Enhancing insect welfare: assessing dietary practices for farmed Insects in the European Union

EUROGROUP FOR ANIMALS

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Foreword

In March 2022, Eurogroup for Animals shared a review of the behavioural needs and welfare of farmed insects, urging the European Union to take up this crucial issue. A year and a half later, the European Parliament's Protein Strategy Own-Initiative Report (INI) stressed that "environmental, health, animal welfare, social and economic effects need to be analysed" when considering the development of the insect farming industry.

With this new review, we take a step forward and a closer look at the key question of dietary practices and associated welfare needs of farmed insects in Europe. This review, by Dr Helen Lambert and Dr Amelia Cornish - animal welfare experts - uses the latest evidence on insect welfare and behaviour science, as well as data directly collected from insect producers, to formulate key recommendations. These constitute a strong basis for future work at industry or legislative level on insect welfare best practices.

1. The need for welfare in a growing industry

Ten species of insects are currently authorised or pending authorisation to be farmed in Europe. As the insect farming industry is rapidly developing and is already rearing insects by the billions, clear rules surrounding the protection of their welfare are still missing, for lack of research and interest from policymakers.

Even the industry organisation (International Platform for Insects as Food and Feed - IPIFF) recognises its limited knowledge, stating that "the current lack of scientific evidence around invertebrate welfare makes it very difficult to develop science-based welfare rules for insect production". At the same time, producers have expressed their support for developing insect welfare best practices and have called for the application of Brambell's model of the Five Freedoms as a starting point.

If we look at industry growth objectives and the tonnage they hope to achieve in 2030, it is equivalent to 45 trillion to 50 trillion individuals reared at any one time. Species of insects authorised for farming purposes in the European Union include some from orders for which the most recent evidence strongly suggest they have the ability to feel pain. We believe that there is an urgency to tackle this issue and act for the welfare of farmed insects.

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2. Key recommendations from the review

With an extensive review of all farmed species in Europe, this study identifies that appropriate diets are essential for ensuring each species' welfare needs are met. For many species, heterogenous diets are necessary to meet the animals' nutritional needs and, although many food residues of agricultural by-products are considered as insect substrates, there is a need for caution as these streams may be unreliable and inconsistent. For health and welfare reasons, these potential substrates need to be processed to avoid any pathogens and contaminants entering the food chain.

The study of multiple diets reveals the need for dietary substrates to be safe, hygienic, nutritional and environmentally suitable. If the available evidence already suggests the use or avoidance of certain diets, there is a need for further research on insects dietary preferences. If mortality rates as a proxy for insect welfare can help in identifying diets that are economically viable yet may be detrimental for welfare, attention should be given to diets that are naturally self-selected by insects.

Other major concerns include forced starvation that arises in the rearing of multiple species, including adult Black Soldier Flies or Crickets. This practice is clearly in stark contrast to the Five Freedoms model that requires “freedom from hunger”. Starvation can, moreover, trigger cannibalistic behaviours. Genetic manipulation has to be approached carefully, as it may affect the expression of natural behaviour in feeding. Experience in genetic manipulation of other farmed animals speaks in favour of proceeding with great caution.

3. Animal welfare should not be sacrificed to production and circularity

Most importantly, this review highlights that research on insect farming is often carried out through a production and economic lens, not a welfare lens. Even though welfare, health, and economic viability can go hand in hand, the strive for profitability should not be made at the expense of the animal’s welfare. When it comes to diets, this review therefore highlights that “diets that are optimum in terms of productivity may be used despite being disastrous in terms of insect welfare.”

It is critical that the insect farming industry does not go down the tragic path taken by the rest of the animal agriculture industry before it. This rising industry is currently at a crossroads in its development, and Eurogroup for Animals strongly calls for welfare issues not to be dismissed or sacrificed in the quest for profitability or circularity.

European legislation currently prevents the use of some substrates for insects on public health grounds. We believe that, in addition to these legitimate health concerns, welfare should also be taken into account when authorising new food streams for farmed insects.

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4. Eurogroup for Animals calls for the application of the precautionary principle and the development of welfare best practices

Although the subject needs to be more fully researched, the scientific community already suggests applying the precautionary principle when considering the capacity of insects to suffer. This means that insects should be treated humanely both as they are reared and at the time of slaughter. Throughout their lifespan, insects should be raised in ways that respect their species-specific needs and behaviours, despite industry productivity considerations.

In addition to applying the precautionary principle and researching sentience and welfare needs before any new authorisation of species for farming purposes, we call on the European Union to address globally the issue of invertebrate animals' welfare. Furthermore, we call on insect producers to take up the subject and work with researchers and animal welfare organisations to develop and implement insect welfare and health best practices .

Diets are an essential part of farmed insects' welfare. Yet other areas need to be given full consideration as evidence calls for improving welfare by acting on rearing densities, methods of slaughter, species-appropriate temperature and humidity levels, among others. Eurogroup for Animals' last review already touched on many of these issues.

5. Insect farming is also a food system issue

Industrial insect production does not only raise welfare concerns. Branded as an environmentally friendly food solution, it does not necessarily go hand in hand with sustainable food systems. In fact, behind what European Commission experts call "an overwhelming lack of knowledge" surrounding the industry, there are a number of issues beyond welfare that require policy makers and the sector to adhere to the precautionary principle.

One of the main arguments in favour of insect farming is that insects can feed on waste and other products not fit for human or animal consumption and upcycle them into protein to be fed to other farmed animals. Notwithstanding ingredients that are not authorised in insect substrates for hygiene and health reasons, this review precisely highlights that not all insect species can thrive on waste or poor quality substrates.

In fact, according to the industry association IPIFF, producers use a number of different ingredients and about three-quarters use fruits, vegetables, and cereal. These are resources that could be used for direct human consumption or fed directly to extensively reared chickens and pigs. Around a third of insect producers use commercial feed which can include soy. Feeding insects ingredients that could be consumed directly by animals or people is inefficient. The European Union is mindful of the need to reduce the food-feed competition and has set out objectives in its Farm to Fork strategy to make the food system more resilient.

The sustainable food system promoted by the EU should focus on reducing the amount of animal products and supplying them from systems with higher welfare standards. Boosting insect farming for animal feed will sustain factory farming with its serious animal welfare concerns. Indeed, the European Commission’s Agricultural Outlook forecasts that the increased supply of insect meal and lower prices could support conventional intensive animal production if the practice is fully commercialised and existing restrictions lifted.

Insect farming is not necessarily compatible with a more sustainable food system. It raises new animal welfare issues as well as supports, rather than limits, intensive animal farming and its consequences on environmentally unsustainable and unhealthy high-animal product diets.

Summary

In the last decade, the use of insects as a source of protein for both humans and livestock has been proposed as an environmentally friendly alternative to farming traditional livestock and feed ingredients such as soy. To date, four species are approved for human consumption in the EU: migratory locusts, house crickets, and the yellow and lesser mealworms, and seven species are authorised for use as feed for aquaculture, poultry and swine animals, which are the black soldier fly, common housefly, yellow and lesser mealworms, silkworms and the house, banded and field crickets.

Producers of insects are considered ‘primary producers’ and must comply with the same safety requirements and hygiene practices as all other food and feed business operators active in the EU's food and feed sectors. Insects farmed within the EU are categorised as ‘farmed animals’ in the EU Animal-By Products (ABP) legislation. This means that insects are subjected to the same restrictions on feed that govern farmed vertebrates, regardless of whether the end product is intended for consumption, feed or biofuel production.

Insects are invertebrate animals with specific and varied dietary needs and, therefore, should have species-specific legislation to ensure their welfare needs are fully met. Currently, legislation regarding insect diets is more focused on hygiene and safety and lacks species-specific standards to govern their welfare.

Overarching welfare issues regarding insect diets

Several overarching welfare concerns regarding the diets of all farmed and pending insect species exist. Firstly, production and research efforts are

focused on production traits and economics rather than welfare, and consequently, the concept of welfare is often disregarded when assessing suitable diets. In this report, we have used proxies for welfare, including survivability, disease resistance, development rates and longevity, which, although may be more production-focused, do also provide important indications for the welfare of the insects in terms of the available data.

Production and welfare can be aligned, and for some insects, some diets are good for welfare and production and may even have wider sustainability benefits. However, with the drive from industry to make insect farming more efficient, profitable, and productive, welfare is often at odds with industry. In terms of diet, this can mean that diets that are optimum in terms of productivity may be used despite being disastrous in terms of insect welfare. For instance, high mortality rates may be acceptable from a production perspective, provided the diet is cheap and development time is short.

There is a significant interest in the use of waste and side streams for rearing insects, as this helps create a highly desirable circular economy and assists with processing the huge amounts of waste created by various industries. The suitability of specific waste streams is discussed in the relevant species sections, but there are some overarching considerations, too. For example, waste streams are often unreliable and inconsistent, leading to variable results. They also need to be processed to remove all hazards and contaminants, and the results may not be applicable at industrial levels.

Black Soldier Fly (*Hermetia illucens*)

Black soldier fly larvae are farmed for livestock feed in the EU. The larvae can consume most substrates and are typically fed a range of by-products, including dairy, vegetable and bakery by-products, brewers' or distillers' grains, and animal feeds. The industry is also seeking to expand into food surplus and animal by-products in the short term, and longer-term plans are in place to explore the use of municipal waste, manure, and slaughterhouse wastes. However, these are not currently allowed in the EU.

The main welfare concerns are focused on finding the right diet for the needs of the larvae, and this includes recognising that the larvae have preferences and will naturally self-select optimum diets from a range of feedstuffs. Furthermore, diets differ in terms of how successful they are in terms of production versus mortality rates, and producers will often accept higher mortality rates for improved productivity at the expense of welfare. There are also significant concerns regarding the adult black soldier flies, as they are not typically fed, despite there being clear evidence of the benefits of feeding them.

Common Housefly (*Musca domestica*)

As with the black soldier fly, the larvae of the common house fly are farmed for livestock feed. Poultry manure is commonly used for larvae, but other substrates, such as offal, pig or cattle manure, blood, wheat bran, or rotten fruit, may also be used or mixed in. However, as many of these substrates are not permitted in the EU, the production of housefly larvae is relatively limited.

Research has explored what properties of manure are important for common housefly production, including moisture content and nutritional composition. However, to date, no research has explored the larvae preferences or considered the welfare impacts of the different feeds. Similarly, there is very little research into the optimum diets for adults, which is an important research area.

Yellow Mealworms (*Tenebrio molitor*), Lesser Mealworms (*Alphitobius diaperinus)* and the Adult Darkling Beetles

The lesser mealworm and the yellow mealworm are larvae from beetles from the darkling beetle family. Mealworms will consume a range of substrates, including decaying grains, cereals, meal scraps, bread, dead insects, feathers, live conspecifics, decaying wood, and grains mixed with animal excrement. Commercially, they are typically fed a homogenous wheat bran diet, which raises significant welfare issues as it fails to provide a nutritional balance. Research shows that mealworms have better welfare and often production outcomes when given heterogeneous diets and show clear preferences when they can self-select their diets.

There is limited research regarding the optimum diets for adult beetles, although what has been done highlights that the typical wheat bran diet cannot meet their nutritional and welfare needs.

Banded Crickets (*Gryllodes sigallatus*), House Crickets (*Acheta domesticus*), and Field Crickets (*Gryllus assimilis*)

Unlike some of the other species, it is the adult cricket that is slaughtered for feed and consumption. There has been much trial and error regarding finding the optimal cricket diets, and much more research is needed. Crickets often undergo dietary restriction in commercial farms, where they are either given inadequate amounts of feed to save money, given inappropriate diets or starved for a period (typically before slaughter). Dietary restriction causes significant welfare concerns, including mortality, poor immunity, and starvation, and can result in outbreaks of cannibalistic behaviour.

There is also significant evidence to show that contaminants, such as pesticide residues and microplastics, are often present in end-products, such as cricket flour. Not only can this have public health concerns, but research indicates that the crickets may also suffer as a result.

Migratory Locust (*Locusta migratoria*)

The migratory locusts have been relatively understudied in comparison with others, and much more research is needed into the welfare implications of feeding them an artificial diet, as opposed to the plant-based diet they have evolved to eat. Research is sorely needed in this area, as to date, much of the research has been focused on how diets affect the flavour and nutritional composition of the locust as a protein source for human consumption rather than on the welfare of the insects themselves.

Western Honeybees (*Apis melifera*)

The Western honeybee (*Apis melifera*) is being considered for human consumption in the EU and potentially as animal feed. In particular, the honeybee drone brood (Apis mellifera male pupae) is currently being proposed as a novel food as a by-product of existing apiculture production rather than a stand-alone industry.

Adult honeybees consume nectar and pollen from flowers. Therefore, access to abundant and varied flowers is necessary for honeybee brood production, immune function, and the general survival of the colony. Today, commercial beekeepers often provide their colonies with an artificial "pollen substitute" to compensate for insufficient nutritional forage in the environment. Artificial diets do not provide the bees with important micronutrients, and as a result, the colonies suffer negative consequences. For example, studies have shown that access to natural forage results in improved health, reduced pathogen loads, and higher overwintering survival rates than artificial diets. More research into the welfare implications of certain diets in honeybee farming is required.

Silkworms (*Bombyx mori*)

Silkworms have so far been farmed for their silk (sericulture). The silkworm pupae, a by-product once the silk thread has been wound off the cocoon, are also edible and are frequently used as food in Asian countries.

Silkworms are monophagous insects that only feed on fresh mulberry leaves (preferably white mulberry species) or sometimes Osage orange leaves. In the 1960s, an artificial diet was developed, commonly called ‘silkworm chow’. Despite efforts to continually improve artificial diet formulae, silkworms reared on artificial diets have higher mortality rates, poorer development and physical weakness, poorer metabolism, reduced disease resistance and low silk yield development compared with those on a natural diet, which is thought to be due to the metabolic utilisation of artificial diets in silkworms being less than that of mulberry leaves. More research is needed to investigate the welfare implications of rearing silkworms on artificial diets.

Conclusion

One overwhelming factor coming from the scientific literature is that there is a considerable amount of trial and error still happening in insect farms concerning the diets used. The consequence of this is that the welfare of the insects suffers, as mortality rates increase when non-optimal diets are used, and this may incur a degree of suffering or ill health. Many unanswered questions remain regarding what is best for insect welfare. Therefore, further research is desperately needed for all farmed and pending species to establish the optimum diets for welfare.

1. Introduction to insect farming

In the last decade, as environmental concerns have increased, the use of insects as a source of protein has become a potentially environmentally friendly alternative to traditional livestock. Those who advocate for farming insects for consumption claim that they are safe and nutritious and that they are more efficient to rear than vertebrate livestock, as they not only need less space, food, and water, but they also convert their food to protein more efficiently.

In terms of financial investments, most attention is on using insects as feed for livestock, which can offer benefits in terms of cost, efficiency and sustainability compared with growing soy and fishmeal. The potential for insects to be reared on waste streams or agricultural byproducts is of particular interest as not only does this reduce waste and emissions and require fewer resources, but it can also result in a desirable circular economy.

2. EU legislation relevant to insect feed

In September 2021, Regulation (EU) 2021/1372 was updated to allow the EU Member States to use processed animal proteins (PAPs) derived from farmed insects as feed for farmed fish, poultry, and pigs within the Regulation (EC) 1069/2009, building on the 2017 authorisation of insect protein for aquafeed (Regulation (EU) 2017/893).

To date, four species are approved for human consumption in the EU: migratory locusts, house crickets, and the yellow and lesser mealworms, and seven species are authorised for use as feed for aquaculture, poultry and swine animals, which are the black soldier fly, common housefly, yellow and lesser mealworms, silkworms and the house, banded and field crickets (European Commission, 2023; IPIFF, 2023).

2.1. Summary of legislation regarding the substrates used as feed for insects

Producers of insects are considered 'primary producers' and must comply with the same safety requirements and hygiene practices as all other food and feed business operators active in the EU's food and feed sectors (IPIFF, 2022) (see table 1).

Insects farmed within the EU are categorised as 'farmed animals' in the EU Animal-By Products (ABP) legislation (see Regulation (EC)1069/2009 and Regulation (EU) 142/2011). This means that insects are subjected to the same restrictions on feed that govern more traditionally farmed species, regardless of whether the end product is intended for consumption, feed or biofuel production (IPIFF, 2022). Regulation (EU) 68/2013 provides a list of feed materials commonly used on the EU market. However, this list is non-exhaustive, and many further restrictions apply. The table below provides a non-exhaustive summary of the key regulations pertaining to the diets and dietary substrates used for insects farmed in the EU.

Table 1. Summary of the key regulations relating to the diets and dietary substrates used for farmed insects in the EU.

Note the following Regulations also apply but were outside of the scope of this report: Regulation (EC) No 396/2005 addresses the maximum pesticide residue limits as applied to feed materials; Regulation (EU) 2019/6 (addresses the addition of veterinary medicinal products via feed or water); Regulation (EU) 2019/4 (addresses the manufacture, placing on the market and use of medicated feed); and Directive 2002/32/EC (addresses undesirable substances in products intended for animal feed).

Regulation	Summarised focus	SUBSTRATES FOR INSECTS FARMED IN THE EU	
		Authorised	Prohibited
Regulation (EC) 1069/2009	Public health and animal health risks associated with animal by-products and derived products	Feed materials of vegetal origin.	Prohibits the use of certain materials of animal origin, such as manure and catering waste.

Regulation (EC) 999/2001	Prevention, control, and eradication of transmissible spongiform encephalopathies	● Rendered animal fat ● Hydrolysed proteins, collagen and gelatine or blood products derived from non-ruminants (or parts of non-ruminants) (including compound feed containing such products) ● Hydrolysed proteins from ruminant hide and skins ● Dicalcium phosphate and tricalcium phosphate of animal origin (including compound feed containing such phosphates) ● Fishmeal ● Former food stuff without meat and/or fish. ● Eggs and egg products ● Milk, milk based-products and milk-derived products ● Honey	Prohibits Processed Animal Proteins (PAPs), in particular: ● PAPs from ruminants ● PAPs from poultry animals ● PAPs from swine animals ● PAPs from famed insects ● PAPs from other non-ruminants.

		Note all precautions must have been taken to prevent the contamination. For example, materials must have been previously processed (e.g. milk must have been pasteurised, and eggs must be cooked).	
[Regulation (EC) 767/2009]()	Sale and use of animal feed	Only allows insects to be fed with 'safe feed.' Safe feed is referred to as feed that does not have a direct adverse effect on the environment or animal welfare, and "a) is sound, genuine, unadulterated, fit for its purpose and of merchantable quality; and (b) is labelled, packaged and presented in accordance with the provisions laid down in this Regulation and other applicable Community legislation."	• Faeces and separated digestive tract content (this includes substrates mixed with frass from insects or dead insects) • Hide treated with tanning substances • Seeds and other plant-propagating materials • Wood and materials derived from wood • Waste derived from urban, domestic, and industrial waste treatment

			• Household waste • Packaging from agri-food products • Protein products obtained from yeasts of the Candida variety cultivated on n-alkanes.
Regulation (EC) 1831/2003	Additives in animal nutrition	Only the feed additives which are authorised for all animal species may be used as feed for insects. Note no specific additives for insects have been defined.	

