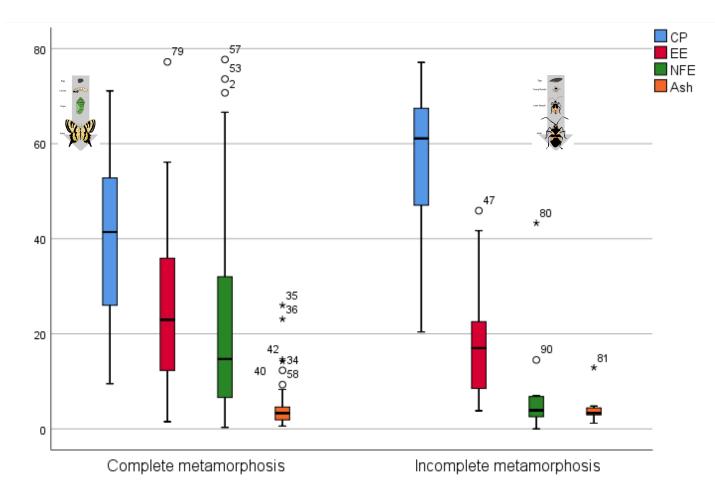
INCOMPLETE Cricket, locust,

- 1. house cricket,
- 2. banded cricket
- 3. field cricket

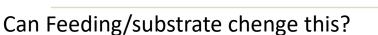


Type of development: source of variability

Box plots displaying mean, median, quartiles, minimum and maximum observations and outliers for nutrient composition (expressed on DM basis) in complete and incomplete metamorphosis insect species (data from Sánchez-Muros et al., 2014). CP – crude protein %; EE – ether extract; NFE nitrogen free extracts;







Challenges to adding insect materials to farm animal feeds

- Variation in nutrient content and nutrient availability between batches/sources...
- Technical aspects/qulaity: processing, feed technology...
- Co-product handling, storage, and transportation...
- Effect on animal performance, end-product quality....
- Safety

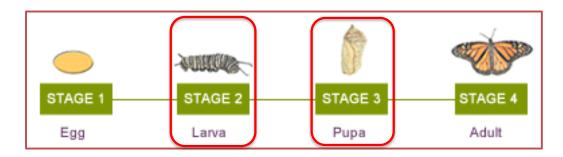




Technological quality: effect of Insect life stage

Rationale of the study:

- to add insect material without processing (high moisture)
- To test the physiological stage: I.E. PREPUPAE VS LARVAE



Tecnologycal treatment investigated: extrusion



Results & discussion **_Experiment 1**



Torque is a measure of the turning force on an object (screw)

premix Insect+wheat 25:75	Crude fat %af	Torque value Ncm	Extrudability
Prepupae	3.15	>400	Not extrudable
Prepupae low oil	3.89	200-400	Not extrudable
Prepupae medium oil	4.63	100-130	ОК
Prepupae high oil	5.37	50-100	ОК
Larvae Caday	4.62	80-120	Best value

BEST MIXTURE Larvae + wheat 25:75 NO OIL

Ottoboni et al., 2018: IJAS



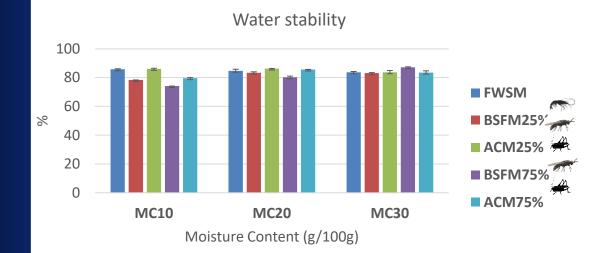
Technological quality: effect of Insect life stage and extrusion

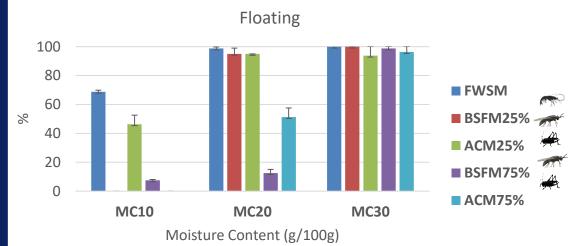
- LARVAE better than pre-pupae (NO OIL needed)
- Fat content in the mixture is a key variable
- Extrusion
 - do not affect CP digestibility
 - increasing OM digestibility
- Results obtained on simple blends!

