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Incorporating whole insect larvae into poultry diets: state of the art and future perspectives

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ABSTRACT

A wide range of insects are being considered as potential candidate sources of dietary protein; however, it was only recently that the European Union, with Regulation (EU) 2017/893 of May 24, first allowed the use of the black soldier fly (Hermetia illucens), the common housefly (Musca domestica), the yellow mealworm (Tenebrio molitor), the lesser mealworm (Alphitobius diaperinus), the house cricket (Acheta domesticus), the banded cricket (Gryllodes sigillatus) and the field cricket (Gryllus assimilis) as feed in aquaculture. With Regulation (EU) 2021/1372 of August 17. their use was extended to poultry and pig feed. Most of the studies on the use of insects in poultry feeding have focused on the benefits of insect meals. However, live insects are part of the natural diet of poultry, so more effort should be directed at evaluating their use further, not only in relation to their effect on poultry performance and product quality, but also from the perspective of bird welfare. No adverse effects have been observed with the dietary inclusion of live larvae at up to 10% of the daily feed intake. However, further research is needed to establish the inclusion levels that are most advantageous for the bird's health, economically sustainable for the breeder and without the unnecessary waste of resources. Live larvae provide fresh, unaltered nutrients and they stimulate birds to express innate behaviours which may improve the birds' welfare. The production and distribution of live larvae may be laborious and expensive, especially for large-scale production systems. Dehydrated larvae are easier to handle, and no complex protocols need to be implemented prior to offering to the bird. By contrast, dehydrated larvae are potentially less attractive to birds.

HIGHLIGHTS

- The use of insects in poultry feed has received significant interest as a potential solution to improve the sustainability of poultry diets.
- Whole insect larvae (live, dehydrated or defrosted) are a promising tool to improve poultry welfare by inducing foraging behaviour and stimulating greater activity.
- The use of dehydrated larvae is promising because of easy handling and biosecurity issues.
- The methods of distributing the whole larvae to the birds need to be customised, thus the poultry equipment manufacturers should expand their market by producing specific automatised dispenser for live or dehydrated insect larvae.

Introduction

The use of insects in poultry feed has received significant interest as a potential solution to improve the sustainability of poultry diets. However, the European Union has limited their use for feed purposes to specific insect species through the introduction of Regulation (EU) 2017/893 of 24 May (European Commission Directorate-General for Health and Food Safety 2017). This law foresees the use of the black soldier fly (BSF, *Hermetia illucens*), the common housefly (HF, *Musca domestica*), the yellow mealworm (YM, Tenebrio molitor), the lesser mealworm (LM, Alphitobius diaperinus), the house cricket (HC, Acheta domesticus), and the banded cricket (BC, Gryllodes sigillatus) and the field cricket (FC, Gryllus assimilis) in aquaculture. The subsequent Regulation (EU) 2021/1372 of 17 August (European Commission Directorate-General for Health and Food Safety 2021) extended the use of these insect species to poultry and pig feedstuffs. At the present time, in Europe, the use of whole dead insects is still not allowed in the production of feed destined to food-producing animals (Regulation EU 1069/2009). The growing interest in the use of insects

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for feed production is also demonstrated by the presence of research projects funded by the European Union promoting innovative and sustainable dietary protein sources and farming systems, in which insects play a key role (Proteinsect 2013; SUSINCHAIN 2019; Poultrynsect 2021; SUSTavianFEED 2021; Advagromed 2022), and by the numerous papers published in the past ten years regarding the benefits that diets containing insects might offer poultry livestock (Gasco et al. 2019; Elahi et al. 2022; Veldkamp et al. 2022; Martínez Marín et al. 2023).