2.2 Review of relevant legislation on substrates of animal origin as feed for insect

As insects are categorised as 'farmed animals' under EU law, using manure, other slaughterhouse or rendering derived products, catering waste or former foodstuff containing meat or fish is not permitted. As a result, the use of waste and side streams for rearing insects, and the desirable circular economies and processing of industrial waste, is not legally permitted in the EU at the moment and excludes many potential environmental benefits of waste-fed insects (Van Zanten et al., 2015). Nonetheless, where relevant, we have discussed the suitability of specific waste streams for each relevant species below, as this is something that the IPIFF (an EU non-profit organisation which represents the interests of the insect production sector) are lobbying for and warrants exploration as changes to legislation may occur in the future, (see the IPIFF Regulatory brochure for more information, IPIFF, 2020).

Furthermore, there may be potential welfare concerns arising from the restrictions placed on feed, particularly regarding the omnivorous species that may have improved welfare outcomes from a more varied diet. However, as the following species-specific sections discuss, waste and side streams are often inconsistent in composition and, therefore, can have variable results. Also, the implications of build-ups of pesticides, heavy metals and veterinary medicines in insects and the associated welfare concerns warrant further investigation (Malematja et al., 2023).

There is a need for further research into the implications of certain substrates on insect welfare. Insects are invertebrate animals with specific and varied dietary needs and, therefore, should have species-specific legislation to ensure their welfare needs are being met. It is also worth noting that the fact that insects are categorised as ‘farmed animals’ is a positive step in terms of their status, as insects, along with all other invertebrates, are still excluded from the Council Directive 98/58/EC concerning the protection of animals kept for farming purposes. Although the motive behind classing them as farmed animals is likely to be based on food safety, the importance of terminology when referring to animals is still of utmost importance (Merskin, 2022). In particular, referring to insects as ‘animals’ and categorising them along with vertebrate-farmed animals strengthens the perception that these ‘mini livestock’ are just as worthy of protective welfare-based legislation. This is particularly important as their omission in the Farming Directive is due to them still widely being considered as insentient. However, this is due to an absence of research and not an absence of evidence (Lambert et al., 2021).

Despite the challenges of establishing sentience in invertebrate species (Klein and Barron, 2016), there is growing momentum regarding scientific evidence of insect sentience (Lambert et al., 2021). Both the precautionary principle and the growing evidence for sentience in insects provide an ethical basis for farming insects in ways that optimise their welfare (Bear, 2020, 2019; Boppré and Vane-Wright, 2019; van Huis, 2019), which means providing feeds and dietary substrates that meet their welfare needs. Insect farming in the EU is only likely to grow, and so research into the impacts of farming, including the feed and substrates used, on the welfare of insects is greatly needed.

3. Overarching considerations and recommendations in relation to feed

3.1. Overarching considerations (non-species-specific)

3.1.1. Proxies for insect welfare

Research and practices regarding insect diets in commercial systems primarily focus on production traits and economics rather than welfare, although interest in welfare is growing. Throughout this report, we have used proxies for welfare in the absence of a more thorough understanding and exploration of the subjective minds of insects in both scientific research and the industry. For instance, mortality rates are an indicator of individual stress, and if the species is sentient, this would likely incur a negative emotional state [14]. As mortality rates can only be considered *post-hoc*, others suggest that the metabolic conditions, specifically oxidative stress responses of larvae growing on different diets, could be used to provide some information on tolerance prior to death, although this is generally understudied [15].

Other proxies throughout this report include disease resistance and immunity, development rates and longevity. Development rates are a particular focus of the industry, and often shorter development periods are preferred, especially for those species slaughtered at an adult stage [e.g., 16,17]. Typically, growth rate is not considered to indicate welfare in animals due to the selective pressures often placed on farmed animals to grow quickly despite being in a poor state of welfare. However, unnaturally slow and even exceptionally fast development periods and growth rates in insects may indicate poor welfare, as the insect may not consume enough nutrients for normal development

[14,18]. Similarly, a particularly low final adult or larval weight can also highlight issues such as an inadequate quantity or quality of diet [14,19]. These factors can impact longevity, particularly if the insect does not have enough nutrients to sustain them through the pupate and adult stages [20]. Lastly, in some species, disease resistance and immunity, or more significantly, a lack of, can also be influenced by the diet, especially when more artificial or processed diets are utilised, which lack the important microbial diversity [21,22].

3.1.2. Production versus welfare

Diet choice by industry may be dictated by numerous factors, including cost, sustainability drivers, preferences for the nutritional composition of the final product, productivity, and resource availability. Whereas the impact on welfare, beyond the proxies mentioned, is often not considered. Furthermore, feeds that may be optimum for insect welfare may not be optimum for production parameters. For instance, mortality rates may be low with one diet, but if growth rate and slaughter weight are also low, a diet may be rendered unsuitable from a production perspective.

3.1.3. Diets as substrates

For some of the species mentioned (i.e., the common house fly larvae, black soldier fly larvae and mealworms, the diet is also the substrate in which they are kept. Therefore, there are additional considerations that apply to these insects. For instance, a substrate may be environmentally sound at a large scale yet be unable to offer the nutritional content required for optimum welfare [14]. Similarly, there may be issues with using some dietary substrates at scale, as although black soldier fly larvae require a moist substrate, if the substrate is too deep, the moisture may pool at the bottom, drowning the larvae, whilst leading to those in the top desiccating from too dry a substrate [14].

3.1.4. Optimising the diets provided

Regardless of whether or not the insect lives in their dietary substrate, their ability to utilise the nutritional content provided can also be impacted by abiotic conditions. For instance, if the environment is not optimal in temperature and humidity, the insects may be unable to access or digest properly due to biological trade-offs prioritising baseline survival over consumption [23,24]. Similarly, the feed needs to be provided in a manner suitable for the species and the stage in their lifecycle, whether by processing to smaller particle sizes or providing a liquid diet, for example. Feed distribution is also an important consideration, as some insects are relatively immobile in the early stages of the life cycle and cannot utilise poorly distributed feed [18].

3.1.5. Circular economies and waste streams

There is a significant interest in the use of waste and side streams for rearing insects, as this helps create a highly desirable circular economy and assists with processing the huge amounts of waste created by various industries. The suitability of specific waste streams is discussed in the relevant species sections, but there are some overarching considerations, too. For instance, waste streams must be processed to ensure they are safe and hygienic. Kitchen waste, for example, which is considered by some to be an ideal source of substrate for the fly larvae [25–27], may contain dangerous or toxic litter (e.g., glass, cigarette butts), which will need to be removed [14]. Furthermore, pesticides may accumulate to unsuitable levels for consumption, which may impact insect welfare and have implications further up the food chain [14,28,29]. Although farmed insects are considered to be less of a risk compared with wild insects, pesticide consumption still poses a significant risk to the welfare of insects. For example, Kolakowski et al. [30] found that 89% of cricket products purchased from Canadian retailers contained pesticide residues, 9% of which were above the legal limit.

Another consideration is that many of the studies into diets using economical waste streams have been performed at laboratory levels and not tested at industrial scales [31]. Not only are there likely to be differences at such scales, but there may also be differences depending on the life stage and sex of the insect, as well as the influence of other abiotic factors [31,32].

It is important to note that many studies for the ‘filth feeding flies’; namely the black soldier fly and the common house fly, are focussed on using manure as feed. Although this is not legally permitted in the EU, bodies such as the IPIFF are lobbying on this [3], so findings regarding the use of manure are potentially relevant should there be changes to legislation in the future. The reason behind the prohibition of manure for farmed animals in the EU is due to the risk of transmitting diseases and infections, for both animal and human health, as manure can contain harmful bacteria such as E. coli and Salmonella.

3.1.6. Genetic manipulation

Technologies for genetic manipulation are developing significantly around the world, and in terms of insects, there are drives to make insect species develop faster, weigh more, and have better feed conversion rates. It is not clear as yet what implications this may have upon their dietary needs, but given that the insect will be required to grow faster and larger, there will no doubt be welfare implications for diet and beyond. Further research is needed in this area to explore the implications further.

3.2. Overarching recommendations regarding insect feed

- Diets (and dietary substrates) should be safe and hygienic for use. Therefore, waste streams must be processed to ensure that any litter is removed, that pesticides are not at dangerous levels, and that it is free of harmful pollutants.
- Substrates must be both nutritionally and environmentally suitable for insect species that live in and consume their substrates (i.e., common house fly larvae, black soldier fly larvae, mealworms, and larvae from other species).
- Diets (and dietary substrates) must be suitable for use at the required scale (i.e., something that works environmentally at a laboratory or bench-top scale may not work on a large commercial scale).
- Research is needed into the importance of providing a heterogenous diet for all relevant species, given the tendency of commercial farms to rely on homogenous diets.

- Research into the welfare implications of contaminants such as pesticides is widely needed, not only in terms of the residue levels residing in the final product but what welfare impacts different contaminants have on the welfare of the insects affected.
- Research is needed into the welfare implications of feed withdrawal prior to slaughter for all species. Initial research suggests that feed withdrawal of over 24 hours can result in cannibalism in some species (e.g., crickets), so this is an area of significant concern.

4. Black Soldier Fly (*Hermetia illucens*)

The black soldier fly (*Hermetia illucens*) (BSF) is approved as feed for farmed animals in the EU, and it is specifically the larvae that are used [29].

4.1. Natural biology of the black soldier fly

The total lifecycle of the black soldier fly (BSF) is around 45 days and comprises four stages: egg, larva, pupa, and adult. The larval stage is around 18 days, the pupae stage is around 14 days, and the adult stage is around nine days, depending on conditions. The female only lays eggs once, ovipositing between 500-900 eggs, and then dies soon after [33].

4.2. Overview of farming systems for black soldier fly

Current industry practices vary significantly and continuously change as this sector grows, adapts, and develops new technologies [14]. Commercial systems typically rear adults separately from the larvae in mesh cages, in greenhouses or in houses with artificial lighting [14].

The larvae are reared in stackable pans, where the 5-day-old larvae are mixed with the dietary substrate and added to the 'larveros' (containers) for waste processing [14]. The BSF larvae (BSFL) live in and consume their substrate, which means that the substrate has to be environmentally and nutritionally appropriate. The substrate typically has a depth of no more than 5cm and can be given either all in one go or incrementally (every 3-5 days) to help prevent overfeeding and rot, the latter being more common in automated systems [34].

4.3. Substrates typically used for black soldier fly larvae

BSFL can consume most substrates and naturally consume a wide range, from coffee grounds to faeces, carrion, and vegetable waste [28,35,36]. Producers are typically reticent to divulge the types of feed they give their BSFL, as they have often tried and tested numerous combinations to find what works for them and their system and what gives the best productivity and nutritional output. According to Barrett et al. [14], BSFL are typically fed a range of by-products, including dairy, vegetable and bakery by-products, brewers' or distillers' grains, and animal feeds. In a systematic review of the influence of different food waste-rearing substrates on protein composition in BSFL, Hopkins et al. [25] found a similar range of substrates being used. In particular, of the 23 articles they reviewed, 16 of them used grain-based ingredients for rearing substrates (e.g., distillers and brewery grains, spent barley, and wheat), 15 used fruit and vegetable ingredients, six used substrates that contained animal-based ingredients, four referred to a generic food or kitchen waste ingredient without giving specifics, and one article mentioned seaweed as an ingredient in the rearing substrate [25].

The industry is also seeking to expand into food surplus and animal by-products in the short term, and longer-term plans are in place to explore the use of municipal waste, manure, and slaughterhouse wastes [14]. Such substrates are not currently permitted in the EU, as insects are governed under the same legislation regarding feedstuff as other farmed animals (see section 2 for legislation and section 3.1 for a discussion on the use of waste streams).

One European company, 'Looop', specialises in creating 'co-products' that come from food and fermentation plants and can be used as insect feed. Their website lists 41 products suitable for insects, from variations of wheat starch to potato steam peels, but does not give any indication in regards to the welfare impacts of the feeds, only that they are certified and comply with relevant laws and regulations.

Neonate larvae may be fed a specialised, higher-quality diet to optimise survival, details of which are not always clear, although starter chick feed is known to be used [37,38].

4.3.1. Substrate processing

All substrates are processed initially to optimise their use for larval rearing, which can include numerous processes, e.g., blending multiple substrates, fermenting or inoculating wastes to improve nutritional value, longevity and decomposition rates, grinding substrates to reach an optimal size, dewatering or rewatering to create the correct moisture levels, and testing for harmful pollutants [34,37]. Processing is an important step, as it can potentially improve larval welfare.

In particular, processing can alter the substrate texture, pH level, moisture and nutrient composition, and microbe levels, which can directly impact comfort and survivability in the larvae [14]. For example, grinding rough substrates can make it safer for larvae by removing the risk of injuries from sharp edges and can make the substrate more porous and looser, which helps retain moisture and allows the larvae to move more freely [39]. Younger larvae also find ground food easier to consume [37,40].

Another form of processing is the addition of certain bacteria, such as *Bacillus subtilis*, which can breakdown hemicellulose and lignin in the substrate, which in turn improves its nutritional value [41,42]. Microbes can also be used to make them more appealing to the larvae, for example, by altering the pH [43]. However, the impact of processing substrates in this way is not fully understood regarding larval welfare, as most investigations focus more on the impact on production parameters such as growth [14].

Another form of processing is to mix substrates to improve their heterogeneity and nutritional value [14]. Plant-based oils, soybean curd residue, glucose, and restaurant wastes have all been tested in this respect [42,44–46].

According to Heinrich Katz, CEO of Hermetia Baruth GmbH and one of the founders of IPIFF, processing the feed given to larvae is critical (H. Katz, personal communication, 11 October, 2023. In particular, he confirmed that the substrate must be processed to ensure that oil is combined and bound into the particles, as otherwise, it can smother the larvae and prevent them from breathing. In his systems, he found that a particle size of no more than 3mm was optimum for feeding, along with a maximum of 8% fat content. He also adds natural additives at this stage to improve the nutritional content of the substrate (H. Katz, personal communication, 11 October 2023).

4.4. The welfare implications of the substrate diets used for BSFL

4.4.1. Natural variation in feedstuffs

Although BSFL may willingly consume a variety of substrates, not all substrates offer the same nutritional value or have the same effects in terms of growth and survival. For instance, the differences in nutritional composition of fruits and vegetables vary geographically and in accordance with the season and strain used. For example, dessert and juicing banana peels yield a 4.5-fold difference in larval weight [43]. Furthermore, different strains of BSF are even known to respond differently to the same substrates [47]. However, according to Katz, all of the EU's BSF come from his system and are, therefore, the same strain (H. Katz, personal communication, 11 October 2023).

4.4.2. High mortality rates versus high performance

High mortality rates are often accepted in the industry, providing that the final weight, biomass, or development time is optimum [14]. For instance, in a test of different combinations of vegetable and butchery waste substrates, researchers found that the 100% vegetable diet performed poorly in terms of production, compared with diets of mixed vegetable and butchery waste, as it resulted in the worst growing performance (live weight, length, thickness and growth rate) [15]. However, the 100% vegetable diet also resulted in the lowest mortality rates compared with the butchery and vegetable mixed diets, potentially rendering it more positive from a welfare perspective. Furthermore, using vegetable wastes also reduced the oxidative stress in the BSFL, further indicating enhanced welfare from feeding vegetable waste [15]. However, these mortality rates (10.61%) were far lower than recorded elsewhere for vegetable wastes (e.g., 23% in [48]), highlighting the impact of other factors.

Others have had more success with vegetable waste by adding horse and sheep manure to vegetable waste, which can improve survival rates by 11% and increase the final body mass of the larvae [17]. Therefore, the impacts of an all-vegetable waste diet are not fully clear. Furthermore, there may also be implications for the breeding adults, as adults reared on vegetable waste as larvae may have insufficient reserves for the pupa and adult life stages (see section 4.4.6.).

4.4.3. Working with incompatible substrates and waste streams

Despite the apparent flexibility of BSFL to feed on different substrates, not only do they show preferences for certain feed, but not all substrates are suitable for BSFL, as some can lead to high mortality levels. For instance, human faeces can result in mortality rates as high as 92% in some feeding regimes [49]. However, given the drive to utilise BSFL as waste processors, efforts have continued to find ways in which certain waste products can be adapted and processed to make them more viable.

One case in point is the use of meat meal, which is also associated with high mortalities, both at the larval stage (60%) and the pupal stage (80%) [20]. However, researchers have explored mixing vegetable waste into meat diets (butchery waste) to balance the resulting lipid and protein levels, which, although still results in higher mortality rates than a 100% vegetable diet, it has improved production value in terms of growth rate and weights [15].

Both dairy manure and chicken manure are also associated with high mortality rates (65% for chicken manure [50] and 45% - 77% for dairy manure [51,52]). However, mixing ~20% of dried food waste into chicken manure can improve survivability to close to 100% [50].

4.4.4. Commercial livestock feeds

Commercial chicken feed, which is considered a high-quality, high carbohydrate substrate for BSFL, may also be used in industry, as it results in fast growth and considerable weight gain in BSFL [53]. However, when degassed sludge (a waste stream) is added, which is higher in proteins and inorganic components, growth slows down as the larvae spend more energy on respiratory purposes [53]. Furthermore, larvae fed solely on degassed sludge do not survive to adulthood as they fail to emerge from the pupa [53].

4.4.5. BSFL preferences for substrates

As with all farmed animals, choice and control are highly limited for BSFL, and depending on the system, they may be fed only one type of mass-rearing diet. One study found that BSFL show preferences for different types of food, specifically pig manure, over the mass-rearing diet [54]. These findings were consistent across age and independent of the type of feed they were previously reared on. The preference for pig manure is likely to be because it is higher in microbes than other substrates (M. Barrett, personal communication, 11 October 2023). However, pig manure is not as nutritious as other substrates, so there may be trade-offs occurring in the larvae regarding microbial and nutrient content. Therefore, this needs to be considered in commercial settings, as the nutritional content of substrates can be improved through inoculation (M. Barrett, personal communication, 11 October 2023).

Furthermore, being prevented from accessing preferred feeds may cause undue suffering in insects, as homogenous diets remove the opportunity for choice and variety but also appear to not be suitable in terms of providing an adequate nutritional profile [14]. Moreover, in vertebrates, providing animals with a choice is considered a way to provide control and a chance for good welfare. If we apply the precautionary principle to insects, then this would mean that giving BSFL access to their preferred feed could have positive welfare benefits.

4.4.6. Impacts of BSFL diet on adult life stages

Whilst most of the BSFL in industry are slaughtered before they pupate, some are kept for breeding and will develop into adults. What the larvae are fed directly impacts the development of the larvae into adults and, potentially, adult welfare. In particular, the quality of food given to BSFL affects development time, pupa survivability, and certain biological traits in adults, such as female fecundity and wingspan [20].

In one study, larvae reared on meat meal took the longest to develop into adults compared with those reared on hen feed or a mixture of meat and hen feed [20]. This was thought to be because the meat meal was too dense and hard to consume, resulting in reduced intake. Furthermore, larvae and pupae mortality were highest (60% and 80%, respectively) on the meat meal diet, dropping to 7% (larval) and 1% (pupae) for the hen diet [20]. In terms of production, the hen feed diet was also more favourable in that the females had fully developed ovaries and larger eggs by day five after emergence, whereas those reared on the mixed diet did not fully develop until day 15 post-emergence [20].

Protein content in the larval diet has the main effect on development time in adult BSFs, but neither protein nor carbohydrate content appears to affect survivability [55]. In particular, development time is shorter when larvae are given a low-protein diet, which can mean that they do not sufficiently build up enough metabolic reserves for adult life, which is particularly relevant if adults are not fed (see sections 4.5-4.6) [55]. Protein also impacts body weight and fecundity, as larvae given diets high in protein (and carbohydrates) are larger both as larvae and as adults, and the adults have higher fecundity compared with those from larvae reared on a low protein and carbohydrate diet [55]. Conversely, the adult emergence rate is not considered to be affected by protein or carbohydrate levels in the larval diet, at least in the intermediate ranges [55].