Ingredient	Control	Fish meal su	Fish meal substituted formulations				
		BSFM25	BSFM50	BSFM75	ACM25	ACM50	ACM75
Sunflower cake	19.0	18.5	18.1	17.6	19.2	19.4	19.6
Maize germ	19.0	18.5	18.1	17.6	19.2	19.4	19.6
Wheat pollard	28.5	27.8	27.1	26.5	28.8	29.1	29.4
FWSM	28.5	20.9	13.6	6.6	21.6	14.5	7.3
BSFM TO THE STATE OF THE STATE	-	9.5	18.4	27.1	-	-	-
ACM	-	-	-	-	6.3	12.5	19.1
Cassava flour	5.0	4.9	4.8	4.6	5	5.1	5.2
Proximate							
Protein	26.6	26.2	25.7	25.3	26.9	27.1	27.4
Fat	11.4	12.7	14	15.2	11.6	11.7	11.9
Fibre	13.7	13.9	14.1	14.5	14.1	14.4	14.8
Ash	9.5	8.7	8	7.3	8.5	7.5	6.5
Carbohydrate	38.8	38.5	38.2	37.7	38.9	39.3	39.4

¹ FWSM = fresh water shrimp meal; BSFM = black soldier fly meal; ACM = adult cricket meal; BSFM25 = BSFM substitutes 25% of the protein supplied by FWSM in control; BSFM50 = BSFM substitutes 50% of the protein supplied by FWSM in control; BSFM75 = BSFM substitutes 75% of the protein supplied by FWSM in control; ACM25 = ACM substitutes 25% of the protein supplied by FWSM in control; ACM50 = ACM substitutes 50% of the protein supplied by FWSM in control; ACM75 = ACM substitutes 75% of the protein supplied by FWSM in control.







technological Feed quality was maintained for all insect meal tested and for all the inclusion levels

Expansion ratio, surface area and volume of pellets were not influenced by insect type and level of substitution

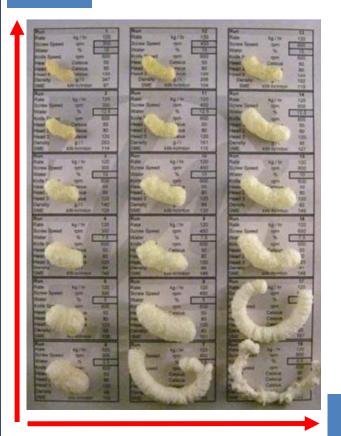
Floating compromised by moisture content in the extruder

Irungu et al., 2018





Moisture



- Insect material an be included in high amount in feed formulas
 - Inclusion up to 25-30% has not detrimental effect on aquafeed technological quality
- Good results with Extrusion
 - Moisture and probably fat content of the blends needs to be adjusted prior to extrusion

Screw speed

Figure courtesy of Dr. Colovic & Wagner Co.





Outline



Legal status



Efficency/Quality



Safety

-Hazards associate with substrate



Challenges and future prospective



Risk profile of insects used as food and feed



Microbiological hazards

Bacteria

Viruses

Parasites

Fungi -> mycotoxins

Prions



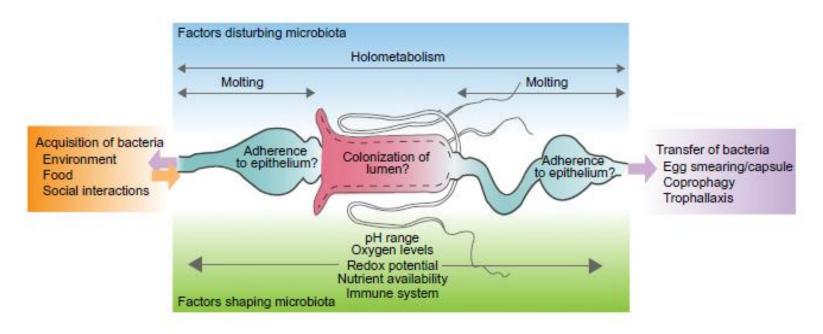
«...hazards associated with other types
of substrate, such as kitchen waste.»