Insects are a highly versatile primary material, as they can be processed and used in various forms, all of which are being tested, including meals (Schiavone et al. 2019; Heuel et al. 2021; Zhao et al. 2022) and fats (Schiavone et al. 2018; Cullere et al. 2019), to promote theit use as source of protein or energy, respectively. Furthermore, whole larvae has been tested as frozen (Moula, Hornick, et al. 2018; Moula, Scippo, et al. 2018; Seyedalmoosavi et al. 2022), dehydrated (Hwangbo et al. 2009; Traore et al. 2020a, 2020b; Ipema et al. 2022), or live (Veldkamp and van Niekerk 2019; Star et al. 2020; Tahamtani et al. 2021; Biasato et al. 2022; Gariglio et al. 2023). Insects form part of the natural diet of poultry, so including live or dehydrated whole insect larvae might offer not only nutritional benefits to the animals but also improve bird welfare (Appleby et al. 2004). The dietary use of live insects may also bring about potential health risks, which may be prevented if appropriate precautions and management measures are taken (Maciel-Vergara and Ros 2017; Maciel-Vergara et al. 2021; Time et al. 2022). For example, insects fed on manure rather than a diet based on vegetable waste are more exposed to pathological agents, which are easily transferred to the bird when eaten (Holt et al. 2007; Hazeleger et al. 2008; Leffer et al. 2010). According to the feeding substrate, some insects tend to accumulate heavy metals, although regular monitoring of the substrate should prevent or reduce this risk (Purschke et al. 2017; Malematja et al. 2023).

Concerning the use of live and/or dehydrated whole insect larvae, their use stand to offer additional benefits by stimulating birds to become more active, evoking elements of natural foraging behaviour, and contribute towards improving their state of welfare and production performances. Nonetheless, most studies to date have concentrated on the use of meals (Biasato et al. 2019; Gariglio et al. 2019; Sayed et al. 2019; Schiavone et al. 2019; Heuel et al. 2021; Zhao et al. 2022; Nieto et al. 2023) and very few have tested bird responses and the potential beneficial effects of the dietary inclusion of live and/or dehydrated whole insect larvae, or addressed the practical aspects of the supplementation (Veldkamp and van Niekerk 2019; Ipema, Gerrits, et al. 2020; Star et al. 2020; Bellezza Oddon et al. 2021; Colombino et al. 2021; Tahamtani et al. 2021; Biasato et al. 2022; Ipema et al. 2022; Bongiorno et al. 2022; Gariglio et al. 2023). The present review aims to summarise the benefits reported to arise from this practice and to outline the aspects in most need of further research. It also provides some practical recommendations.

Poultry feeding behaviour

The established, traditional systems for poultry management are often deemed to be the best systems by farmers, but these practices do not usually take into account species-specific knowledge about natural behaviours, the expression of which is needed to guarantee a high level of bird welfare. In the case of domestic fowl, it should be kept in mind that these species exhibit behaviours highly similar to their wild ancestors; indeed, none of their ancestors' behavioural patterns have been lost and no new behaviours introduced, although learning processes and bird-environment interactions are necessary for them to learn and express specific behaviours (Appleby et al. 2004).

Wild and feral poultry dedicate the majority of the day to foraging for food, covering many kilometres a day within a well-defined and familiar area (Appleby et al. 2004). Foraging behaviour also permits a bird to achieve a balanced diet by leading them to feed on a wide range of food items, including seeds, herbage, invertebrates and fruit (Appleby et al. 2004). Given the opportunity to do so, domestic birds are similarly capable of seeking and selecting appropriate food items to fulfil their nutritional requirements (Hughes and Whitehead 1979; Appleby et al. 2004).