4.5. Substrates typically used for BSF adults (BSF)

Adult BSFs are often described as non-feeders, relying upon fat stores from the larval stage for the adult phase, as they are thought not to have functioning mouthparts [56]. As a result, adult BSF are not typically given any food, although moist, rotting food may be provided to stimulate oviposition [14,33,56]. In fact, the family of solider flies (*Stratiomyidae*) actually have fully developed muscoid (sponge-like) type mouthparts and are described as nectar and pollen feeders [57]. Although adult BSFs can utilise stored fat reserves, they can still physically eat and will exploit resources in the wild [57]. In particular, BSF may colonise beehives, exploiting them as an energy resource for both adults and larvae [57]. Moreover, although adult BSFs may have difficulty absorbing solid particles, they can use salivary secretions to aid absorption [57].

4.6. The welfare implications of the diets (or absence of diet) given to BSF adults

As adult BSFs can reproduce without feeding, they are not typically fed in industry and are left to rely on the energetic and nutrient reserves they developed in the larval phase [23]. This means that adult BSFs could experience trade-offs as they must allocate resources and energy expenditure into either reproductive output or somatic maintenance to prolong their lifespan [23]. However, mated females tend to live longer than unmated females, which may result from the male's ejaculates containing an array of nutrients [58]. However, the opposite is the case for the males, as mated males have reduced longevity compared with unmated males due to the high energetic cost of copulation and courtship behaviour [23].

In a comparison of four diets: 1) no diet (control), 2) water, 3) a mix of sugar, bacteriological peptone and milk, or 4) agar with sugar, the female adult BSFs fed the agar or milk mix diets lived for 4-5 days more, compared with the control or water diets [57]. There was no significant effect on males, although water, agar or milk diets all led to an increased longevity of ~3 days compared with the control diet. Furthermore, both the milk and agar diets also increased the number of eggs the females laid by three-fold, compared with the water or control diets, partly because the period of oviposition was longer [57]. However, it is important to note that due to the biology of adult BSF, they would not likely have been able to fully access the agar diet, as they are unable to suck up a gelatinous substrate. Therefore, a liquid substrate could have even better results.

Macavei et al. [59] had similar results when they fed adult BSFs no food, water, or sugar water. Here, both the males and the females lived longer when fed sugar water compared with those fed water or nothing [59]. Similar findings were found in a small-scale laboratory setting, where feeding the newly emerged adults with sugar water increased the males' longevity by 3-fold and the females by 2-fold, compared with just providing water [60]. The differences were even greater when compared with no food (7-fold for males and 4-fold for females) [60].

Although, to date, research into adult diets has been focussed on longevity and egg laying, some pending studies have explored preferences in BSF adults. For instance, Wadell et al. have found that BSF adults have preferences for carbohydrates and proteins that are commonly used in the rearing of other flies, such as *Drosophila melanogaster* (personal communication, 14 October 2023). In particular, they have found strong preferences for 5% molasses (carbohydrates) compared with 10% molasses, 5 or 10% honey or dextrose) with preferences strengthening over age. As for protein, they found that the adults showed strong preferences for 5% heat-treated yeast, over 10% yeast, and 5 or 10% agar or casein). It is unsurprising that yeast is preferred by adults as yeast is the most prevalent protein source for insects in the natural world, and therefore, both species-appropriate and evolutionary relevant (M.Barrett, personal communication, 11 October 2023). In terms of ratios, their preferences have been found to be similar to other adult dipterans (1:4 and 1:8 protein to carbohydrate ratios) (Waddell et al., personal communication, 14 October, 2023).

As longevity is improved by feeding BSF adults, a lack of food may lead to the unpleasant experiences of starvation and frustration in these animals. In fact, according to Dr Barrett, Director of the Insect Welfare Research Society, the absence of feed for adult BSFs represents a significant welfare concern, and an absence of food results in them dying from starvation rather than being humanely culled (M. Barrett, personal communication, 11 October 2023). Furthermore, an anecdotal observation from Edward Waddell et al. (personal communication, 14 October 2023) is that the "BSF adults appear to gravitate towards the feeder location in the mating cage when food is provided". Such behaviour could provide additional evidence for the importance of providing feed to adult BSFs.

There is a clear evolutionary and biological benefit from feeding adult BSFs, and so the common misconception that adult BSFs are incapable of feeding is a harmful feature of the industry. Furthermore, most producers have trialled feeding adults only with sugar-water (H. Katz, personal communication, 11 October 2023), a pure carbohydrate diet. A pure carbohydrate diet can provide energy for copulation and flight, particularly for males, but female BSF require significant amounts of protein to produce eggs, so it is unlikely to provide significant benefits to female longevity and fecundity (M. Barrett, personal communication, 11 October 2023).

4.7. Recommendations for the black soldier fly larvae and adults

In addition to the overarching recommendations in section 3.2., the following apply to the black soldier fly larvae and adults.

- Substrates should be processed to an optimal size for the size of the larvae (e.g., 3mm) for consumption.
- Substrates should be processed to ensure any oils are bound into the particles to prevent suffocation of the larvae.
- BSFL diets should be heterogeneous and high in nutrients, containing a ratio of carbohydrates and proteins that are suitable for the life stage.
- The use of non-digestible substrate should be minimal, and at least 50% nutritional content in any substrate should be maintained. Substrates with large amounts of non-digestible matter should be inoculated with beneficial microbes to aid digestion (providing suitable evidence available to assess welfare risks.
- Microbial inoculation is likely to have favourable results, especially when processing has removed some of the nutritional content. However, further research is needed in this area.
- Certain substrates have been proven to have poor survivability and should be avoided unless further research can find ways to process and improve these substrates to promote welfare (e.g., meat meal, animal faeces, and dairy manure).
- Commercial feeds, especially when mixed with other substrates to form a heterogeneous diet, can be a favourable diet for BSFL.
- Vegetable waste can be a favourable diet for BSFL; however, additional protein may need to be provided (i.e., yeast), especially if the larvae are to be used for breeding purposes.

- BSFL intended for breeding should be given sufficient protein at the larval stage to ensure sufficient reserves are built up for pupation and the adult stages.
- Adult BSF must be fed and should be given diets comprised of both proteins and carbohydrates in liquid form to allow feeding, but in feeders that prevent drowning. Pending further research, the optimum ratios appear to be 1:4 and 1:8 P:C[1] (females and males). Optimum protein sources appear to be yeast, and optimum carbohydrates appear to be 5% molasses.

[1] P:C – Protein: Carbohydrate ratio

5. Common Housefly (*Musca domestica*)

The common housefly (*Musca domestica*) (CHF) is approved for livestock feed in the EU. As with the black soldier fly, the larvae are used for feed, and the adults for reproduction. The larvae are currently used in feed for fish, swine, poultry and pets, as well as for manure management [61].

5.1. Natural biology of the common housefly

The lifespan of the common housefly is between 20-30 days for all four stages, depending upon the conditions [62]. The larval stage takes between four to 13 days at optimal temperatures, and adults can live between 15-25 days. The female CHF can lay eggs up to five times, laying around 130 eggs each time [62].

5.2. Overview of farming systems for the common housefly

Farming systems vary for CHF, but more intensive systems rear the stock flies in sterile cages, each with over 750,000 flies [63]. Tanks or crates are filled with the substrate and kept moist to attract the flies to oviposit. The eggs or larvae are then removed and placed into a rearing area, where they are kept at specific temperatures and humidity levels to aid growth before being harvested just before they become pupae [33,63]. Some systems use natural oviposition, placing rearing beds in the open air with suitable substrates to attract adult flies [64].

5.3. Substrates typically used for CHF larvae

Poultry manure is commonly used for larvae, but other substrates, such as offal, pig or cattle manure, blood, wheat bran, or rotten fruit, may also be used or mixed in [33,65]. For instance, Koné et al. [64] found that chicken manure mixed with litter and water; sheep manure with fresh blood from an abattoir and water; and fish offal and water were the most successful substrates and that fish offal or blood added to chicken manure also worked well [64]. Note the use of many of these substrates is not permitted in the EU, and it appears that EU production of CFH larvae is not widespread as a result.

5.4. The welfare implications of the substrate diets used for CHF larvae

5.4.1. The impacts of different manure substrates

Although manure as a feeding substrate is not currently permitted in the EU, it is still a popular substrate for rearing CHF larvae elsewhere, as not only is it an attractive substrate for them, but the larvae are also useful as manure management agents [24]. However, the development and welfare on manure substrates can vary depending on the type used [24].

In one study performed under laboratory conditions, swine, poultry and calf manure produced significantly more adult flies, resulted in shorter development times, and produced larger and heavier pupae compared with the other types of manure used (cow, dog, goat, and horse, which were ranked second, and composted swine manure which was totally ineffective [66]. Furthermore, mortality rates were significantly lower with swine manure (2.5%), compared with all other substrates, apart from chicken manure (5% mortality). In particular, larval mortality was 17% on cow manure, 11% on calf, 46% on horse, 63% on goat, 23% on dog faeces, and 100% on composted swine manure [66].

Similarly, another study that tested eight different manures also found that poultry manure had the lowest mortality rate (9%), followed by calf (14%), dog (16%), buffalo (22%), cow (23%), sheep and goat (25%), and horse (30%) [67]. It is important to note that the mortality rates between studies differ despite using the same species' manure, which is due to the natural variation depending on diets given to the livestock species, as well as the effect of the environmental climate upon the larvae [24]. Furthermore, both studies were performed at a bench-top scale of 30 larvae; therefore, their results may not apply to large-scale commercial systems.

In a large-scale study involving tens of thousands of larvae, swine manure (which performs well in small-scale studies) resulted in 40% mortality [68]. Another study had mortality rates ranging from 10% to 40% on poultry manure, with mortality positively correlated with larval densities [69]. More recently, Miranda et al. [51] found that mortality rates were the lowest (26%) when larvae were fed the control diet (Gainesville diet), which was comprised of 50% wheat bran, 30% alfalfa meal, and 20% corn meal. In comparison, there was a 27% mortality rate from swine manure, 22% from poultry manure, and 50% from dairy manure [24].

Therefore, not only is it important to consider the differences in using substrates at small or large scales, but also the nutritional content of manures is likely to vary according to the diets of the livestock it derived from.

5.4.2. Importance of moisture content

Moisture content is considered an important component of substrate for CHF larvae and a reason why some manures are wholly unsuitable [24]. For instance, the upper moisture threshold for CHF larvae is reportedly between 70-80% on poultry manure [70]. In contrast, cattle manure has a moisture content of 85%, which may be why survivability was so low (50%) in Miranda et al.'s study [24]. Conversely, the swine and control diets in the study had moisture content levels of 72% and 70%, respectively, which are within the acceptable range for CHF larvae, and so may be why they had higher survivability rates (73% swine and 74% control) [24].

5.4.3. Importance of nutrient content

Moisture content is not the only factor influencing survivability and development in CHF larvae, as the nutritional value of the substrate also plays an important role [67]. CHF larvae nutritionally require live bacteria and will choose to live in microbe-rich environments, living amongst and ingesting bacteria [71]. However, not all microbes are digestible, and the microbial communities are also affected by the nutrient composition of the manure itself [24,71]. In their study, Miranda et al. [24] found that swine manure (which incidentally had 73% survivability) had nearly twice as much nitrogen as in the control diet (74% survivability), poultry manure had over five times the amount of phosphorus and three times as much potassium (67% survivability), and dairy manure had over twice as much potassium (50% survivability), compared with the control diet. However, they suggested that excess nutrients may, in fact, be detrimental to development or indigestible by the larvae [24]. It is likely that these differences in the manure composition also led to variations in the effects on the larvae's development and survivability, but further research is needed to explore this.

5.4.4. Preferences in CHF larvae

No research has been performed into the preferences of CHF larvae for substrates, but based on findings from the black soldier fly, it is likely that they will also show preferences [72].

5.5. Substrates typically used for CHF adults

As adult CHF are considered pests, there is little research regarding their diet and welfare in the scientific literature, as most of the focus on diet is from a pest control perspective [72]. However, both small and large-scale studies refer to keeping the adults on mixtures of sugar and milk powder, along with a water source [e.g., 24,62,66,67,73,74], and this appears to be the convention in commercial systems, too [75].

5.6. The welfare implications of the substrate diets used for CHF adults

The use of milk and sugar is an expensive feed, and so alternatives have been explored. For instance, other foods (i.e., pineapple, papaya, banana, and honey) have been studied to determine their viability as replacements [75]. The honey diet led to the lowest mortality rate (27% in 10 days), compared with 37% for the milk and sugar diet and 57-62% for the fruits. There were minimal differences in egg hatching, pupation and emergence rates [75]. Therefore, in terms of welfare, it is likely that the honey diet has the most potential, as evidenced by the lowest mortality rate, although there is scope for improvement given that the mortality rate was 27%. However, given the focus of the industry is on production, it is worth noting that the study found that pineapple, whilst causing high mortality rates, was comparable with the honey and the milk and sugar diets in terms of fecundity [75]. The use of pineapple is unlikely in European insect farms due to the cost of importation, compared with the study's location in West Africa, where pineapples were a cheap substrate. However, similar results may be found with local fruits, which may have a similar effect in terms of high mortality rates and fecundity, which could be a welfare concern. Furthermore, as seen with the BSF, milk-sugar diets or fruit diets may not provide sufficient protein for the females, and therefore, research should explore the provision of yeast on the welfare and production parameters.

5.7. Recommendations for the common house fly

Note. It is likely that some of the recommendations for BSF are relevant here, but the following recommendations are restricted to what can be derived from the scientific literature.

- Further research is needed to determine the effects of providing CHF larvae with a heterogenous diet compared with a homogeneous diet.
- Chicken and swine manure and the Gainsville diet have all had positive effects on larvae survivability compared with other diets. However, none of them achieved optimum survival rates of >95%, and other factors such as densities and environmental conditions play a large role.
- Research is needed into the preferences of CHF larvae to determine whether they can self-select more suitable diets for survivability and welfare.
- Adult CHFs should be fed either a honey diet or a milk-sugar mix (1:1 ratio) unless research indicates another suitable substrate that results in higher survival rates.
- Adult CHFs require a water source in addition to dietary and oviposition substrates.

6. Yellow Mealworms (*Tenebrio molitor*), Lesser Mealworms (*Alphitobius diaperinus)* and the Adult Darkling Beetles

In the EU, the yellow mealworm (*Tenebrio molitor*) and the lesser mealworm (*Alphitobius diaperinus*) are approved for feed for aquaculture, pigs and poultry and human consumption, with the lesser mealworm being approved in January 2023.[2]

The lesser mealworm (LM) and the yellow mealworm (YM) are larvae from beetles from the darkling beetle family. As they come from the same taxonomic family, there are similarities in production and dietary needs, so they are grouped into one section.

[2] EUR-Lex - 32023R0058 - EN - EUR-Lex (europa.eu)

6.1. Natural biology of yellow and lesser mealworms and the adult darkling beetles

Depending upon the nutritional and abiotic conditions, a yellow mealworm can take anywhere from 2 months to 4 years to go from egg to pupa [18]. The adult beetles can live for another 1-6 months [18]. Females may lay five to eight eggs daily and about 300 in their lifetime [76].

The lesser mealworm can take 6-14 weeks to develop into adults, and the adult beetle can live for 3-12 months. The female adult lays, on average, 200-400 eggs in her lifetime at a rate of 1-5 a day [77].

Adult beetles are flightless, nocturnal, and photophobic and will naturally live in the substrates where they reproduce [78,79].

6.2. Overview of farming systems for yellow and lesser mealworms and the adult darkling beetles

Mealworm larvae are typically raised in large plastic bins or stackable crates, insulated to maintain the heat generated by the mealworms [29]. Larvae intended for breeding are typically reared separately [80,81]. The adult beetles are usually kept in large bins or crates, and the eggs are removed and placed into separate bins for larvae rearing [29].

6.3. Substrates typically used for yellow and lesser mealworms

YM generally consume decaying grains but will also eat cereals, meal scraps, bread, dead insects, feathers, live conspecifics, decaying wood, and grains mixed with animal excrement [18].

In commercial systems, YM's are fed diets comprising 70-85% carbohydrates, typically wheat bran, and sometimes added protein, fruits, and vegetables to provide moisture and micronutrients [18,82]. Wheat bran is an expensive rearing substrate, which can limit its use [82]. Consequently, mealworms are increasingly reared on mixed-grain diets, which are already considered safe for food and feed [83]. However, there are concerns that this will lead to competition in the future, so there is a push to focus on using side streams [83].

6.4. The welfare implications of the substrate diets used for mealworms

6.4.1. Homogenous diets lead to suboptimal levels of macronutrients

Inadequate nutrition for both larvae and adult beetles was identified as a significant concern in a review by Barrett et al. [18]. In particular, homogenous diets can fail to supply appropriate macronutrient ratios, which, in turn, may lead to an increase in cannibalism among conspecifics [18,77]. Both LM and YM are cannibalistic and will even collectively attack and kill small vertebrates if their preferred foods are unavailable, and cannibalism is a major cause of loss in production [77,84,85].

Wheat bran is the most common component of mealworm diet in commercial systems, and wheat-bran-only diets are commonly seen [18,86–88]. Compared with other homogenous diets (i.e., flour, maise flour, lucerne, and oats), wheat bran has the highest survivability rate in YM, as well as the highest individual weight and fecundity from adult females [82]. However, although wheat bran contains all the nutrients needed for mealworm growth, they are only in suboptimal proportions [79,86,89]. Therefore, despite its popularity in the industry, a wheat bran-only diet results in longer development times and higher mortality rates for mealworms compared with mixed diets [89,90].

Conversely, heterogenous diets comprised of cereal (typically wheat bran), protein (such as soy protein or beer yeast), and a water source (typically vegetables like potatoes) result in increased survival rates in both LM and YM, compared with homogenous diets [18,90]. As a result, many scientific studies recommend including fresh feed, such as chopped vegetables and yeast, which offer a source of water as well as additional nutritional value [e.g., 86,87,91–93].

6.4.2. Balancing macronutrient compositions in the two species of mealworms

Balancing the macronutrient compositions is an important challenge in industry, as small differences can make significant impacts, and there are also key species differences. For instance, in YM larvae, the addition of protein improves pupal weight gain, reduces total development time, and significantly increases survival rates [90,94,95]. In particular, a high-protein and low-starch diet (i.e., maise DDGS (maise distillers' dried grains with solubles), beer yeast, bread remains, and spent grains) results in a survival rate of 92% (compared with 71% and 85% in the control diets), reduces the development time from 117-123 days down to 95 and increases pupal weight from 0.144-0.149g to 0.161g [90].

Conversely, LM larvae respond differently to higher protein diets, with improvements primarily observed in survivability and development time, while other production metrics remain unaffected [90]. Furthermore, a low-protein, low-starch diet composed of beer yeast, bread remains, and spent grains produces the highest LM survival rates, elevating them from 79-82% (as observed in the control or commercial diet) to 97% [90]. Additionally, compared to one of the control diets, the low-protein and low-starch diet significantly reduces LM's development time, although it does not influence pupal or adult weight [90]. However, this experimental diet was freeze-dried first, which is not economically viable on a large scale [83].