EFSA Scientific Committee. (2015). Risk profile related to production and consumption of insects as food and feed. *EFSA Journal*, 13(10).



Insect GUT

Factors afecting gut insect microbiota and mycobiota

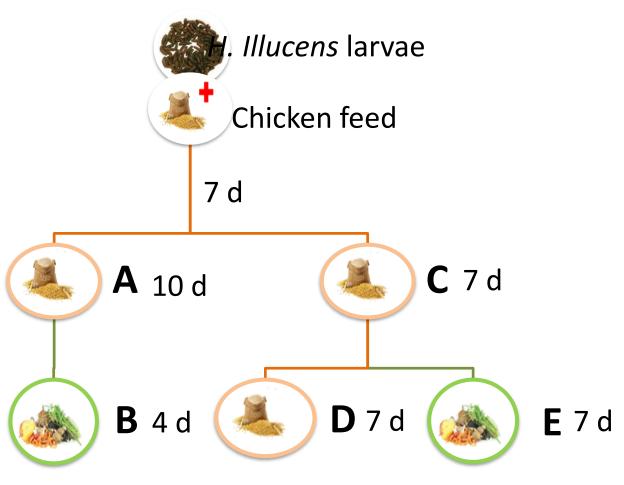


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In insects with distinct larval, pupal, and adult stages, there is a radical remodeling of the gut at metamorphosis...

Aim: Evaluate the impact of the substrate on the intestinal fungal community in *H. Illucens*

Experimental design



Isolation of yeasts and moulds from the intestinal content of larvae Typing of isolates by **ITS-RFLP** Inhibition assays to test the killer phenotype of isolated yeast strains Analysis of fungal community by Next Generation Sequencing 454

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Results

Molecular characterisation

Pichia yeast were present in insect exposed to vegetable waste

Mould isolates were associated with the species

Geotrichum candidum:

Group of larvae	Species	ITS RFLP profile
Δ.	Trichosporon jirovecii	1
Chicken feed	Rhodotorula mucilaginosa	2
17gg	Trichosporon asahii	3
	Pichia fermentans	4
	Saccharomyces servazzii	5
Chicken feed and	Saccharomyces spencerorum	6
vegetable waste	Pichia kluyveri	7
17gg+4gg	Trichosporon asahii	3
	Pichia kudriavzevii	8
	Candida tropicalis	9
С	Meyerozyma guilliermondii	10
Chicken feed 14gg	Geotrichum candidum	11
	Trichosporon asahii	3
D	Trichosporon asahii	3
Chicken feed	Meyerozyma guilliermondii	10
21gg	Geotrichum candidum	11
_	Pichia kluyveri	7
E Chicken feed and	Meyerozyma guilliermondii	10
vegetable wast		4
14gg+7gg	Saccharomyces servazzii	5



-No foodborne disease has been linked to the consumption of products containing *G. candidum* (Pottier et al., 2008).



Results

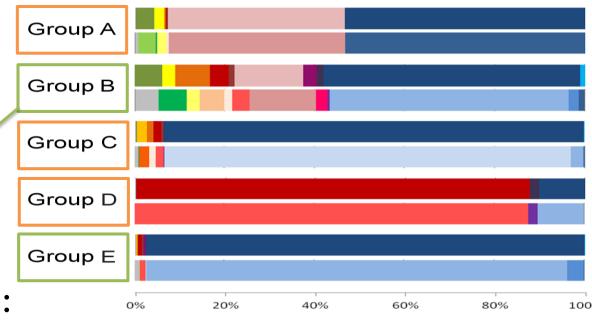
Next Generation Sequencing

Taxonomic composition of intestinal mycobiota of HI larvae

(fungal ribosomal ITS region)

Chicken feed (17d) + Vegetable waste (4d):

The greatest fungal diversity







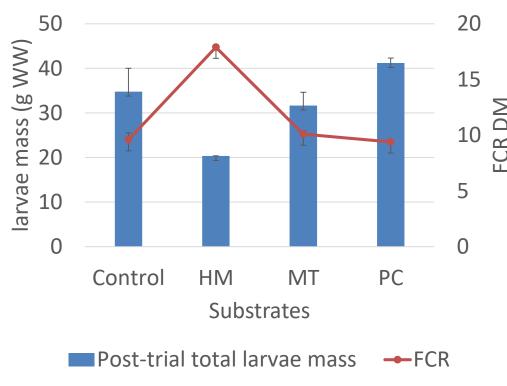
Impact of substrate contamination with mycotoxins, heavy metals and pesticides on the growth performance of BSF larvae

Purschke et al., (2017). Food Add Cont: Part A, 34(8), 1410-1420.