When foraging, birds peck and scratch at the ground, eating the various food sources it comes across. In the case of ducks, the birds use their bills to sort out food items from the water (Coppinger 1970; Moreby et al. 2006; Mench 2009), and when they find hard, unusual or an unknown tasty food, they use it to soak these objects in the water to render it more edible (Castillo et al. 2020). The time dedicated to feeding might be influenced by the type of food the bird is able to find and consume in order to satisfy its needs. In the case of edible invertebrates, some authors have associated the availability of this food source to bird food preferences, but it is known that certain elements also condition the birds' choices,

such as food colour, size, movement and consistency (Green 1984). Food selection also seems to be guided by visual and tactile senses. In the case of newly hatched chicks, the birds do not immediately recognise food items, though they are prompt to peck, and this behaviour, combined with the hen's call, makes them learn to recognise edible items (Evans 1975; Appleby et al. 2004). In domestic fowl, studies have demonstrated that sweet flavours are not particularly attractive, and acidic, bitter and salty food items are generally rejected. Indeed, according to the food taste, the bird is able to refuse potentially toxic foodstuffs (Halpern 1962; Appleby et al. 2004). How the bird behaves when it finds an insect will depend on the characteristics of the insect itself, how it is presented, and the bird's previous experience of it. To this regard, various conditions have been tested; for instance, grev partridge chicks offered dead insects preferred those larger in size and of a green-yellow colour (Moreby et al. 2006); whereas when offered red-black or greenyellow ant pupae, the latter colours were preferred (Potts 1986; Moreby et al. 2006). Quails, on the other hand, avoid red coloured insects, preferring green or black arthropods (Moreby et al. 2006). Moreover, grey partridge chicks were observed to reject certain dead insect species (Lasius sp., Bembidion lampros. and Lema melanophera), whereas they were guickly eaten up if presented live. Pheasants were found to prefer moderately large live insects (Lygus lineolaris, Adelphocoris lineolatus) over small or very large ones (moths and grasshoppers, respectively) (Whitmore et al. 1986). As described in previous reports, the prey handling procedures required may also influence the bird's preferences. For example, for chicks to be able to eat a whole mealworm they need to perform a complex sequence of actions, thus they tend to prefer chickbill-size preys that can be eaten entirely at once (Moreby et al. 2006). Studies have also shown that unknown food items are often aversive to birds, and that the degree of rejection is related to the birds' previous experiences and how the food differs from the one the bird is used to Coppinger (1970).

As discussed above, avian feeding behaviour is a vast and complex topic which farmers and breeders, and indeed all those dealing with poultry breeding and production, should deepen their knowledge of in order to gain a better understanding of a certain bird's behaviours. This would equip them to manage feed distribution systems better, according to that species' specific needs. This concept should be applied not only to feeding but to all aspects of bird biology and husbandry since it stands to benefit both the bird and the farmer.

Whole insect larvae production and processing

Insects are proficient at converting agricultural wastes into animal protein, drastically reducing gas emissions and waste mass (Veldkamp et al. 2022). Insects have a high protein and fat content and provide a source of other important nutrients such as macro and micro minerals and vitamins. For instance, the crude protein values of insects' larvae range from 35% to 57% for black soldier flv, from 43%-68% for the housefly, and from 44% to 69% for the yellow mealworm. Crude fat is at around 35% in the black soldier fly, 4%-32% in the housefly, and 23%-47% in the vellow mealworm (Veldkamp et al. 2012; Hawkey et al. 2021). The nutritional composition of the insect depends on the diet fed to the insect and its developmental stage (Hawkey et al. 2021; Oonincx and Finke 2021; Sverguzova et al. 2023). Insects also contain bioactive compounds such as antimicrobial peptides, fatty acids and polysaccharides, which might provide their consumers with protection against oxidative tissue damage and defend against microbial threats (Veldkamp et al. 2022).

In Europe, the limited use of just a few insect species in feedstuffs destined for poultry species is primarily associated with the current legislation and safety matters, gaps in our knowledge about the suitability of insect species for different poultry species, the adaptability of insect species to being reared on specific substrates and/or under controlled conditions, and practical issues associated with the management of large-scale production systems (Van Huis 2016; Bjone and Fitches 2021). As mentioned above, the main insect species currently used are: BSF (order Diptera) at the larval and prepupae stages and HF at the larval stage; YM (order Coleoptera) at the larval stage; and adult HC (order Orthoptera) (Van Huis 2016).