Overall, Barrett et al. [18] suggest that yellow mealworms require a minimum of 20% protein in their diet for optimum nutrition and that this is to increase to around 40% minimum for the later instar[3] stages [18]. It is not clear whether this can also be applied to the lesser mealworms.

Another macronutrient of importance is fat, as high-fat diets appear to perform the best in terms of survival rates when combined with high amounts of protein [18,96], and the opposite occurs if combined with low protein levels, highlighting the critical importance of balancing the macronutrients and the species-specific needs [18,96].

[3] Instar refers to the developmental stage between moults. The first instar is the larvae after hatching from the egg, the second comes after the first moult, and so on until pupation.

6.4.3. The potential of side streams

Gianotten et al. [83] explored the impact of side-stream diets that had not been freeze-dried, as would be the case in commercial systems. They found that LM can be successfully reared on both single-side-stream diets and mixtures of side-streams [83]. In particular, the most economically viable and productive diets were considered to be wheat middlings and rapeseed meal, and a mix of both, along with brewery grains (for moisture), resulted in the best growth rates [83]. The researchers concluded that there was potential to use agri-food side streams, such as rapeseed meal and wheat middlings, for mealworm feed without adverse effects and with good production rates. The researchers found that doing so also offered the potential to reduce production costs and subsequently reduce competition over other feed applications [83].

6.4.4. Mealworm preferences

Despite the focus in the industry on rearing mealworms on unsuitable homogenous diets, there are production benefits to rearing mealworms on heterogeneous diets, and the larvae themselves show a distinct preference for variety. For instance, when they can self-select, YM preferentially consumes a 4:1 ratio of wheat bran: dry potato flakes, which results in the shortest development time and the second-best population growth (compared with other ratios) [97]. Another study found that YMs prefer 20% egg protein diets over soy protein diets or those that mix salmon, peanut or canola oil with soy and egg protein [94]. However, further research is needed to test their preferences in relation to yeast, which is the most commonly reported source of protein used with YML [79].

6.4.5. Accessibility of different feeds

As mealworms become more mobile as they age, the placement of food is an important consideration, as younger larvae may be unable to access all dietary resources if they are not within easy reach [18]. Younger larvae may also struggle with consuming large particles if they are not ground to a suitable size [18].

6.4.6. Feeding mealworms plastics and polymers

Mealworms are known to be able to survive and reproduce on diets comprised entirely of polymer [98–100]. However, there are significant welfare concerns associated with these diets, including slow growth rates [101], higher mortality and cannibalism rates [102–105], and increased metabolic stress [106] as a result of malnutrition. However, researchers have successfully included bran additives to increase the nutritional value, which increases survival and development rates [103,107] and reduces cannibalism [105].

6.4.7. Processing foods to reduce infection risk and improve nutritional value

When mealworms are fed food scraps, kitchen discards, and local feed sources, they have a higher risk of infection from protozoan fungal pathogens compared with those fed fresh produce or specialised feed [108]. Therefore, there are health and disease risks associated with the types of feed given to mealworms, with waste foods being associated with a higher risk of disease outbreaks [18].

Mealworm feed can be heat-treated to around 60-90°C to significantly reduce *Gregarina* spore viability and, therefore, the risk of infection from them [109]. Heat treatment can also inactivate Cry toxins from *B. thuringiensis* [110], as well as viral, fungal and bacterial diseases, and the presence of parasites [18]. However, heat treatment also kills the beneficial microorganisms in the feed, so it is recommended that producers consider inoculating heat-treated feed with beneficial microbes [18]. Heat-treating mealworm feed may also have positive impacts down the food chain for those consuming the mealworms, whether that’s livestock or humans, although this requires further research and investigation.

There is limited research in this area, but there are indications that some probiotics used for vertebrates could improve mealworms' welfare and production parameters [18]. For instance, studies show increased larval weight gain [111,112], reduction of growth and presence of bacterial pathogens [111], and potentially antibiotic or antifungal properties, as seen through the increased survivability of YM infected with pathogenic fungus following the consumption of probiotics [113].

6.4.8. Naturally occurring mycotoxins

Mycotoxins are naturally occurring toxins produced by certain moulds and can be found growing on a range of crops and foodstuffs, including cereals [114]. Mycotoxins in naturally occurring contaminated grains, such as those fed to mealworms, are often higher than when tested in laboratory conditions [115]. High concentrations can be particularly toxic to mealworms [116] and even more so when there is more than one mycotoxin [117]. Furthermore, most of the studies exploring the effects of mycotoxins on mealworms are conducted on later-instar larvae, whereas the early-instar larvae are thought to be more susceptible [118]. Therefore, in lieu of further research, these feeds cannot be considered safe for early-instar larvae [18].

6.4.9. Access to water sources

There are multiple benefits for both welfare and production, from giving mealworms access to water sources, such as vegetables, fruits, hydrated gels or sprays [81]. In particular, providing a source of water has been shown to result in increased growth, survival rates and body masses in YM [96,119–121], decreased development times, and increased adult eclosion rates and longevity [119–121]. The provision of water becomes more important in environments with low to moderate relative humidity [120] but has important benefits across all environments and enhances the benefits of appropriate nutrition across different strains of mealworms [96].

6.4.10. Impacts of mealworm diet on adult life stages

When mealworms are provided with protein, this also impacts the adult life stages, as beetles given protein as mealworms show increased egg production (from 3-4 eggs/day to 6-7 eggs/day) [94]. Consequently, 40% protein in diets is considered to be optimum for late-instar mealworms intended for breeding [18].

6.5. Substrates typically used for adult darkling beetles

The adult beetles are typically fed wheat bran, which they also use as an oviposition substrate [18]. Supplementary protein sources are infrequently reported [18].

6.6. The welfare implications of the substrate diets used for adult beetles

The composition of macronutrients in the adult beetle's diet is also important, as it can directly impact the lifespan of the beetle and their reproductive performance [18]. For instance, males fed a 1:1 (P:C) ratio and females with a ratio of either 1:1 or 1:5 have the longest average lifespan [122]. The typical wheat bran diet given to adults in commercial systems only provides a P:C ratio of between 1:3.1 and 1:4.3, a significant deficit in terms of the optimum ratio required for longevity and, thereby, welfare [18].

6.7. Recommendations for yellow and lesser mealworms

- Diets should be heterogeneous, allowing the mealworms to perform natural nutrient self-selection behaviours.
- Diets should contain supplementary protein (e.g., soy, yeast, or egg), increasing in proportion as the mealworms age.

- Substrate and foods should be suitably spread throughout the mealworm enclosure to accommodate for the limited mobility of younger mealworm larvae.
- The feeding of polymers and plastics should be avoided, and if used, then substantial dietary additives should be included to provide the required nutrition for optimum welfare.
- Waste food and kitchen scraps should not be used, given the heightened risk of infection from fungal pathogens.
- Mealworm feed should be heat-treated (to 60-90°C) to reduce viral, fungal, and bacterial infections. However, as heat treating kills the beneficial microorganisms, heat-treated feed should be inoculated with beneficial microbes to promote nutritional value.
- Diets should be supplemented with appropriate probiotics.
- Mycotoxin-contaminated grains should not be fed unless diluted to levels known to be safe for mealworm consumption and should be avoided for all early-instar mealworms.
- Continuous access to water must be given, and within 5cm from each individual, to allow for the limited mobility of younger mealworm larvae.
- Late instar mealworms, and especially those intended for breeding, should be given higher proportions of protein (e.g., 40%) to ensure higher welfare (and production) in the adult beetles.
- Adult darkling beetles should not be fed a homogenous diet of wheat bran but should be supplemented to ensure a ratio of 1:1 (P:C) for males and either 1:1 or 1:5 for females.

7. Banded Crickets (*Gryllodes sigallatus*), House Crickets (*Acheta domesticus*), and Field Crickets (*Gryllus assimilis*)

The banded cricket (*Gryllodes sigallatus*), the house cricket (*Acheta domesticus*), and the field cricket (*Gryllus assimilis*) are all approved in the EU for use as livestock feed, and the house cricket is approved for human consumption.

7.1. Natural biology of the banded, house and field crickets

Crickets hatch out of the egg as nymphs, which look like small versions of adults but without wings and their full reproductive capacities. As the nymphs grow, they shed their exoskeleton in moults, which can occur 8-10 times before adulthood [123]. Adult crickets have fully developed wings and reproductive systems.

7.2. Overview of the farming systems for crickets

Crickets are often reared in plastic bins, with cardboard materials (rearing matrices) inside to provide more surface area and a place for females to oviposit [29]. There are usually damp sponges in the containers to create humidity whilst preventing drowning [123]. The rearing process is typically comprised of one or two stages; in one-step systems, the ‘pinhead’ crickets (newly hatched nymphs) stay in the same unit until harvesting, whereas the two-step approach separates the crickets at instar four or five to reduce density [124]. Feed is usually provided in trays at the top of the rearing matrix or in reservoirs accessible through a mesh screen [124,125].

7.3. Cricket diets in commercial systems

The diet of wild crickets is not fully understood [126], but they are thought to prefer plant material, including fruits, vegetables, seeds, grasses, and fungi. They are considered omnivores, as they will also occasionally scavenge on dead insects [127–129] and show cannibalistic behaviour [130].

Farmed crickets are often fed commercial livestock feed, such as chicken feed [131–134]. Commercial cricket feeds are also being developed worldwide, and the ingredients vary according to relevant legislation. For example, EU cricket diets do not have animal-derived ingredients besides fish, milk and eggs [135]. In general, specially developed cricket diets are comprised of ingredients like wheat middling, corn meal, oats, brewer's yeast, soybean meal, molasses, beet pulp, whey, plant oil, alfalfa meal, along with salts, vitamins and minerals. Animal products such as fish meal, fish oil, porcine and bone meal, and animal fat may also be added [29].

Crickets are typically starved or given only fresh vegetables for the last 24-48 hours before slaughter to clear the digestive system, improve taste, or save money [10,128,136–138].

7.4. The welfare implications of the commercial diets for banded, house, and field crickets

7.4.1. Preferences in crickets

Although crickets can be reared on various diets, they show clear preferences, which can have welfare implications during farming [32]. Furthermore, their preferences change throughout their lifespan. For instance, when given a choice, adult house crickets opt for rice bran, corn, buckwheat, and dry cabbage from a selection of feedstuffs [139]. In comparison, juveniles have the following order of preferred foods: yeast, followed by soybean meal, animal liver powder, and lastly, wheat middling [140]. The preferences of crickets for different feeds may be determined by different factors such as nutrient composition and ease of consumption, but as control and choice are important considerations in welfare, recognising and responding to these preferences can be a valuable contribution to welfare [32].

7.4.2. Dietary restriction

Dietary restriction may be deliberate (i.e., feed withdrawal before slaughter or on a long-term basis to save costs) or incidental (i.e., through inappropriate feeding techniques, such as providing food particles that are too large or via improper dietary composition). Dietary restriction can have multiple welfare impacts on crickets, including increasing mortality rates, reducing fitness and growth, and increasing the chances of cannibalistic behaviours [130,140–142]. For example, in juvenile field crickets, two days of ad libitum feed, followed by two days of no feed, resulted in lower-quality mating songs as adults, which is thought to indicate poor body condition and smaller size [142]. Whereas in adults, dietary restriction significantly decreases body condition in males [142] and decreases energy allocation to reproductive development in females [143].

Providing a varied and nutritionally balanced diet for crickets is vital for their welfare, and diets that utilise only a few agricultural by-products can often result in dietary restriction, leading to poor welfare outcomes [31]. For instance, crickets fed on diets mainly composed of straw have mortality rates of <99% [132]. Inappropriate nutritional composition in cricket diets can have significant welfare implications as a result of inappropriate macro or micronutrient provisioning [141].

7.4.2.1. Macronutrients

When house crickets are given a heterogenous diet and can self-select, the juveniles eat around 30.7% protein, 58.7% carbohydrate, and 10.6% lipids [139]. Studies have also found that juvenile house crickets require at least 20-30% protein, 32-47% carbohydrates, and 3.2-5.2% fat for optimal growth and survival [140].

In comparison, commercial livestock feed, such as chicken feed, has a far lower protein level of between 15.2 – 22% [131–134]. Whereas specialised commercial cricket diets typically have a low protein content of less than 20% [125], despite this being below the recommended range for growth [140] and below what is preferred by house crickets [134]. Although increasing the protein levels of cricket feed may increase costs, there are clear welfare advantages in doing so and significant implications in providing inadequate levels.

Although providing the right protein level is important, balancing protein levels with other macronutrients, such as carbohydrates, is also key. Furthermore, there are trade-offs when diets are high in protein, as female house crickets fed high-protein diets (3P:1C) will lay more eggs over their lifetime but will also die younger as a result, compared with those on lower-protein diets (1P:1C) [129]. In males, high protein diets result in faster development, larger body sizes, and early reproductive investment, and high carbohydrate diets result in longer-term reproductive investment and increased longevity [144,145].

7.4.2.2. Micronutrients

Micronutrients also contribute to cricket health and welfare, although little information is reported on industry practices [32]. As with macronutrients, self-selection behaviour provides valuable indications. For instance, crickets prefer feed sources high in vitamin C, sterol and manganese [139], and these micronutrients are also known to correlate with increases in biomass in house crickets [139]. Vitamin B-12 also appears to be important for cricket welfare, yet it is hard to consume from a plant-only diet. However, fresh and dried pumpkin are good sources and are preferred by crickets over a 16% protein chicken feed [146]. Phosphorus may also be of importance, not only to health but also for behavioural expression [32]. In particular, house crickets reared on a diet higher in phosphorus (1%) have better body condition and mass [147], the females lay more eggs [148], and the adult males signal more frequently [32], fulfilling an important instinctive behaviour, compared with those on a diet with 0.2% phosphorus. In comparison, commercial cricket feeds typically have a phosphorus content of between 0.72-0.84% [149,150], which may be suboptimal. However, the benefits of phosphorus supplementation vary across species [151], so further research is needed [32].

7.4.3. Potential welfare risks associated with dietary contaminants

There are multiple risks associated with cricket feeds that have yet to be fully explored in terms of the welfare impacts. For instance, mycotoxins (secondary metabolites produced by fungi) may cause both welfare concerns and human safety issues, but research is lacking, and exploration into the risks is considered to be an urgent matter [32,116,133].

Pesticide residues are known to be an issue in crickets, as evidenced by their presence in end-products such as cricket flour [30] (see also section 3.1.5.). Sublethal levels of the pesticide malathion change the natural behaviour of house crickets, making them more active and decreasing their use of shelters [152]. In vertebrates and some other invertebrates, such a behaviour change is considered evidence of pain and discomfort [153–155], and therefore, the same possibility should be considered for crickets [154].

Non-lethal levels of malathion also cause changes in behaviour, such as increasing non-directional movements (i.e., grooming, twitching and convulsing), compared with the controls [152]. The twitching and convulsing seen in these insects could be a critical indication of poor health, given the negative impacts such behaviour has on fitness. Furthermore, self-grooming in insects is a protective behaviour typically performed to remove parasites and pathogens, and when vertebrates perform protective grooming, it is considered an indicator of pain [155]. Moreover, an increase in grooming behaviour is also an indicator of negative emotional states [156].

Therefore, applying the same consideration to crickets would suggest that they find both sublethal and non-lethal levels of malathion to be aversive and result in negative states such as pain. However, further research is needed, as there are also some indications that sublethal doses of some pesticides can positively affect cricket welfare for those individuals experiencing other stressors, such as suboptimal diet and temperature, although findings to date are mixed [32].

The literature also indicates that microplastics could cause welfare issues in crickets, as microplastic fibres reduce growth in females [157]. Given that European agricultural land is estimated to be one of the largest reservoirs of microplastic pollution globally [158], the likelihood of microplastics entering the food and feed crops used for crickets is high [159]. Therefore, the welfare impacts of microplastics on crickets deserve further attention.

Research into the bioaccumulation of heavy metals in crickets is another area that needs attention [160]. The issue is complex as the prevalence and concentration of heavy metals are impacted by different farming and processing techniques, and producers do not readily report their levels [161,162]. Some insect species can protect themselves against the negative effects of heavy metals [163], but research is needed to determine whether this is the case for crickets [32].

Pharmaceutical drugs have been found in samples of crickets, and although they were below the legal residue limits, it is unclear what impact these drugs may have on the welfare of the crickets [32,164].

7.4.4. Implications of the types of diets typically used in industry for crickets

7.4.4.1. Waste streams and by-products

As with other insect species, there is a significant drive to utilise cost-effective, sustainable diets such as by-products from agriculture for crickets [125,137]. This can be a positive move for cricket welfare, providing that consideration is given to the macronutrient ratio and that high-quality food waste ingredients are used [32]. For instance, Morales-Ramos et al. [139] found that a diet composed mainly of agricultural by-products was economical, in line with the self-selected macronutrient ratio for the house cricket, and resulted in high growth rates [139]. However, poor-quality waste ingredients such as unprocessed post-consumer organic waste, straw from wheat, chicken manure, weed plants, maize or rice, can all have poor welfare outcomes for crickets, including very high mortality rates and poor development [132,137].

7.4.4.2. Commercial livestock feed

Given its homogenous nature, commercial livestock feed is unlikely to offer ideal welfare outcomes for crickets, as it does not allow crickets to self-select [32,165]. However, some producers do supplement these diets with other ingredients, including agricultural by-products, fruits and vegetables, fish meal and grains [136,137,166–169]. Not only does this improve the heterogeneity of the diet, allowing the crickets to self-select their optimal nutrition for their life stage and sex, but providing choice may also instigate consummatory pleasure if the crickets are given their preferred feed [170]. However, supplemented diets can only improve welfare if they provide the right ratios of macronutrients (see section 7.4.2.1.), as too low levels in protein, for example, may result in cannibalistic behaviours, which are detrimental to welfare [32].

7.5. Recommendations for house, banded, and field crickets

- Dietary restriction of any form should not be practised due to the negative welfare impacts it causes. If required before slaughter, feed withdrawal should not occur for longer than 24 hours, although research is still needed into the welfare effects of fasting for this period.

- The nutritional composition of diets for house crickets at different life stages is understood and, therefore, should be adhered to. Research is still needed for banded and field crickets.
- Research is needed into the impacts of the different feed contaminants outlined in this section.
- Diets should contain sources of micronutrients known to be important to cricket welfare, i.e., vitamin C, sterol, manganese, phosphorus and B-12.
- High-quality agricultural by-products appear to be a viable feed source for house crickets.
- Commercial livestock feed should not be used without supplementation to improve the heterogeneity and nutritional composition of the diet.

8. Migratory Locust (*Locusta migratoria*)

The migratory locust (*Locusta migratoria*) has recently been authorised in the European Union as a novel food. It is the adult locust that is harvested for consumption.

8.1. Natural biology of the migratory locust

The life cycle of a migratory locust includes four stages: egg, nymph, fledgling, and adult. Female locusts lay up to 30-60 eggs, which hatch as nymphs and undergo five moults. After the fifth moult, the soft and fragile fledgling locust emerges. Fledgelings feed extensively on green plants for around two weeks, after which they are considered adults. The adults are mostly migratory and continuously feed. The total life expectancy of adult locusts is about 8 to 10 weeks [171].

The migratory locust undergoes a process known as density-dependent 'polyphenism', which means when the population density is low, locusts behave as solitary individuals. However, when the population density becomes high, individuals undergo physiological and behavioural changes and behave gregariously, banding together in swarms of adults [172–174].