- Heavy metals reduce larvae mass
- Mycotoxins and pesticides does not affect larvae growth

Control and contaminated substrates, containing defined amounts of :

- heavy metals (HM)
- mycotoxins (MT)
- pesticides (PC)





Rearing larvae on contaminated substrates

Purschke et al., (2017).

Mycotoxins and pesticides have neither been accumulated in the larval tissue

Significant bioaccumulation of Cd and Pb was observed in the larvae.

Bio-accumulation Factors:

Cadmium: >9

Lead: > 2

	Heavy metal (HM) – contaminated substrates		
	Initial substrate (mg kg-1)	Residual substrate (mg kg-1)	
Chrome	15.2	19.9 ± 3.0	
Nickel	15.2	19.7 ± 3.0	
Arsenic	3.0	3.8 ± 0.6	
Cadmium	1.5	1.8 ± 0.3	
Mercury	0.2	0.3 ± 0.08	
Lead	15.2	19.8 ± 3.0	
	Control (mg kg-1)	нм (mg kg–1)	
Chrome	0.064 ± 0.01	3.4 ± 0.5	
Nickel	0.048 ± 0.007	4.2 ± 0.6	
Arsenic	< LOQ of 0.024	2.8 ± 0.4	
Cadmium	0.048 ± 0.007	13.7 ± 2.1	
Mercury	< LOQ of 0.012	0.1 ± 0.03	
Lead	0.032 ± 0.005	35.6 ± 5.3	



Summary: Safety



- Insects for feed are processed with their GIT content, which can harbour different species of transmissible microorganisms.
 - Insect mycobiota and microbiota can be enriched/modulated during farming and processing.
- Chemical hazards, like pesticides, fluorine, heavy metals, and dioxins, merit specific evaluation.
 - Bioaccumulation risks





Outline



Legal status



Efficency/Quality



Safety



Challenges and future prospective







- Theuse of appropriate and tailored substrates could lead to the production of a premium feed specialty, providing new opportunities for raw materials and diet formulations.
 - E.g. The case of BSF on fish offal
- This implies that a standardization of the rearing protocols is needed
- Setting type of substrate /Time (e.g. Growth vs finishing phase)
- Limits; ABP regulation

	Ingredients		
Fatty acids (%)	Normal black soldier fly prepupae	Enriched black soldier fly prepupae	
12:0	23.6	37.1	
14:0	5.1	6.3	
16:0	19.8	17.3	
16:1n7	6.3	7.6	
18:0	6.5	2.0	
18:1n9	22.7	18.8	
18:2n6	6.8	5.9	
18:3n3	0.0	0.5	
18:4n3	0.0	0.5	
20:5n3	0.1	3.5	
22:5n3	0	0.35	
22:6n3	0	1.7	

All values are reported as means of duplicate analysis



General conclusions (1/2)

- Insects
 - -can be used as PAP or animal fats
 - Can upgrade waste biomasses/streams to valuable feed ingredients
 - —are able to process efficiently substrate high in moisture and fibre.
 - –have not detrimental effect on feed technological quality
 - -Safety evaluation cannot be considered as complete



General conclusions (2/2)

- Source of variability
 - Insect species
 - Substrate
 - Exposure time
 - Harvesting growing phase
 - Types of development metamorphosis



...To be defined/adressed

- Balance diet is needed ??(chicken feed results)
 - -Requirements??
 - –Feeding phases (growing, finishing....)
 - Feed speciality production
- FCR not defined (from 1.5 to 16!!)
- Micro-livestock features (environment, density,..)





Thanks for the your attention

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