Environmental conditions and the quality of the diet influence not only the length of the insect's life stages but also their nutritional value (Makkar et al. 2014; Gold et al. 2020; Bjone and Fitches 2021). The larval stage duration for BSF employs 13 days–5 months, 4–7 days for HF, and 3–4 months for YM (Bjone and Fitches 2021). HC becomes an adult at 70 days (Bjone and Fitches 2021). The advantages and disadvantages associated with different insect species depend on various circumstances and the intrinsic characteristics of each individual species. For instance,

HF larvae undergo a significantly faster development compared to BSF larvae; however, their pupal weight is only one-fifth that of BSF pupae (Geden et al. 2021; Salam et al. 2022). For in-depth information on the breeding and use of the various insect species, other dedicated reviews are available (Rumpold and Schlüter 2013; Makkar et al. 2014; Gasco et al. 2019; Bjone and Fitches 2021; Elahi et al. 2022; Van Huis 2022; Liceaga 2022).

Another important matter concerning the supplementation of poultry diets with live insects concerns the logistics of having larvae in an alive state at the right time. Inducing larvae to enter diapause can aid this problem as it extends the period in which they can be kept alive. In the case of BSF larvae (BSFL), diapause is induced at 16°C (Holmes et al. 2016); whereas for YM larvae (YML), a temperature of 6°C is required (Bellezza Oddon et al. 2021). To end diapause and supply the larvae to the birds in an 'alive' state, their metabolism must be reactivated, achieved by exposing them to a temperature of 28°C for 10 min (Bellezza Oddon et al. 2021).

Whole larvae might also be offered in the dehydrated state, making them easier to handle and store, though some nutritional properties of the larvae might change depending on the drying processing conditions and the insect species used (Liceaga 2021). The main drying techniques used are oven-drying, microwave-drying, freeze-drying, and fluidised bed drying (Kröncke et al. 2018; Melgar-Lalanne et al. 2019). For instance, the method mainly used for drying YML on the industrial scale is freeze-drying, which assures a product with a good nutritional value, texture, colour and aroma, but the disadvantage of this method is that it requires long processing times, it is costly, and, in larvae with a high fat content, the lipids are susceptible to oxidation (Kröncke et al. 2018; Melgar-Lalanne et al. 2019). However, only small differences were observed in the nutritional value of YML dehydrated by different drying techniques compared with those of fresh larvae (Kröncke et al. 2018). An additional aspect which should be considered is the safety of the dehydrated larvae, which should be guaranteed by the processing method (Wakefield et al. 2021). Very little is known about the optimised processing conditions for drying whole larvae of different insect species and the effects on the end-products; therefore, further research should be directed towards clarifying these matters.

Freezing the larvae is another way of preserving them until they are offered to the bird. However, the characteristics required of the final product, the type of freezing process adopted and the related safety concerns should again be considered on an insect-byinsect basis. Furthermore, the cold chain cannot be interrupted, thus suitable transport and storage conditions are needed. The temperature at which larvae freeze may also differ according to the insect species involved; for instance, a temperature of -20°C was shown to be insufficient to freeze whole LM larvae fully as certain enzymes remained active, leading to browning reactions (Wessels et al. 2020). In fact, the ice melting point of these larvae was -32.5 °C (Wessels et al. 2020). The activation of certain enzymatic pathways, such as the melanisation process, was also observed in BSFL subjected to the same freezing temperature, which again resulted in changes to the nutritional quality of proteins (Leni et al. 2019). Differences in insect protein functionalities and colour development in LM and YM stored at -18 °C have also been reported (Janssen 2018; Wessels et al. 2020). Once again, further studies are needed to identify the freezing conditions which result in the stable storage insect larvae species for specific of different applications.

Finally, as the demand for insects is in constant expansion, intensive production systems are likely going to be the ones undergoing the most growth, and in the same way that the welfare of vertebrate species is an important concern for livestock production systems, insect welfare at all life cycle stages should also be considered, necessitating the adoption of suitable management practices and protocols (Delvendahl et al. 2022; Barrett and Fischer 2023; Klobučar and Fisher 2023).