8.2. Overview of farming systems for the migratory locust

Locusts are the second most commonly consumed insect by humans, after crickets [173]. In many countries, wild locusts are widely collected during outbreaks [172]. When farmed, migratory locusts are reared in controlled, indoor systems. The adult locusts are mated, and the eggs are separated to allow the nymphs to be grown under monitored temperature and humidity conditions in stainless steel containers [175]. Adults are separated from the substrates and faeces at about 3–5 weeks old to be harvested.

8.3. Migratory locust diets in commercial systems

Migratory locusts are generalist herbivores and eat a plant-based diet [173,176,177]. Artificial diets have successfully been used within laboratory conditions for a long time. For example, in 1960, Dadd developed a successful artificial diet comprised of cellulose, sucrose, dextrin, a salt mixture, cholesterol, linoleic acid, casein, peptone, egg albumen, ascorbic acid, and ten water-soluble vitamins of the B complex. Indigestible material in the form of cellulose was also found to be vital for proper growth [178]. This artificial diet is still often used today (see 179–181].

8.4. The welfare implications of the diets used for migratory locusts

8.4.1. Dietary needs at different life stages

Locusts have different preferences depending on whether they are in the gregarious or solitarious phase, which may be triggered in farming conditions depending on the stocking densities used. For instance, desert locusts (*Schistocerca gregaria)*, locusts in the gregarious phase tended to eat a wider variety of plants compared to when they were in the solitarious phase [182]. Moreover, when desert locusts were given a diet with an unbalanced protein-to-carbohydrate ratio, the gregarious locusts were more likely to eat the artificial diets, despite their nutrient imbalances, compared to solitarious locusts [183]. Also, when they were fed a diet high in carbohydrates and low in protein, gregarious nymphs showed poorer survivability compared to solitarious insects [183]. The researchers suggested this could be due to them having evolved to trade-off the cost of eating an excess of a nutritionally unbalanced diet against the probability of encountering foods of complementary composition in the future. And so, because solitarious locusts are less mobile and live in environments with few host-plant species than their gregarious counterparts, they tend to opt to minimise nutritional error rather than maximising nutrient intake [183].

8.4.2. Contaminants

According to the safety report by the EFSA [175], no antimicrobials, veterinary medicinal products or solvents are reported to be used during the entire production process of migratory locusts [175]. Although, as discussed in sections 3.1.5. and 7.4.3, these contaminants may enter through agricultural sources.

8.4.3. Nutritional deficiencies impact immune function

Migratory locusts are herbivores and prefer carbohydrate-rich diets [173,176,177]. However, overall dietary nutrient levels have been found to impact their immunity and deficiencies in locusts cause weaknesses in immune function [184]. In particular, locusts are more susceptible to the Beauveria fungus when dietary protein is inadequate and show less anti-bacterial activity when the levels of carbohydrates are inadequate [184]. Reports of microbial contamination in farmed locusts have previously been attributed to uncertified insect feed substrates [172,185], but the issue can be exacerbated by crowded conditions in farms, which render the locusts more susceptible to disease [172,184,185].

8.4.4. Welfare implications of typical diets

8.4.4.1. Artificial diets

Migratory locusts have high energetic demands and tend to perform better on carbohydrate-rich diets [173,176,177]. In particular, diets high in carbohydrates allow locusts to store fats, resulting in overall improved flight performance [180]. The artificial diet given to locusts in farms may be able to meet their needs if carefully balanced, as wild juvenile locusts relocated to laboratories have been found to prefer an artificial diet comprised of 54% cellulose and 4% vitamins and salts, compared with the local plant diets [177]. The artificial diet also resulted in their survivability rate doubling on average, compared with those fed the plant diet under laboratory conditions [177].

8.4.4.2. Plant diets

Although less likely to be used in industry, plant diets may still feature in some parts of the industry and represent a more natural alternative to the artificial diet more commonly offered. In comparisons of different food plants on the growth and development of Oriental migratory locus (*L. migratoria manilensis*), maize leaves have the highest food utilisation rate, highest weight gain, and lowest mortality rate, followed by goosegrass, soybean leaves, and pakchoi [186]. Plants of the family Gramineae (grasses) are often the primary hosts of locusts in nature and, therefore, can be considered the most suitable food plant for locusts, with promising outcomes in terms of growth performance compared to locusts fed on Dicotyledon (flowering plants) [186].

8.5. Recommendations for the substrate diet used for migratory locusts

- Migratory locusts are generalist herbivores and should be fed a wide variety of plants, preferably wild grasses found in their wild habitat.
- When considering the plants used as feed, attention should be paid to the carbohydrate-to-protein ratio of the plant species.
- While migratory locusts can be fed artificial diets, fresh plants should be fed at crucial times of growth.
- More research directly comparing the welfare implications of artificial diets and fresh plants on migratory locusts is needed. Research to date has focused on the chemical composition of migratory locusts in terms of human consumption, with little research focused on the welfare implications for the animals of various substrates used in production.

9. Western Honeybees (*Apis melifera*)

The Western honeybee (*Apis melifera*) is being considered for human consumption in the EU and potentially as animal feed in the future. In particular, it is the honeybee drone brood (*Apis mellifera* male pupae) that is currently being proposed as a novel food.

9.1. Natural biology of the Western honeybee

The Western honeybee naturally lives in colonies in beehives constructed with numerous hexagonal chambers. The Western honeybee's life cycle consists of four stages: egg, larva, pupa, and adult. The queen bee lays eggs, and once they are hatched, the workers feed the larvae. After about six days, the larvae begin the final stage of pupation in a capped or sealed cell. The duration of pupation varies depending on whether the larva will be a drone (15 days), worker (12 days), or queen (7 days) bee; the bees then emerge from the cells sometime after eclosions [187,188].

9.2. Overview of farming systems for the Western honeybee

Current industry practices for farming honeybees or apiculture involve the keeping of honeybees in artificial hives of various designs in open-air systems [189]. In general, these artificial beehives are upright constructions with different segments. The inside boxes often contain an artificial, wax-coated foundation where bees can deposit honey or grow young [189].

Drone broods are produced in surplus in beehives during honey production. The main purpose of the male drone brood is to mate with young queens to maintain the reproduction of bee colonies. In many countries, farmers will discard naturally occurring drone brood pupae as they do not produce any products such as honey and, therefore, have no commercial significance. Also, removing the drone brood from hives can help control bee parasites of the genus Varroa, which prefer the drone brood [187,190,191]. Once the larvae begin to pupate, the drone brood are capped by worker bees within beeswax cells of combs built by the bee colony. It is the pupae drone brood that is being considered edible in the EU [192].

Honeybees naturally respond to changes in the ratio of pollen supply and protein demand in the hive and will vary what type of bee is produced accordingly (e.g., worker, drone) [190]. Drone brood production requires a good pollen supply to meet their protein demand. Drone larvae are more costly for bees to produce and maintain, and so colonies regulate drone production according to food availability and season. Optimal drone brood production is dependent on many factors, including climate, colony status, and forage availability conditions. The amount of drone brood stock in the colony also affects drone production, so regularly harvesting them is one way for farmers to ensure that drone production continues [190].

Currently, the production of drone broods for feed and food is not approved in the EU. However, given that there are economic and biological control advantages to be obtained, it is likely that they will be approved in the future. The following welfare discussion focuses on their use in apiculture, as drone brood production is likely to be a by-product of existing apiculture production rather than a stand-alone industry.

9.3. Western honeybee diets in commercial systems

Adult honeybees consume nectar and pollen from flowers. Therefore, access to abundant and varied flowers is necessary for honeybee brood production, immune function, and the general survival of the colony [193]. However, climate change and highly cultivated landscapes are negatively affecting floral availability and diversity and, in turn, bee nutrition [193]. Today, commercial beekeepers often provide their colonies with an artificial “pollen substitute” to compensate for insufficient nutritional forage in the environment [193]. While the composition of artificial bee diet formulations varies, they often incorporate protein-rich ingredients such as soy, pea, yeast, casein, egg, and microalgae [193,194].

9.4. The welfare implications of the diets used for the Western honeybee

Artificial diets do not provide the bees with important micronutrients, and as a result, the colonies suffer negative consequences. For example, studies have shown that access to natural forage results in improved health, reduced pathogen loads, and higher overwintering survival rates compared with artificial diets [193,195].

A study by Kumar and Agrawal [196] compared the performance of honeybee colonies fed with various artificial diets in India and found that a diet comprised of equal parts of soy flour, brewer's yeast, soy protein hydrolysate, sugar, and glucose was consumed in the largest quantities, and had the best outcomes in terms of colony parameters, such as egg laying, honey stores and bee coverage area [196]. Similarly, Kumar and Kumar [197] also found that an artificial bee diet comprised of equal parts defatted soy flour, parched gram, brewer's yeast, soy protein hydrolysate, and two parts sugar showed favourable outcomes for honeybees in terms of strength, health and the build-up of colonies during dearth periods, compared to Haydak's 1967 formulae [197].

9.5. Recommendations for the Western honeybee

- Honeybees should be reared in outdoor systems that allow for adults to collect nectar and pollen from flowers.
- Where necessary to account for poor forage in the natural environment, supplementation with artificial diets should be allowed.
- Artificial pollen substitutes must provide adequate protein.

- More research into the welfare implications of certain diets in honeybee farming is required.

10. Silkworms (*Bombyx mori*)

The silkworm (*Bombyx mori*) was added to the list of authorised insect species for use as protein in aquaculture feed in 2021.[4]

10.1. Natural biology of the silkworm (Bombyx mori)

The total lifecycle of the silkworm is completed in 6-8 weeks and consists of four stages of growth: egg (10-25 days), larva (20-33 days), pupa (10-14 days), and the adult moth (5-10 days). The moth lays the eggs on the leaves of a mulberry tree, and once the eggs hatch, the larvae feed on the tree's leaves continuously for about 20-33 days before spinning cocoons to transform into pupae. The adult moths emerge from these cocoons to mate, and the females

[4] https://eur-lex.europa.eu/legal-content/EN/TXT/?uri=CELEX%3A32021R1925&qid=1696410564947

can lay up to 500 eggs. Adult moths cannot fly and do not eat due to their reduced or absent mouthparts [198–200].

10.2. Overview of the farming systems for silkworms

Silkworms have so far been farmed for their silk (sericulture) at large scales in automatic rearing systems that allow for year-round production on trays that can be arranged in tiers [198,201]. The silkworm pupae, a by-product once the silk thread has been wound off the cocoon [202], are also edible and are frequently used as food in Asian countries [203].

10.3. Silkworm diets in commercial systems

Silkworms are monophagous insects that only feed on fresh mulberry leaves (preferably white mulberry species) or sometimes Osage orange leaves [204]. Consumption rates and the type of leaf vary as the larvae grow, as the first few instar phases are associated with low consumption (around 2% of that consumed throughout the entire larval period), focusing more on young, tender leaves [205]. The fourth and fifth stages are when the most mulberry leaves are ingested, around 10% in the fourth stage and around 88% in the fifth stage.

In the 1960s, an artificial diet was developed and is commonly referred to as 'silkworm chow' [204–206]. According to Taylor Battistella from Everything Silkworms, the development of silkworm chow is very difficult and something they failed to achieve even in a laboratory setting (Battistella, personal communications, 9 October 2023). As a result, the composition in viable mixes is not shared by those who have succeeded. Battistella commented that the only way to develop a successful chow diet would be through thorough scientific testing and analysis in a research-approved setting, given the number of vitamins, antibiotics and supplements required in the formulation. However, silkworm chow can be purchased online, and some websites provide a step-by-step guide to making silkworm chow using dried mulberry leaves, soy flour and maise/corn meal mixed with water.[5]

In general, the artificial diet used for rearing silkworms contains mulberry flour as its main ingredient, along with other ingredients such as soybean meal, corn starch, citric acid, ascorbic acid, agar-agar, trace elements and vitamins, sitosterol, antibiotics and preservatives; however, the composition generally varies [207].

A study by Qin et al. [208] utilised the following artificial diet composition: 30% defatted soybean powder, 25% mulberry leaf powder, 19.6% starch, 10% cellulose powder, 8% agar, 2% vitamin C, 2% citric acid; 1.5% vitamin B complex; 1% mineral salts complex; 0.5% Potassium Sorbate; 0.2% choline chloride; 0.2% Calcium propionate. It was mixed with three times the amount of water (ratio by weight) to the unprocessed artificial diet and heated at 100 °C for 35 minutes [208].

[5] https://www.wikihow.com/

10.4. The welfare implications of the artificial diet used for silkworm larvae

10.4.1. Poor performance

The welfare risks of rearing silkworms on artificial diets are not yet fully understood [207]. Despite efforts to continually improve artificial diet formulae, silkworms reared on artificial diets have higher mortality rates, poorer development and physical weakness, poorer metabolism, reduced disease resistance and low silk yield development compared with those on a natural diet, which is thought to be due to the metabolic utilisation of artificial diets in silkworms being less than that of mulberry leaves [21,22,208,209]. Taylor Battistella from Everything Silkworms sells silkworm chow on his website and explains that silkworm chow is not as nutritious as feeding silkworms mulberry leaves (Battistella, personal communications, 9 October 2023). However, it can be a practical option, especially for smaller-scale farms in urban settings.

One study compared the growth and development of the domesticated silkworm at the protein level when fed either fresh mulberry leaves or an artificial diet [210]. They found that different diets could alter the expression of proteins, which can affect immunity, digestion, and the absorption of nutrients in silkworms [210]. This results in a lower cocoon yield, low survival rate of young larvae, and poor resistance to specific pathogens seen in silkworms reared on artificial diets [210].

In another study, Qin and colleagues [208] compared the faecal metabolome of silkworms fed on artificial diets compared to silkworms reared with mulberry leaves to investigate the stunted growth and the low efficiency of silk protein synthesis of silkworms fed with artificial diet. They found 57 differentially expressed metabolites, 39 of which were upregulated and 18 were downregulated in the silkworms fed mulberry leaves, compared with the artificial diets [208]. In particular, most of the amino acids, carbohydrates and lipids associated with physical development and silk protein biosynthesis were enriched in silkworms reared on mulberry leaves [208].

10.4.2. Impact on gut microbiota

Silkworms reared on artificial diets have been found to show compromised disease resistance and immunity and require greater care during preparation and disinfection [21,22,208,209]. Today, antibiotics may be readily added to the diets of silkworms to address disease risks in production [211]. However, antibiotics and increasingly sterile rearing conditions are also negatively impacting the intestinal microbial diversity in the silkworms, which is suggested to be associated with various negative outcomes for the silkworms [21].

A study comparing the gut microbiota between silkworms reared on fresh mulberry leaves and an artificial diet found that the abundance of dominant bacteria in the gut microbiota differed due to diet and silkworm strains [21]. The gut microbiota diversity was lower in the silkworm strains reared on the artificial diet, compared with fresh mulberry leaves, which is thought to contribute to the poor development, metabolism, and disease resistance seen with the artificial diet [21]. Intestinal microbial diversity is also adversely affected by the sterile feeding environments associated with the rearing of silkworms on artificial diets and the addition of antibiotics to the diets [21].

10.4.3. Silkworm preferences for substrates

According to a study by Zhang and colleagues [204], the species-specific feeding preference is attributed to the gene GR66, which encodes a putative bitter gustatory receptor (GR) responsible for the mulberry-specific feeding preference. The study found that a gene mutation in silkworms can change their eating habits, with GR66 mutant larvae found to eat other plant species and fresh fruits and grain seeds. Such findings are thought to be promising in the search for alternative food sources for the mass rearing of silkworms [204].

10.5. Recommendations for the Silkworm

- Silkworms are monophagous insects and should ideally only feed on fresh mulberry leaves.
- If artificial diets are required, supplementation with fresh mulberry leaves should be encouraged, and care should be taken to monitor the welfare of the silkworms on the artificial diet.
- More research should be conducted to investigate the welfare implications of rearing silkworms on artificial diets.

11. Conclusion

One overwhelming factor coming from the scientific literature is that there is a considerable amount of trial and error still happening in insect farms with regard to the diets used, especially as one diet may work on one farm and not on another. The consequence of this is that the welfare of the insects suffers, as mortality rates increase when non-optimal diets are used, and this may incur a degree of suffering or ill health. As considerable progress is being made in insect breeding, this also plays a large role as different strains will adapt differently [47]. The EU appears to be more focused on breeding BSF and mealworms, and consequently, there is more known about the dietary needs of these species. However, even for them, many unanswered questions remain regarding what is best for their welfare. Further research is desperately needed for all farmed and pending species to establish the optimum diets for welfare.

References

1. European Commission Approval Insect Novel Food Available online: https://food.ec.europa.eu/safety/novel-food/authorisations/approval-insect-novel-food_en (accessed on 15 October 2023).
2. IPIFF Insects As Feed EU Legislation – Aquaculture, Poultry & Pig Species Available online: https://ipiff.org/insects-eu-legislation/ (accessed on 15 October 2023).
3. IPIFF *IPIFF Guide-on-Good-Hygiene-Practices for European Union (EU) Producers of Insects as Food and Feed*; International Platform of Insects for Food and Feed, 2022; p. 121.
4. Van Zanten, H.H.E.; Mollenhorst, H.; Oonincx, D.G.A.B.; Bikker, P.; Meerburg, B.G.; De Boer, I.J.M. From Environmental Nuisance to Environmental Opportunity: Housefly Larvae Convert Waste to Livestock Feed. *Journal of Cleaner Production* **2015**, *102*, 362–369, doi:10.1016/j.jclepro.2015.04.106.
5. IPIFF *The Insect Sector Milestones towards Sustainable Food Supply Chains*; 2020.
6. Malematja, E.; Manyelo, T.G.; Sebola, N.A.; Kolobe, S.D.; Mabelebele, M. The Accumulation of Heavy Metals in Feeder Insects and Their Impact on Animal Production. *Science of The Total Environment* **2023**, *885*, 163716, doi:10.1016/j.scitotenv.2023.163716.
7. Merskin, D. She, He, Not It: Language, Personal Pronouns, and Animal Advocacy. *Journal of World Languages* **2022**, *8*, 391–408, doi:10.1515/jwl-2022-0018.
8. Lambert, H.; Elwin, A.; D'Cruze, N. Wouldn't Hurt a Fly? A Review of Insect Cognition and Sentience in Relation to Their Use as Food and Feed. *Applied Animal Behaviour Science* **2021**, *243*, 105432, doi:10.1016/j.applanim.2021.105432.