Farmers' perceptions about the feeding of domestic poultry with insects

The extent to which farmers are willing to include live insects in their birds' diets might depend on different factors. For instance, while farmers expressed interest in the use of insects in poultry diets, they expressed less willingness at the idea of rearing their own insects (Sebatta et al. 2018). When farmers keep birds under free-range systems, the birds already have the opportunity to scavenge for insects, so the advantages that might be gained from providing additional insects in their feed are much less or indeed absent. Farmers are aware of the nutritional value of insects, and those rearing their birds under semi- or fully-intensive systems were also those most likely to be interested in buying insect-based feeds (Sebatta et al. 2018; Chia et al. 2020). The need to engage with complex new technologies, such as insect rearing practices, might discourage farmers from using live insects as a feedstuff, mainly due to a lack of knowledge about these practices (Selaledi et al. 2021). Indeed, the ease of handling and storage of dehydrated larvae makes them more advantageous. Dehydrated larvae remain a product with good nutritive value, and although birds might find them less attractive than live larvae, the farmer's work would undoubtfully be simplified. That said, the prices of dehydrated larvae are still too high. However, considering the continuous increase in demand for insects as feedstuffs, the number of companies producing them will also hopefully increase to meet this demand, enabling their prices to drop too. Nevertheless, all these recent considerations necessitate additional research into the potential utilisation of dehydrated larvae, addressing both the perspectives of farmers and animals.

Consumers' perceptions about the feeding of domestic poultry with insects

Consumers were shown to support the use of dietary insects in livestock practices (Proteinsect 2013; Menozzi et al. 2021; Rumpold 2021; Spartano and Grasso 2021; Sogari et al. 2022), and do not, therefore, present an obstacle to their use. Preferences on the use of insects in poultry diets might be influenced by a wide variety of factors, such as the product price, the degree of knowledge about insect production practices and the benefits which might be derived from their use, animal welfare, environmental issues, social and cultural contexts, disgust towards the idea of feeding birds with insects, and potential changes in the taste of the resulting products obtained from these birds (Rumpold 2021; Spartano and Grasso 2021). For instance, eggs from hens receiving insects as part of their diet were well accepted in the UK (Spartano and Grasso 2021), mainly motivated by the derived environmental benefits and the potential to improve bird welfare. A similar outcome emerged in Italian consumers regarding meat from ducks being fed insects as part of their diet (Sogari et al. 2022). However, as evidenced by Rumpold (2021), consumers have not traditionally been aware about the feed ingredients in the livestock sector, so informing the consumer that the product they are buying was produced with dietary insects might present a useful marketing strategy to encourage customers to buy those products.

Welfare implication and biological effects of dietary whole insect larvae supplementation in poultry feeding practice

Studies on dietary insects have mainly focused on broiler chickens, evaluating the ideal quantities to offer, insect species, the modality of supply, and the benefits derived from their use. The following insect species have been studied for their inclusion in the diet of broiler chicken: BSF (Ipema, Bokkers, et al. 2020; Ipema, Gerrits, et al. 2020; Bellezza Oddon et al. 2021; Colombino et al. 2021; Biasato et al. 2022; Ipema et al. 2022; Seyedalmoosavi et al. 2022), YML (Pichova et al. 2016; Bellezza Oddon et al. 2021; Colombino et al. 2021; Biasato et al. 2022), HF (Hwangbo et al. 2009), and LM (Despins and Axtell 1995). Most of which involved the dietary inclusion of live larvae (Tables 1 and 2), with the exception of the study by Hwangbo et al. (2009), who used dehydrated HF larvae, and Ipema et al. (2022), who used both live and dehydrated BSFL. Live BSFL were also tested in slow growing chickens (Bongiorno et al. 2022), autochthonous chicken (Dankwa et al. 2002; Star et al. 2020; Fiorilla et al. 2023), laying hens (Tahamtani et al. 2021), turkeys (Veldkamp and van Niekerk 2019), and in ducks (Gariglio et al. 2023) (Table 2). In addition to BSFL, YML were also included in the testing of ducks (Gariglio et al. 2023).