9. Klein, C.; Barron, A.B. Insects Have the Capacity for Subjective Experience. *Animal Sentience* **2016**, *1*, doi:10.51291/2377-7478.1113.
10. Bear, C. Approaching Insect Death: Understandings and Practices of the UK's Edible Insect Farmers. *Society and Animals* **2019**, *27*, 751–768, doi:10.1163/15685306-00001871.
11. Bear, C. Making Insects Tick: Responsibility, Attentiveness and Care in Edible Insect Farming. *Environment and Planning E: Nature and Space* **2020**, *0*, 251484862094532, doi:10.1177/2514848620945321.
12. Boppré, M.; Vane-Wright, R.I. Welfare Dilemmas Created by Keeping Insects in Captivity. In *The welfare of invertebrate animals*; Springer: Cham, 2019; pp. 23–67.
13. van Huis, A. Welfare of Farmed Insects. *Journal of Insects as Food and Feed* **2019**, *5*, 1–12, doi:10.3920/jiff2020.0061.
14. Barrett, M.; Chia, S.Y.; Fischer, B.; Tomberlin, J.K. Welfare Considerations for Farming Black Soldier Flies, Hermetia Illucens (Diptera: Stratiomyidae): A Model for the Insects as Food and Feed Industry. *JIFF* **2023**, *9*, 119–148, doi:10.3920/JIFF2022.0041.
15. Addeo, N.F.; Vozzo, S.; Secci, G.; Mastellone, V.; Piccolo, G.; Lombardi, P.; Parisi, G.; Asiry, K.A.; Attia, Y.A.; Bovera, F. Different Combinations of Butchery and Vegetable Wastes on Growth Performance, Chemical-Nutritional Characteristics and Oxidative Status of Black Soldier Fly Growing Larvae. *Animals* **2021**, *11*, 3515, doi:10.3390/ani11123515.
16. Bjørge, J.D.; Overgaard, J.; Malte, H.; Gianotten, N.; Heckmann, L.H. Role of Temperature on Growth and Metabolic Rate in the Tenebrionid Beetles Alphitobius Diaperinus and Tenebrio Molitor. *Journal of Insect Physiology* **2018**, *107*, 89–96, doi:10.1016/j.jinsphys.2018.02.010.
17. Julita, U.; Suryani, Y.; Kinasih, I.; Yuliawati, A.; Cahyanto, T.; Maryeti, Y.; Permana, A.D.; Fitri, L.L. Growth Performance and Nutritional Composition of Black Soldier Fly, Hermetia Illucens (L), (Diptera :

Stratiomyidae) Reared on Horse and Sheep Manure. *IOP Conf. Ser.: Earth Environ. Sci.* **2018**, *187*, 012071, doi:10.1088/1755-1315/187/1/012071.

18. Barrett, M.; Godfrey, R.K.; Schnell, A.; Fischer, B. Farmed Yellow Mealworm (Tenebrio Molitor; Coleoptera: Tenebrionidae) Welfare: Species-Specific Recommendations for a Global Industry 2023.
19. Adámková, A.; Adámek, M.; Mlček, J.; Borkovcová, M.; Bednářová, M.; Kouřimská, L.; Skácel, J.; Vítová, E. Welfare of the Mealworm (Tenebrio Molitor) Breeding with Regard to Nutrition Value and Food Safety. *Potravinarstvo Slovak Journal of Food Sciences* **2017**, *11*, 460–465.
20. Gobbi, P.; Martínez-Sánchez, A.; Rojo, S. The Effects of Larval Diet on Adult Life-History Traits of the Black Soldier Fly, Hermetia Illucens (Diptera: Stratiomyidae). *Eur. J. Entomol.* **2013**, *110*, 461–468, doi:10.14411/eje.2013.061.
21. Dong, H.-L.; Zhang, S.-X.; Chen, Z.-H.; Tao, H.; Li, X.; Qiu, J.-F.; Cui, W.-Z.; Sima, Y.-H.; Cui, W.-Z.; Xu, S.-Q. Differences in Gut Microbiota between Silkworms (Bombyx Mori) Reared on Fresh Mulberry (Morus Alba Var. Multicaulis) Leaves or an Artificial Diet. *RSC Adv.* **2018**, *8*, 26188–26200, doi:10.1039/C8RA04627A.
22. Dong, H.-L.; Zhang, S.-X.; Tao, H.; Chen, Z.-H.; Li, X.; Qiu, J.-F.; Cui, W.-Z.; Sima, Y.-H.; Cui, W.-Z.; Xu, S.-Q. Metabolomics Differences between Silkworms (Bombyx Mori) Reared on Fresh Mulberry (Morus) Leaves or Artificial Diets. *Sci Rep* **2017**, *7*, 10972, doi:10.1038/s41598-017-11592-4.
23. Harjoko, D.N.; Hua, Q.Q.H.; Toh, E.M.C.; Goh, C.Y.J.; Puniamoorthy, N. A Window into Fly Sex: Mating Increases Female but Reduces Male Longevity in Black Soldier Flies. *Animal Behaviour* **2023**, *200*, 25–36, doi:10.1016/j.anbehav.2023.03.007.
24. Miranda, C.D.; Cammack, J.A.; Tomberlin, J.K. Large-Scale Production of House Fly, *Musca Domestica* (Diptera: Muscidae), Larvae Fed 3 Manure

Types. *Journal of Economic Entomology* **2023**, *116*, 1102–1109, doi:10.1093/jee/toad099.

25. Hopkins, I.; Newman, L.P.; Gill, H.; Danaher, J. The Influence of Food Waste Rearing Substrates on Black Soldier Fly Larvae Protein Composition: A Systematic Review. *Insects* **2021**, *12*, 608, doi:10.3390/insects12070608.
26. Van Huis, A.; Rumpold, B.A.; Van Der Fels-Klerx, H.J.; Tomberlin, J.K. Advancing Edible Insects as Food and Feed in a Circular Economy. *JIFF* **2021**, *7*, 935–948, doi:10.3920/JIFF2021.x005.
27. Varelas, V. Food Wastes as a Potential New Source for Edible Insect Mass Production for Food and Feed: A Review. *Fermentation* **2019**, *5*, 81, doi:10.3390/fermentation5030081.
28. Diener, S.; Zurbrügg, C.; Tockner, K. Bioaccumulation of Heavy Metals in the Black Soldier Fly, Hermetia Illucens and Effects on Its Life Cycle. *JIFF* **2015**, *1*, 261–270, doi:10.3920/JIFF2015.0030.
29. Rowe, A. *Insects Raised for Food and Feed — Global Scale, Practices, and Policy — Rethink Priorities*; 2020.
30. Kolakowski, B.M.; Johaniuk, K.; Zhang, H.; Yamamoto, E. Analysis of Microbiological and Chemical Hazards in Edible Insects Available to Canadian Consumers. *Journal of Food Protection* **2021**, *84*, 1575–1581.
31. Sorjonen, J.M.; Karhapaa, M.; Holm, S.; Valtonen, A.; Roininen, H. Performance of the House Cricket (Acheta Domesticus) on by-Product Diets in Small-Scale Production. *Journal of Insects as Food and Feed* **2022**, *8*, 289–294.
32. Rowe, E.; López, K.Y.R.; Robinson, K.M.; Baudier, K.M.; Barrett, M. Farmed Cricket (Acheta Domesticus, Gryllus Assimilis, and Gryllodes Sigillatus; Orthoptera) Welfare Considerations: Recommendations for Improving Global Practice 2023.

33. Makkar, H.P.S.; Tran, G.; Heuzé, V.; Ankers, P. State-of-the-Art on Use of Insects as Animal Feed. *Animal Feed Science and Technology* **2014**, *197*, 1–33, doi:10.1016/J.ANIFEEDSCI.2014.07.008.
34. Yang, S. Case Presentation of Large-Scale Production of Black Soldier Fly Larvae. *The Black Soldier Fly Company* 2017.
35. Nguyen, T. Influence of Diet on Black Soldier Fly (Hermetia Illucens Linnaeus) (Diptera: Stratiomyidae) Life History Traits, UniversityofWindsor, 2010.
36. Tomberlin, J.K.; Sheppard, D.C.; Joyce, J.A. *Black Soldier Fly (Diptera: Stratiomyidae) Colonization of Pig Carrion in South Georgia*; Citeseer, 2005;
37. Dortsman, B.; Diener, S.; Verstappen, B.; Zurbrügg, C. *Black Soldier Fly Biowaste Processing - A Step by Step Guide*; Eawag: Swiss Federal Institute of Aquatic Science and Technology: Dübendorf, Switzerland, 2017;
38. Yang, S. Explanation of an Industrial Application of Black Soldier Fly Converting Food Waste into Valuable Products. *EVO Conversion Systems - The Black Soldier Fly Company* 2019.
39. Palma, L.; Ceballos, S.J.; Johnson, P.C.; Niemeier, D.; Pitesky, M.; VanderGheynst, J.S. Cultivation of Black Soldier Fly Larvae on Almond Byproducts: Impacts of Aeration and Moisture on Larvae Growth and Composition. *J Sci Food Agric* **2018**, *98*, 5893–5900, doi:10.1002/jsfa.9252.
40. Tomberlin, J. The Story of a Plant- How Kudzu Started a Black Soldier Fly Colony. *A Science Blog About Black Soldier Fly* 2017.
41. Rehman, K.; Rehman, R.U.; Somroo, A.A.; Cai, M.; Zheng, L.; Xiao, X.; Rehman, A.U.; Rehman, A.; Tomberlin, J.K.; Yu, Z. Enhanced Bioconversion of Dairy and Chicken Manure by the Interaction of Exogenous Bacteria and Black Soldier Fly Larvae. *Journal of environmental management* **2019**, *237*, 75–83.

42. Zheng, L.; Hou, Y.; Li, W.; Yang, S.; Li, Q.; Yu, Z. Biodiesel Production from Rice Straw and Restaurant Waste Employing Black Soldier Fly Assisted by Microbes. *Energy* **2012**, *47*, 225–229.
43. Isibika, A.; Vinnerås, B.; Kibazohi, O.; Zurbrügg, C.; Lalander, C. Pre-Treatment of Banana Peel to Improve Composting by Black Soldier Fly (Hermetia Illucens (L.), Diptera: Stratiomyidae) Larvae. *Waste Management* **2019**, *100*, 151–160.
44. Li, W.; Li, M.; Zheng, L.; Liu, Y.; Zhang, Y.; Yu, Z.; Ma, Z.; Li, Q. Simultaneous Utilization of Glucose and Xylose for Lipid Accumulation in Black Soldier Fly. *Biotechnol Biofuels* **2015**, *8*, 117, doi:10.1186/s13068-015-0306-z.
45. Rehman, K.; Rehman, A.; Cai, M.; Zheng, L.; Xiao, X.; Somroo, A.A.; Wang, H.; Li, W.; Yu, Z.; Zhang, J. Conversion of Mixtures of Dairy Manure and Soybean Curd Residue by Black Soldier Fly Larvae (Hermetia Illucens L.). *Journal of Cleaner Production* **2017**, *154*, 366–373.
46. Vogel, H.; Müller, A.; Heckel, D.G.; Gutzeit, H.; Vilcinskas, A. Nutritional Immunology: Diversification and Diet-Dependent Expression of Antimicrobial Peptides in the Black Soldier Fly Hermetia Illucens. *Developmental & Comparative Immunology* **2018**, *78*, 141–148.
47. Zhou, F.; Tomberlin, J.K.; Zheng, L.; Yu, Z.; Zhang, J. Developmental and Waste Reduction Plasticity of Three Black Soldier Fly Strains (Diptera: Stratiomyidae) Raised on Different Livestock Manures. *Journal of Medical Entomology* **2013**, *50*, 1224–1230, doi:10.1603/ME13021.
48. Nguyen, T.T.X.; Tomberlin, J.K.; Vanlaerhoven, S. Influence of Resources on Hermetia Illucens (Diptera: Stratiomyidae) Larval Development. *Journal of Medical Entomology* **2013**, *50*, 898–906, doi:10.1603/ME12260.
49. Banks, I.J.; Gibson, W.T.; Cameron, M.M. Growth Rates of Black Soldier Fly Larvae Fed on Fresh Human Faeces and Their Implication for Improving Sanitation. *Tropical Med Int Health* **2014**, *19*, 14–22, doi:10.1111/tmi.12228.

50. Lee, K.-S.; Yun, E.-Y.; Goo, T.-W. Optimization of Feed Components to Improve Hermetia Illucens Growth and Development of Oil Extractor to Produce Biodiesel. *Animals* **2021**, *11*, 2573, doi:10.3390/ani11092573.
51. Miranda, C., D.; Cammack, J., A.; Tomberlin, J., K. Mass Production of the Black Soldier Fly, Hermetia Illucens (L.), (Diptera: Stratiomyidae) Reared on Three Manure Types. *Animals* **2020**, *10*, 1–11.
52. Myers, H.M.; Tomberlin, J.K.; Lambert, B.D.; Kattes, D. Development of Black Soldier Fly (Diptera: Stratiomyidae) Larvae Fed Dairy Manure. *Environmental entomology* **2014**, *37*, 11–15.
53. Laganaro, M.; Bahrndorff, S.; Eriksen, N.T. Growth and Metabolic Performance of Black Soldier Fly Larvae Grown on Low and High-Quality Substrates. *Waste Management* **2021**, *121*, 198–205, doi:10.1016/j.wasman.2020.12.009.
54. Parodi, A.; Van Dijk, K.; Van Loon, J.J.A.; De Boer, I.J.M.; Van Schelt, J.; Van Zanten, H.H.E. Black Soldier Fly Larvae Show a Stronger Preference for Manure than for a Mass-Rearing Diet. *Journal of Applied Entomology* **2020**, *144*, 560–565, doi:10.1111/jen.12768.
55. Barragan-Fonseca, K.B.; Gort, G.; Dicke, M.; van Loon, J.J.A. Effects of Dietary Protein and Carbohydrate on Life-History Traits and Body Protein and Fat Contents of the Black Soldier Fly Hermetia Illucens. *Physiological Entomology* **2019**, *44*, 148–159, doi:10.1111/phen.12285.
56. Abd El-Hack, M.E.; Shafi, M.E.; Alghamdi, W.Y.; Abdelnour, S.A.; Shehata, A.M.; Noreldin, A.E.; Ashour, E.A.; Swelum, A.A.; Al-sagan, A.A.; Alkhateeb, M.; et al. Black Soldier Fly (Hermetia Illucens) Meal as a Promising Feed Ingredient for Poultry: A Comprehensive Review. *Agriculture (Switzerland)* **2020**, *10*, 1–31, doi:10.3390/agriculture10080339.
57. Bertinetti, C.; Samayoa, A.C.; Hwang, S.Y. Effects of Feeding Adults of Hermetia Illucens (Diptera: Stratiomyidae) on Longevity, Oviposition, and Egg Hatchability: Insights into Optimizing Egg Production. *Journal of Insect Science* **2019**, *19*, 1–7, doi:10.1093/jisesa/iez001.

58. Karlsson, B. Nuptial Gifts, Resource Budgets, and Reproductive Output in a Polyandrous Butterfly. *Ecology* **1998**, *79*, 2931–2940, doi:10.1890/0012-9658(1998)079[2931:NGRBAR]2.0.CO;2.
59. Macavei, L.I.; Benassi, G.; Stoian, V.; Maistrello, L. Optimization of Hermetia Illucens (L.) Egg Laying under Different Nutrition and Light Conditions. *PLoS ONE* **2020**, *15*, 1–18, doi:10.1371/journal.pone.0232144.
60. Nakamura, S.; Ichiki, R.T.; Shimoda, M.; Morioka, S. Small-Scale Rearing of the Black Soldier Fly, Hermetia Illucens (Diptera: Stratiomyidae), in the Laboratory: Low-Cost and Year-Round Rearing. *Appl Entomol Zool* **2016**, *51*, 161–166, doi:10.1007/s13355-015-0376-1.
61. van Huis, A.; Oonincx, D.G.A.B.; Rojo, S.; Tomberlin, J.K. Insects as Feed: House Fly or Black Soldier Fly? *Journal of Insects as Food and Feed* **2020**, *6*, 221–229, doi:10.3920/JIFF2020.X003.
62. Francuski, L.; Jansen, W.; Beukeboom, L.W. Effect of Temperature on Egg Production in the Common Housefly. *Entomologia Experimentalis et Applicata* **2020**, *168*, 513–522, doi:10.1111/eea.12912.
63. van Huis, A.; Van Itterbeeck, J.; H, K.; Mertens, E.; Halloran, A.; Muir, G.; Vantomme, P. *Edible Insects: Future Prospects for Food and Feed Security*; Food and Agriculture Organization of the United Nations: Rome, 2013.
64. Koné, N.; Sylla, M.; Nacambo, S.; Kenis, M. Production of House Fly Larvae for Animal Feed through Natural Oviposition. *Journal of Insects as Food and Feed* **2017**, *3*, 177–186, doi:10.3920/JIFF2016.0044.
65. Hussein, M.; Pillai, V.V.; Goddard, J.M.; Park, H.G.; Kothapalli, K.S.; Ross, D.A.; Ketterings, Q.M.; Brenna, J.T.; Milstein, M.B.; Marquis, H.; et al. Sustainable Production of Housefly (Musca Domestica) Larvae as a Protein-Rich Feed Ingredient by Utilizing Cattle Manure. *PLoS ONE* **2017**, *12*, 1–19, doi:10.1371/journal.pone.0171708.

66. Larraín, P.; Salas, C. House Fly (Musca Domestica L.) (Diptera Muscidae) Development in Different Types of Manure. *Chilean J. Agric. Res.* **2008**, *68*, doi:10.4067/S0718-58392008000200009.
67. Khan, H.A.A.; Shad, S.A.; Akram, W. Effect of Livestock Manures on the Fitness of House Fly, Musca Domestica L. (Diptera: Muscidae). *Parasitol Res* **2012**, *111*, 1165–1171, doi:10.1007/s00436-012-2947-1.
68. Čičková, H.; Pastor, B.; Kozánek, M.; Martínez-Sánchez, A.; Rojo, S.; Takáč, P. Biodegradation of Pig Manure by the Housefly, Musca Domestica: A Viable Ecological Strategy for Pig Manure Management. *PLOS ONE* **2012**, *7*, e32798, doi:10.1371/journal.pone.0032798.
69. Zi-zhe, C.; De-po, Y.; Sheng-qing, W.; Yong, W.; Reaney, M.J.T.; Zhi-min, Z.; Long-ping, Z.; Guo, S.; Yi, N.; Dong, Z.; et al. Conversion of Poultry Manure to Biodiesel, a Practical Method of Producing Fatty Acid Methyl Esters via Housefly (Musca Domestica L.) Larval Lipid. *Fuel* **2017**, *210*, 463–471, doi:10.1016/j.fuel.2017.08.109.
70. Fatchurochim, S.; Geden, C.J.; Axtell, R.C. Filth Fly (Diptera) Oviposition and Larval Development in Poultry Manure of Various Moisture Levels. *Journal of Entomological Science* **1989**, *24*, 224–231.
71. Nayduch, D.; Burrus, R.G. Flourishing in Filth: House Fly–Microbe Interactions Across Life History. *Annals of the Entomological Society of America* **2017**, *110*, 6–18, doi:10.1093/aesa/saw083.
72. Kortsmit, Y.; Van Loon, J.J.A.; Dicke, M. Preference of Black Soldier Fly Larvae for Feed Substrate Previously Colonised by Conspecific Larvae. *J Applied Entomology* **2023**, *147*, 336–345, doi:10.1111/jen.13112.
73. Bahrndorff, S.; Kjærsgaard, A.; Pertoldi, C.; Loeschcke, V.; Schou, T.M.; Skovgrd, H.; Hald, B. The Effects of Sex-Ratio and Density on Locomotor Activity in the House Fly, Musca Domestica. *Journal of Insect Science* **2012**, *12*, 1–12, doi:10.1673/031.012.7101.
74. Miranda, C.D.; Cammack, J.A.; Tomberlin, J.K. Interspecific Competition between the House Fly, Musca Domestica L. (Diptera: Muscidae) and

Black Soldier Fly, Hermetia Illucens (L.) (Diptera: Stratiomyidae) When Reared on Poultry Manure. *Insects* **2019**, *10*, 440, doi:10.3390/insects10120440.

75. Ganda, H.; Abihona, H.A.; Zannou-Boukari, E.T.; Kenis, M.; Chrysostome, C.A.A.M.; Mensah, G.A. Influence of Adult Diet on Biological Parameters of the Housefly, Musca Domestica L. (Diptera: Muscidae). *JoBAZ* **2020**, *81*, 46, doi:10.1186/s41936-020-00181-z.
76. Andersen, J.; Berggreen, I.E.; Heckmann, L.L. *Recommendations for Breeding and Holding of Regular Mealworm Recommendations for Breeding and Holding of Regular Mealworm, Tenebrio Molitor*; INBIOM, 2017; pp. 1–7.
77. Sammarco, B.C.; Hinkle, N.C.; Crossley, M.S. Biology and Management of Lesser Mealworm Alphitobius Diaperinus (Coleoptera: Tenebrionidae) in Broiler Houses. *Journal of Integrated Pest Management* **2023**, *14*, 2, doi:10.1093/jipm/pmad003.
78. Balfour, C.E.; Carmichael, L. The Light Reactions of the Meal Worm (Tenebrio Molitor Linn). *The American Journal of Psychology* **1928**, *40*, 576–584.
79. Ribeiro, N.; Abelho, M.; Costa, R. A Review of the Scientific Literature for Optimal Conditions for Mass Rearing Tenebrio Molitor (Coleoptera: Tenebrionidae). *Journal of Entomological Science* **2018**, *53*, 434–454, doi:10.18474/JES17-67.1.
80. Morales-Ramos, J.A.; Rojas, M.G. Effect of Larval Density on Food Utilization Efficiency of Tenebrio Molitor (Coleoptera: Tenebrionidae). *Journal of economic entomology* **2015**, *108*, 2259–2267.
81. Ortiz, J.C.; Ruiz, A.T.; Morales-Ramos, J.A.; Thomas, M.; Rojas, M.G.; Tomberlin, J.K.; Yi, L.; Han, R.; Giroud, L.; Jullien, R.L. Insect Mass Production Technologies. In *Insects as sustainable food ingredients*; Elsevier, 2016; pp. 153–201.

82. Langston, K.; Selaledi, L.; Yusuf, A. Evaluation of Alternative Substrates for Rearing the Yellow Mealworm Tenebrio Molitor (L). *Int J Trop Insect Sci* **2023**, doi:10.1007/s42690-023-01061-z.
83. Gianotten, N.; Soetemans, L.; Bastiaens, L. Agri-Food Side-Stream Inclusions in the Diet of Alphitobius Diaperinus Part 1: Impact on Larvae Growth Performance Parameters. *Insects* **2020**, *11*, 79, doi:10.3390/insects11020079.
84. Crook, P.G.; Novak, J.A.; Spilman, T.J. The Lesser Mealworm, Alphitobius Diaperinus, in the Scrotum of Rattus Norvegicus, with Notes on Other Vertebrate Associations (Coleoptera, Tenebrionidae; Rodentia, Muridae). *The Coleopterists Bulletin* **1980**, *34*, 393–396.
85. Deruytter, D.; Coudron, C.L.; Teerlinck, S. Influence of Crate Size, Oviposition Time, Number of Adults and Cannibalism on the Reproduction of Tenebrio Molitor. *Journal of Insects as Food and Feed* **2019**, *5*, 247–255, doi:10.3920/JIFF2019.0018.
86. Fasce, B.; Ródenas, L.; López, M.C.; Moya, V.J.; Pascual, J.J.; Cambra-López, M. Nutritive Value of Wheat Bran Diets Supplemented With Fresh Carrots and Wet Brewers' Grains in Yellow Mealworm. *J Insect Sci* **2022**, *22*, 7, doi:10.1093/jisesa/ieac022.
87. Gourgouta, M.; Rumbos, C.I.; Michail, V.; Athanassiou, C.G. Valorization of Agricultural Side-Streams for the Rearing of Larvae of the Lesser Mealworm, Alphitobius Diaperinus (Panzer). *Sustainability* **2022**, *14*, 7680, doi:10.3390/su14137680.
88. Kotsou, K.; Rumbos, C.I.; Baliota, G.V.; Gourgouta, M.; Athanassiou, C.G. Influence of Temperature, Relative Humidity and Protein Content on the Growth and Development of Larvae of the Lesser Mealworm, Alphitobius Diaperinus (Panzer). *Sustainability* **2021**, *13*, 11087, doi:10.3390/su131911087.
89. Morales-Ramos, J.A.; Rojas, M.G.; Shapiro-Nan, D.I.; Tedders, W.L. Developmental Plasticity in Tenebrio Molitor (Coleoptera:

Tenebrionidae): Analysis of Instar Variation in Number and Development Time under Different Diets. *Journal of Entomological Science* **2010**, *45*, 75–90, doi:10.18474/0749-8004-45.2.75.

90. van Broekhoven, S.; Oonincx, D.G.A.B.; van Huis, A.; van Loon, J.J.A. Growth Performance and Feed Conversion Efficiency of Three Edible Mealworm Species (Coleoptera: Tenebrionidae) on Diets Composed of Organic by-Products. *Journal of Insect Physiology* **2015**, *73*, 1–10, doi:10.1016/j.jinsphys.2014.12.005.
91. Baek, S.; Perez, A.E.; Turcotte, R.M.; White, J.B.; Adedipe, F.; Park, Y.-L. Response of Tenebrio Molitor (Coleoptera: Tenebrionidae) Adults to Potato: Implications for Monitoring and Sampling. *Journal of Stored Products Research* **2015**, *60*, 5–10, doi:10.1016/j.jspr.2014.11.002.
92. Deruytter, D.; Coudron, C. l.; Claeys, J. The Influence of Wet Feed Distribution on the Density, Growth Rate and Growth Variability of Tenebrio Molitor. *Journal of Insects as Food and Feed* **2021**, *7*, 141–149, doi:10.3920/JIFF2020.0049.
93. Liu, C.; Masri, J.; Perez, V.; Maya, C.; Zhao, J. Growth Performance and Nutrient Composition of Mealworms (Tenebrio Molitor) Fed on Fresh Plant Materials-Supplemented Diets. *Foods* **2020**, *9*, 151, doi:10.3390/foods9020151.
94. Morales-Ramos, J.A.; Rojas, M.G.; Shapiro-Llan, D.I.; Tedders, W.L. Use of Nutrient Self-Selection as a Diet Refining Tool in Tenebrio Molitor (Coleoptera: Tenebrionidae). *Journal of Entomological Science* **2013**, *48*, 206–221.
95. Oonincx, D.G.; van Broekhoven, S.; van Huis, A.; van Loon, J.J. Correction: Feed Conversion, Survival and Development, and Composition of Four Insect Species on Diets Composed of Food by-Products. *PLoS One* **2019**, *14*, e0222043.
96. Oonincx, D.G.; Van Broekhoven, S.; Van Huis, A.; van Loon, J.J. Feed Conversion, Survival and Development, and Composition of Four Insect

Species on Diets Composed of Food by-Products. *PloS one* **2015**, *10*, e0144601.

97. Morales-Ramos, J.A.; Rojas, M.G.; Shapiro-Ilan, D.I.; Tedders, W.L. Self-Selection of Two Diet Components by Tenebrio Molitor (Coleoptera: Tenebrionidae) Larvae and Its Impact on Fitness. *Environmental Entomology* **2011**, *40*, 1285–1294.
98. Cucini, C.; Funari, R.; Mercati, D.; Nardi, F.; Carapelli, A.; Marri, L. Polystyrene Shaping Effect on the Enriched Bacterial Community from the Plastic-Eating Alphitobius Diaperinus (Insecta: Coleoptera). *Symbiosis* **2022**, *86*, 305–313, doi:10.1007/s13199-022-00847-y.
99. Mitra, B.; Das, A.; Mitra, B.; Das, A. *The Ability of Insects to Degrade Complex Synthetic Polymers*; IntechOpen, 2023; ISBN 978-1-80355-613-0.
100. Nukmal, N. Effect of Styrofoam Waste Feeds on the Growth, Development and Fecundity of Mealworms (Tenebrio Molitor). *OnLine Journal of Biological Sciences* **2018**, *18*, 24–28.
101. Billen, P.; Khalifa, L.; Van Gerven, F.; Tavernier, S.; Spatari, S. Technological Application Potential of Polyethylene and Polystyrene Biodegradation by Macro-Organisms Such as Mealworms and Wax Moth Larvae. *Science of the Total Environment* **2020**, *735*, 139521.
102. Jin, L.; Feng, P.; Cheng, Z.; Wang, D. Effect of Biodegrading Polyethylene, Polystyrene, and Polyvinyl Chloride on the Growth and Development of Yellow Mealworm (Tenebrio Molitor) Larvae. *Environ Sci Pollut Res* **2022**, *30*, 37118–37126, doi:10.1007/s11356-022-24957-8.
103. Lou, Y.; Li, Y.; Lu, B.; Liu, Q.; Yang, S.-S.; Liu, B.; Ren, N.; Wu, W.-M.; Xing, D. Response of the Yellow Mealworm (Tenebrio Molitor) Gut Microbiome to Diet Shifts during Polystyrene and Polyethylene Biodegradation. *Journal of Hazardous Materials* **2021**, *416*, 126222.
104. Wang, Y.; Luo, L.; Li, X.; Wang, J.; Wang, H.; Chen, C.; Guo, H.; Han, T.; Zhou, A.; Zhao, X. Different Plastics Ingestion Preferences and Efficiencies of Superworm (Zophobas Atratus Fab.) and Yellow

Mealworm (Tenebrio Molitor Linn.) Associated with Distinct Gut Microbiome Changes. *Science of the Total Environment* **2022**, *837*, 155719.

105. Yang, L.; Gao, J.; Liu, Y.; Zhuang, G.; Peng, X.; Wu, W.-M.; Zhuang, X. Biodegradation of Expanded Polystyrene and Low-Density Polyethylene Foams in Larvae of Tenebrio Molitor Linnaeus (Coleoptera: Tenebrionidae): Broad versus Limited Extent Depolymerization and Microbe-Dependence versus Independence. *Chemosphere* **2021**, *262*, 127818.
106. Tsochatzis, E.D.; Berggreen, I.E.; Nørgaard, J.V.; Theodoridis, G.; Dalsgaard, T.K. Biodegradation of Expanded Polystyrene by Mealworm Larvae under Different Feeding Strategies Evaluated by Metabolic Profiling Using GC-TOF-MS. *Chemosphere* **2021**, *281*, 130840.
107. Yang, S.-S.; Wu, W.-M.; Brandon, A.M.; Fan, H.-Q.; Receveur, J.P.; Li, Y.; Wang, Z.-Y.; Fan, R.; McClellan, R.L.; Gao, S.-H. Ubiquity of Polystyrene Digestion and Biodegradation within Yellow Mealworms, Larvae of Tenebrio Molitor Linnaeus (Coleoptera: Tenebrionidae). *Chemosphere* **2018**, *212*, 262–271.
108. Galecki, R.; Sokol, R. A Parasitological Evaluation of Edible Insects and Their Role in the Transmission of Parasitic Diseases to Humans and Animals. *PLoS ONE* **2019**, *14*, 1–19, doi: https://doi.org/10.1371%2Fjournal.pone.0219303.
109. MacDougall, M.M. A Study of Temperature Effects on Gregarines of Tenebrio Molitor Larvae. *The Journal of Parasitology* **1942**, *28*, 233–240.
110. Ujvary, I. Pest Control Agents from Natural Products. In *Handbook of pesticide toxicology*; Academic Press: London, 2010; pp. 119–229.
111. Lecocq, A.; Natsopoulou, M.E.; Berggreen, I.E.; Eilenberg, J.; Heckmann, L.-H.L.; Nielsen, H.V.; Stensvold, C.R.; Jensen, A.B. Probiotic Properties of an Indigenous Pediococcus Pentosaceus Strain on Tenebrio Molitor

Larval Growth and Survival. *JIFF* **2021**, *7*, 975–986, doi:10.3920/JIFF2020.0156.

112. Rizou, E.; Kalogiouri, N.; Bisba, M.; Papadimitriou, A.; Kyrila, G.; Lazou, A.; Andreadis, S.; Hatzikamari, M.; Mourtzinos, I.; Touraki, M. Amelioration of Growth, Nutritional Value, and Microbial Load of Tenebrio Molitor (Coleoptera: Tenebrionidae) through Probiotic Supplemented Feed. *Eur Food Res Technol* **2022**, *248*, 727–739, doi:10.1007/s00217-021-03925-5.
113. Dahal, S.; Jensen, A.B.; Lecocq, A. Effect of Probiotics on Tenebrio Molitor Larval Development and Resistance against the Fungal Pathogen Metarhizium Brunneum. *Insects* **2022**, *13*, 1114.
114. WHO Mycotoxins Available online: https://www.who.int/news-room/fact-sheets/detail/mycotoxins (accessed on 9 October 2023).
115. Guo, Z.; Pfohl, K.; Karlovsky, P.; Dehne, H.-W.; Altincicek, B. Dissemination of Fusarium Proliferatum by Mealworm Beetle Tenebrio Molitor. *PLos ONE* **2018**, *13*, doi:10.1371/journal.pone.0204602.
116. Niermans, K.; Meyer, A.M.; Hoek-van den Hil, E.F.; van Loon, J.J.A.; van der Fels-Klerx, H.J. A Systwmatic Literature Review on the Effects of Mycotoxin Exposure on Insects and on Mycotoxin Accumulation and Biotransformation. *Mycotoxin Research* **2021**, *37*, 279–295.
117. Canteri de Souza, P.; Caloni, C.C.; Wilson, D.; Almeida, R.S. An Invertebrate Host to Study Fungal Infections, Mycotoxins and Antifungal Drugs: Tenebrio Molitor. *J Fungi* **2018**, *4*, 125.
118. Abado-Becognee, K.; Fleurat-Lessard, F.; Creepy, E.E.; Melcion, D. Effects of Fumonisin B1 on Growth and Metabolism of Larvae of the Yellow Mealworm, Tenebrio Molitor. *Entomologia Experimentalis et Applicata* **1998**, *86*, 135–143.
119. Mellanby, K. Water Content and Insect Metabolism. *Nature* **1958**, *181*, 1403–1403.

120. Murray, D.R.P. THE IMPORTANCE OF WATER IN THE NORMAL GROWTH OF LARVAE OF TENEBRIO MOLITOR. *Entomologia Experimentalis et Applicata* **1968**, *11*, 149–168, doi:10.1111/j.1570-7458.1968.tb02041.x.
121. Urs, K.C.D.; Hopkins, T.L. Effect of Moisture on Growth Rate and Development of Two Strains of Tenebrio Molitor L.(Coleoptera, Tenebrionidae). *Journal of Stored Products Research* **1973**, *8*, 291–297.
122. Rho, M.S.; Lee, K.P. Behavioural and Physiological Regulation of Protein and Carbohydrates in Mealworm Larvae: A Geometric Analysis. *Journal of Insect Physiology* **2022**, *136*, 104329, doi:10.1016/j.jinsphys.2021.104329.
123. Hanboonsong, A.; Durst, P. *Guidance on Sustainable Cricket Farming*; FAO: Bangkok, 2020; ISBN 9789251337394.
124. Kyllonen, T.; Manzanares, A. Insect Production and Operations Management. In *The basics of edible insect rearing*; Wageningen Academic Publishers: Wageningen, 2022.
125. Ortiz, J.C.; Ruiz, A.T.; Morales-Ramos, J.A.; Thomas, M.; Rojas, M.G.; Tomberlin, J.K.; Yi, L.; Han, R.; Giroud, L.; Jullien, R.L. Insect Mass Production Technologies. In *Insects as sustainable food ingredients*; Elsevier, 2016; pp. 153–201.
126. Fontanetti, C.S.; Zefa, E. Morphological Characterization of the Proventriculus of Gryllus Assimilis Fabricius (Orthoptera, Gryllidae). *Revista Brasileira de Zoologia* **2004**, *17*, 793–798.
127. Gangwere, S.K. A Monograph on Food Selection in Orthoptera. *Transactions of the American Entomological Society* **1961**, *87*, 67–230.
128. Hanboonsong, Y.; Jamjanya, T.; Durst, P.B. *Six-Legged Livestock: Edible Insect Farming, Collection and Marketing in Thailand*; Food and Agriculture Organization of the United Nations: Regional office for Asia and the Pacific, 2013; pp. 1–69;.
129. Gutierrez, Y.; Fresch, M.; Ott, D.; Brockmeyer, J.; Scherber, C. Diet Composition and Social Environment Determine Food Consumption,

Phenotype and Fecundity in an Omnivorous Insect. *Royal Society Open Science* **2020**, *7*.

130. Collavo, A.; Glew, R.; Huang, Y.; Chuang, L.; Bosse, R.; Paoletti, M. House Cricket Small-Scale Farming. In *Ecological implications of minilivestock*; CRC Press, 2005.
131. Nakagaki, B.J.; Defoliart, G.R. Comparison of Diets for Mass-Rearing Acheta Domesticus (Orthoptera:Gryllidae) as a Novelty Food, and Comparison of Food Conversion Efficiency with Values Reported for Livestock. *Journal of Economic Entomology* **1991**, *84*, 891–896.
132. Lundy, M.E.; Parrella, M.P. Crickets Are Not a Free Lunch: Protein Capture from Scalable Organic Side-Streams via High-Density Populations of Acheta Domesticus. *PLoS ONE* **2015**, *10*, e0118785, doi:10.1371/journal.pone.0118785.
133. Fernandez-Cassi, X.; Supeanu, A.; Vaga, M.; Jansson, A.; Boqvist, S.; Vagsholm, I. The House Cricket (Acheta Domesticus) as a Novel Food:A Risk Profile. *Journal of Insects as Food and Feed* **2019**, *5*, 137–157.
134. Sorjonen, J.M.; Valtonen, A.; Hirvisalo, E.; Karhapaa, M.; Lehtovaara, V.J.; Lindgren, J.; Marnila, P.; Mooney, P.; Maki, M.; Siljander-Rasi, H.; et al. The Plant-Based by-Product Diets for Mass-Rearing Acheta Domesticus and Gryllus Bimaculatus. *Plos One* **2019**, *14*.
135. Veldkamp, T.; Claeys, J.; Haenen, O.; van Loon, J.; Spranghers, T. *The Basics of Edible Insect Rearing*; Wageningen Academic Publishers: The Netherlands, 2021;
136. Durst, P.B.; Hanboonsong, Y. Small-Scale Production of Edible Insects for Enhanced Food Security and Rural Livelihoods: Experience from Thailand and Lao People’s Democratic Republic. *Journal of Insects as Food and Feed* **2015**, *1*, 25–31.
137. Miech, P. Cricket Farming: An Alternative for Producing Food and Feed in Cambodia. Doctoral Thesis, Swedish University of Agricultural Sciences, 2018.

138. Vandeweyer, D.; Wynants, E.; Crauwels, S.; Verreth, C.; Viaene, N.; Claes, J.; Lievens, B.; Van Campenhout, L. Microbial Dynamics during Industrial Rearing, Processing, and Storage of Tropical House Crickets (Gryllodes Sigillatus) for Human Consumption. *Applied and environmental microbiology* **2018**, *84*.
139. Morales-Ramos, J.A.; Rojas, M.G.; Dossey, A.T.; Berhow, M. Self-Selection of Food Ingredients and Agricultural by-Products by the House Cricket, Acheta Domesticus (Orthoptera:Gryllidae): A Holistic Approach to Develop Optimized Diets. *PLOS One* **2020**, *15*.
140. Patton, R.L. Oligidic Diets for Acheta Domesticus (Orthoptera:Gryllidae). *Annals of the Entomological Society of America* **1967**, *60*, 1238–1242.
141. Lyn, J.C.; Naikkhwah, W.; Aksenov, V.; Rollo, C.D. Influence of Two Methods of Dietary Restriction on Life History Features and Aging of the Cricket Acheta Domesticus. *Age* **2011**, *33*, 509–522.
142. Whattam, E.M.; Bertram, S.M. Effects of Juvenile and Adult Condition on Long-Distance Call Components in the Jamaican Field Cricket, Gryllus Assimilis. *Animal Behaviour* **2011**, *81*, 135–144, doi:10.1016/j.anbehav.2010.09.024.
143. Zera, A.J.; Harshman, L.G. The Physiology of Life History Trade-Offs in Animals. *Annual review of ecology and systematics* **2001**, *32*, 95–126.
144. Hunt, J.; Brooks, R.; Jennions, M.D.; Smith, M.J.; Bentsen, C.L.; Bussiere, L.F. High-Quality Male Field Crickets Invest Heavily in Sexual Display but Die Young. *Nature* **2004**, *432*, 1024–1027.
145. Reifer, M.L.; Harrison, S.J.; Bertram, S.M. How Dietary Protein and Carbohydrate Influence Field Cricket Development, Size and Mate Attraction Signalling. *ANimal Behaviour* **2018**, *139*, 137–146.
146. Bawa, M.; Songsermpong, S.; Kaewtapee, C.; Chanput, W. Effect of Diet on the Growth Performance, Feed Conversion, and Nutrient Content of the House Cricket. *Journal of insect science* **2020**, *20*, 10.

147. Visanuvimol, L.; Bertram, S.M. How Dietary Phosphorus Availability during Development Influences Condition and Life History Traits of the Cricket, Acheta Domesticus. *Journal of Insect Science* **2011**, *11*, 1–17.
148. Visanuvimol, L.; Bertram, S.M. Dietary Phosphorus Availability Influences Female Cricket Lifetime Reproductive Effort. *Ecological Entomology* **2010**, *35*, 386–395.
149. Finke, M.D.; Dunham, S.U.; Cole, J.S. Evaluation of Various Calcium-Fortified High Moisture Commercial Products for Improving the Calcium Content of Crickets, Acheta Domesticus. *Journal of Herpetological Medicine and Surgery* **2004**, *14*, 17–20.
150. Finke, M.D.; Dunham, S.U.; Kwabi, C.A. Evaluation of Four Dry Commercial Gut Loading Products for Improving the Calcium Content of Crickets, Acheta Domesticus. *Journal of Herpetological Medicine and Surgery* **2005**, *15*, 7–12.
151. Harrison, S.J.; Raubenheimer, D.; Simpson, S.J.; Godin, J.-G.J.; Bertram, S.M. Towards a Synthesis of Frameworks in Nutritional Ecology: Interacting Effects of Protein, Carbohydrate and Phosphorus on Field Cricket Fitness. *Proceedings of the Royal Society B: Biological Sciences Royal SOciety* **2014**, *281*.
152. Neuman-Lee, L.A.; Hopkins, G.R.; Brodie, E.D.; French, S.S. Sublethal Contaminant Exposure Alters Behaviour in a Common Insect: Important Implications for Trophic Transfer. *Journal of Environmental Science and Health, Part B* **2013**, *48*, 442–448.
153. Elwood, R.W.; Appel, M. Pain Experience in Hermit Crabs? *Animal Behaviour* **2009**, *77*, 1243–1246.
154. Sherwin, C.M. Can Invertebrates Suffer? Or, How Robust Is Argument-by-Analogy? *Animal Welfare* **2001**, *10*, 103–118.
155. Sneddon, L.U.; Elwood, R.W.; Adamo, S. a.; Leach, M.C. Defining and Assessing Animal Pain. *Animal Behaviour* **2014**, *97*, 201–212, doi:10.1016/j.anbehav.2014.09.007.

156. Smolinsky, A.N.; Bergner, C.L.; LaPorte, J.L.; Kalueff, A.V. Analysis of Grooming Behaviour and Its Utility in Studying Animal Stress, Anxiety and Depression. **2009**, 21–36.
157. Fudlosid, S.; Ritchie, M.W.; Muzzatti, M.J.; Allison, J.E.; Provencher, J.; MacMillan, H.A. Ingestion of Microplastic Fibres, but Not Microplastic Beads, Impacts Growth Rates in the Tropical House Cricket Gryllodes Sigillatus. *Frontiers in Physiology* **2022**, *13*.
158. Lofty, J.; Muhawenimana, V.; Wilson, C.A.M.E.; Ouro, P. Microplastics Removal from a Primary Settler Tank in a Wastewater Treatment Plant and Estimations of Contamination onto European Agricultural Land via Sewage Sludge Recycling. *Environmental Pollution* **2022**, *304*, 119198–119198.
159. Oliveri Conti, G.; Ferrante, M.; Banni, M.; Favara, C.; Nicolosi, I.; Cristaldi, A.; Fiore, M.; Zuccarello, P. Micro- and Nano-Plastics in Edible Fruit and Vegetables. The First Diet Risks Assessment for the General Population. **2020**, *Environmental Research*, 109677–109677.
160. Zheng, D.; Liu, X.; Jin, D.; Li, H.; Li, X. Mercury Bioaccumulation in Arthropods from Typical Community Habitats in a Zinc-Smelting Area. *Environmental Geochemistry and Health* **2018**, *40*, 1329–1337.
161. EFSA Risk Profile Related to Production and Consumption of Insects as Food and Feed. *EFSA Journal* **2015**, *13*, 4257, doi:10.2903/j.efsa.2015.4257.
162. van der Fels-Klerx, H.J.; Camenzuli, L.; Belluco, S.; Meijer, N.; Ricci, A. Food Safety Issues Related to Uses of Insects for Feeds and Foods. *Comprehensive Reviews in Food Science and Food Safety* **2018**, *17*, 1172–1183.
163. Argasinski, K.; Bednarska, A.; Laskowski, R. The Toxicokinetics Cell Demography Model to Explain Metal Kinetics in Terrestrial Invertebrates. *Ecotoxicology* **2012**, *21*, 2186–2194.

164. De Paepe, E.; Wauters, J.; Van Der Borght, M.; Claes, J.; Huysman, S.; Croubels, S.; Vanhaecke, L. Ultra-High-Performance Liquid Chromatography Coupled to Quadrupole Orbitrap High-Resolution Mass Spectrometry for Multi-Residue Screening of Pesticides, (Veterinary) Drugs and Mycotoxins in Edible Insects. *Food Chemistry* **2019**, *293*, 187–196.
165. Barrett, M.; Chia, S.Y.; Fischer, B.; Tomberlin, J.K. Welfare Considerations for Farming Black Soldier Flies, Hermetia Illucens (Diptera: Stratiomyidae): A Model for the Insects as Food and Feed Industry. *Journal of Insects as Food and Feed* **2022**, 1–30, doi:10.3920/JIFF2022.0041.
166. Megido, R.C.; Alabi, T.; Nieus, C.; Blecker, C.; Danthine, S.; Bogaert, J.; Haubruge, E.; Francis, F. Optimisation of a Cheap and Residential Small-Scale Production of Edible Crickets with Local by-Products as an Alternative Protein-Rich Human Food Source in Ratanakiri Province, Cambodia. *Journal of the Science of Food and Agriculture John Wiley & Sons, Ltd* **2016**, *96*, 627–632.
167. van Huis, A.; Tomberlin, J.K. *Insects as Food and Feed: From Production to Consumption*; Wageningen Academic Publishers: Wageningen, NL, 2016.
168. Oloo, J.A.; Ayieko, M.; Nyongesah, J.M. Acheta Domesticus (Cricket) Feed Resources among Smallholder Farmers in Lake Victoria Region of Kenya. *Food Science and Nutrition* **2020**, *8*, 69–78.
169. Reverberi, M. Edible Insects: Cricket Farming and Processing as an Emerging Market. *Journal of Insects as Food and Feed* **2020**, *6*, 211–220, doi:10.3920/jiff2019.0052.
170. Berridge, K. Food Reward: Brain Substrates of Wanting and Liking. *Neuroscience & Biobehavioral Reviews* **1996**.
171. Department of Primary Industries Biology and Behaviour of the Australian Plague Locust Available online:

https://www.dpi.nsw.gov.au/emergencies/locusts/locust-biology-and-behaviour/biology (accessed on 18 October 2023).

172. Egonyu, J.P.; Subramanian, S.; Tanga, C.M.; Dubois, T.; Ekesi, S.; Kelemu, S. Global Overview of Locusts as Food, Feed and Other Uses. *Global Food Security* **2021**, *31*, 100574, doi:10.1016/j.gfs.2021.100574.
173. Le Gall, M.; Overson, R.; Cease, A. A Global Review on Locusts (Orthoptera: Acrididae) and Their Interactions With Livestock Grazing Practices. *Frontiers in Ecology and Evolution* **2019**, *7*.
174. Song, H. Density-Dependent Phase Polyphenism in Nonmodel Locusts: A Minireview. *Psyche: A Journal of Entomology* **2010**, *2011*, e741769, doi:10.1155/2011/741769.
175. EFSA; Turck, D.; Castenmiller, J.; De Henauw, S.; Hirsch-Ernst, K.I.; Kearney, J.; Maciuk, A.; Mangelsdorf, I.; McArdle, H.J.; Naska, A.; et al. Safety of Frozen and Dried Formulations from Migratory Locust (Locusta Migratoria) as a Novel Food Pursuant to Regulation (EU) 2015/2283. *EFSA Journal* **2021**, *19*, e06667, doi:10.2903/j.efsa.2021.6667.
176. Brosemann, J.; Overson, R.; Cease, A.J.; Millerwise, S.; Le Gall, M. Nutrient Supply and Accessibility in Plants: Effect of Protein and Carbohydrates on Australian Plague Locust (Chortoicetes Terminifera) Preference and Performance. *Frontiers in Insect Science* **2023**, *3*.
177. Talal, S.; Cease, A.J.; Youngblood, J.P.; Farington, R.; Trumper, E.V.; Medina, H.E.; Rojas, J.E.; Fernando Copa, A.; Harrison, J.F. Plant Carbohydrate Content Limits Performance and Lipid Accumulation of an Outbreaking Herbivore. *Proc Biol Sci* **2020**, *287*, 20202500, doi:10.1098/rspb.2020.2500.
178. Dadd, R.H. The Nutritional Requirements of Locusts—I Development of Synthetic Diets and Lipid Requirements. *Journal of Insect Physiology* **1960**, *4*, 319–347, doi:10.1016/0022-1910(60)90057-3.

179. Simpson, S.J.; Abisgold, J.D. Compensation by Locusts for Changes in Dietary Nutrients: Behavioural Mechanisms. *Physiological Entomology* **1985**, *10*, 443–452, doi:10.1111/j.1365-3032.1985.tb00066.x.
180. Talal, S.; Parmar, S.; Osgood, G.M.; Harrison, J.F.; Cease, A.J. High Carbohydrate Consumption Increases Lipid Storage and Promotes Migratory Flight in Locusts. *Journal of Experimental Biology* **2023**, *226*, jeb245351, doi:10.1242/jeb.245351.
181. Chyb, S.; Simpson, S. j. Dietary Selection in Adult Locusta Migratoria. *Entomologia Experimentalis et Applicata* **1990**, *56*, 47–60, doi:10.1111/j.1570-7458.1990.tb01380.x.
182. Despland, E.; Simpson, S.J. Food Choices of Solitarious and Gregarious Locusts Reflect Cryptic and Aposematic Antipredator Strategies. *Animal Behaviour* **2005**, *69*, 471–479, doi:10.1016/j.anbehav.2004.04.018.
183. Simpson, S.J.; Raubenheimer, D.; Behmer, S.T.; Whitworth, A.; Wright, G.A. A Comparison of Nutritional Regulation in Solitarious- and Gregarious-Phase Nymphs of the Desert Locust Schistocerca Gregaria. *Journal of Experimental Biology* **2002**, *205*, 121–129, doi:10.1242/jeb.205.1.121.
184. Srygley, R.B. Diet Drives the Collective Migrations and Affects the Immunity of Mormon Crickets and Locusts: A Comparison of These Potential Superspreaders of Disease. *Integrative and Comparative Biology* **2016**, *56*, 268–277, doi:10.1093/icb/icw035.
185. EFSA Risk Profile Related to Production and Consumption of Insects as Food and Feed. *EFSA Journal* **2015**, *13*, doi:10.2903/j.efsa.2015.4257.
186. Wang, Q.; Liu, Y.; Yin, X. Comparison of Gut Bacterial Communities of Locusta Migratoria Manilensis (Meyen) Reared on Different Food Plants. *Biology (Basel)* **2022**, *11*, 1347, doi:10.3390/biology11091347.
187. Choi, J.-S. Nutrition, Safety, Health Functional Effects, and Availability of Honeybee (Apis Mellifera L.) Drone Pupae. *Insects* **2021**, *12*, 771, doi:10.3390/insects12090771.

188. Britannica, T. Honeybee Available online: https://www.britannica.com/animal/honeybee (accessed on 7 October 2023).
189. Schukraft, J. Managed Honey Bee Welfare: Problems and Potential Interventions Available online: https://rethinkpriorities.org/publications/managed-honey-bee-welfare-problems-and-potential-interventions (accessed on 12 October 2023).
190. Jensen, A.B.; Evans, J.; Jonas-Levi, A.; Benjamin, O.; Martinez, I.; Dahle, B.; Roos, N.; Lecocq, A.; Foley, K. Standard Methods for Apis Mellifera Brood as Human Food. *Journal of Apicultural Research* **2019**, *58*, 1–28, doi:10.1080/00218839.2016.1226606.
191. Schiel, L.; Wind, C.; Ulmer, M.; Braun, P.G.; Koethe, M. Honey Bee Drone Brood Used as Food. *Peer Review* **2022**.
192. IPIFF *Regulation (EU) 2015/2283 on Novel Foods: Briefing Paper on the Provisions Relevant to the Commercialisation of Insect-Based Products Intended for Human Consumption in the EU*; IPIFF, 2021.
193. Ricigliano, V.A.; Williams, S.T.; Oliver, R. Effects of Different Artificial Diets on Commercial Honey Bee Colony Performance, Health Biomarkers, and Gut Microbiota. *BMC Veterinary Research* **2022**, *18*, 52, doi:10.1186/s12917-022-03151-5.
194. Ricigliano, V.A.; Cank, K.B.; Todd, D.A.; Knowles, S.L.; Oberlies, N.H. Metabolomics-Guided Comparison of Pollen and Microalgae-Based Artificial Diets in Honey Bees. *J. Agric. Food Chem.* **2022**, *70*, 9790–9801, doi:10.1021/acs.jafc.2c02583.
195. DeGrandi-Hoffman, G.; Chen, Y.; Rivera, R.; Carroll, M.; Chambers, M.; Hidalgo, G.; de Jong, E.W. Honey Bee Colonies Provided with Natural Forage Have Lower Pathogen Loads and Higher Overwinter Survival than Those Fed Protein Supplements. *Apidologie* **2016**, *47*, 186–196, doi:10.1007/s13592-015-0386-6.

196. Kumar, R.; Agrawal, O.P. Comparative Performance of Honeybee Colonies Fed with Artificial Diets in Gwalior & Panchkula Region. *Journal of Entomology and Zoology Studies* **2014**.
197. Kumari, I.; Kumar, R. Pollen Substitute Diet for Apis Mellifera: Consumption and Effects on Colony Parameters in Sub-Tropical Himalaya. *Indian Journal Of Agricultural Research* **2019**, doi:10.18805/IJARe.A-5369.
198. Everything Silkworms Available online: https://everythingsilkworms.com.au/ (accessed on 9 October 2023).
199. Britannica, T.E. of E. Silkworm Moth Available online: https://www.britannica.com/animal/silkworm-moth (accessed on 5 October 2023).
200. Mahanta, D.; Komal, J.; Samal, I.; Bhoi, T.; Dubey, V.; Pradhan, K.; Nekkanti, A.; Gouda, M.; Saini, V.; Negi, N.; et al. Nutritional Aspects and Dietary Benefits of “Silkworms”: Current Scenario and Future Outlook. *Frontiers in Nutrition* **2023**, *19*, 1121508.
201. *Forest Insects as Food: Humans Bite Back ; Proceedings of a Workshop on Asia-Pacific Resources and Their Potential for Development, 19-21 February 2008, Chiang Mai, Thailand*; Durst, P.B., FAO, Eds.; RAP publication; Food and Agriculture Organization of the United Nations, Regional Office for Asia and the Pacific: Bangkok, Thailand, 2010; ISBN 978-92-5-106488-7.
202. Wu, X.; He, K.; Velickovic, T.C.; Liu, Z. Nutritional, Functional, and Allergenic Properties of Silkworm Pupae. *Food Sci Nutr* **2021**, *9*, 4655–4665, doi:10.1002/fsn3.2428.
203. Zhou, J.; Han, D. Safety Evaluation of Protein of Silkworm (Antheraea Pernyi) Pupae. *Food and Chemical Toxicology* **2006**, *44*, 1123–1130, doi:10.1016/j.fct.2006.01.009.
204. Zhang, Z.-J.; Zhang, S.-S.; Niu, B.-L.; Ji, D.-F.; Liu, X.-J.; Li, M.-W.; Bai, H.; Palli, S.R.; Wang, C.-Z.; Tan, A.-J. A Determining Factor for Insect Feeding

Preference in the Silkworm, Bombyx Mori. *PLoS Biol* **2019**, *17*, e3000162, doi:10.1371/journal.pbio.3000162.

205. Takeda, S. Chapter 232: Sericulture. In *Encyclopedia of Insects (Second Edition)*; 2009; pp. 912–914.
206. Panthee, S.; Paudel, A.; Hamamoto, H.; Sekimizu, K. Advantages of the Silkworm As an Animal Model for Developing Novel Antimicrobial Agents. *Frontiers in Microbiology* **2017**, *8*.
207. Nikolova, T. Growing Mulberry Silkworm with Artificial Diet with Added Extract Tribulus Terrestris L. *Bulgarian Journal of Agricultural Science* **2020**, *26*, 1041–1046.
208. Qin, D.; Wang, G.; Dong, Z.; Xia, Q.; Zhao, P. Comparative Fecal Metabolomes of Silkworms Being Fed Mulberry Leaf and Artificial Diet. *Insects* **2020**, *11*, 851, doi:10.3390/insects11120851.
209. Qin, L.; Qi, J.; Shen, G.; Qin, D.; Wu, J.; Song, Y.; Cao, Y.; Zhao, P.; Xia, Q. Effects of Microbial Transfer during Food-Gut-Feces Circulation on the Health of Bombyx Mori. *Microbiol Spectr* **2022**, *10*, e02357-22, doi:10.1128/spectrum.02357-22.
210. Zhou, Z.-H.; Yang, H.-J.; Chen, M.; Lou, C.-F.; Zhang, Y.-Z.; Chen, K.-P.; Wang, Y.; Yu, M.-L.; Yu, F.; Li, J.-Y.; et al. Comparative Proteomic Analysis between the Domesticated Silkworm (Bombyx Mori) Reared on Fresh Mulberry Leaves and on Artificial Diet. *J Proteome Res* **2008**, *7*, 5103–5111, doi:10.1021/pr800383r.
211. Rahmathulla; Nayak, P.; Vindya; Himantharaj; Rajan Influence of Antibiotic on Feed Conversion Efficiency of Mulberry Silkworm (Bombyx Mori L.). *Animal Biology* **2006**, *56*, 13–22, doi:10.1163/157075606775904731.

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