Other authors (Moula, Scippo, et al. 2018; Moula, Hornick, et al. 2018; Sevedalmoosavi et al. 2022) employed defrosted whole BSF larvae. In studies in which dehydrated BSFL were offered to chicks, some of them were initially observed to avoid the larvae. Dehydrated larvae are rigid, and the authors hypothesised that this may render them difficult to eat, and their taste and smell may also have been altered, rendering them being less appetising to the birds (Ipema et al. 2022). Very young chicks tend to prefer very small items to peck at, so the relatively large dimensions of larvae might have contributed towards their not being consumed by all birds (Hogan 1973; Appleby et al. 2004). Indeed, the physical characteristics of food items (particle size, taste, smell, and colour) affect their palatability to birds (Appleby et al. 2004). Moreover, chicks are naturally inclined to peck at items with a glossy aspect—a preference which helps them locate sources of water and stay hydrated. Dry larvae lack the glossy aspect of live specimens, and this together with the fact that they are inert may render them less attractive to chicks (Mench 2009). However, in a study by Fiorilla et al. (2023), no distinction in preference between live and dehydrated BSFL among autochthonous chickens was observed. This

| sented are a | l indicated, to | ogether with the | observed effects | on the birds. | טווט משב, טובנמוץ וווט | מאטון ומנכי טוטאואטון וובקעבורטי מות נווב ווובנווטת מווטעטון אוווכון נווב | ב ומו אמב אאבוב לחוב- |
|-----------------|---------------------|--------------------|-----------------------|-----------------|----------------------------|--|---|
| Insect sp. | Age (d) | Quantity | | Freq. | Method | Effects | Reference |
| BSFL | 0-35 | 8% | DFI (DM) | 4 times | Scattered | \uparrow activity / > interesting than dehydrated larvae due to movement, odour, palatable / < daily pellet intake / > final bw and dm conversion ratio / hock burn incidence | (lpema et al. 2022) |
| BSFL | 4-38 | 596 | DFI (DM) | once | Plate | foraging / †birds activity / ↓ behaviours attributable to frustration/ no effects on plumage status, leg health, excreta corticosterone metabolites/ improved fcr / final bw and bwg not affected / no effect on slaughter performance / no negative effect on haematological and serum parameters / no histopathological alterations / intestinal morphometric indices unaffected / ↑ relative spleen weight/ slight improvement in caecal microbiota / no negative effect on mucin | (Bellezza Oddon et al. 2021; Colombino et al. 2021; Biasato et al. 2022) |
| BSFL | 0-42 | 5% | DFI (DM) | 4 times | Scattered | composition or local immune response activity due to foraging/ improved welfare even at commercial stocking density / no effect on foot pad dermatitis or hock burn | (Ipema, Bokkers, et al. 2020) |
| | | 5% | | 7 times | | f activity due to foraging / improved welfare even at commercial stocking density / ↓ fearfulness / temporarily ↓ growth rate / no infuence on final bw / no effect on foot pad dermatitis or hock burn | |
| | | 10% | | 4 times | | incidence 1 activity due to foraging / temporarily ↓ growth rate / no influence on 6nal hur incidance | |
| BSFL | 0-42 | 5% | DFI (DM) | 2 times | Scattered | The provided and the section of the provided the provided of the provided of the provided provided the provided | (Ipema, Gerrits, |
| | | 5% | | 4 times | | ↓ resulting / watking and being active ↓ over time ↑ active behaviours (walking, standing idle, ground pecking, foraging) / ↓ resting / walking and being active ↓ over time / improved leg | EL 41. 2020) |
| | | 10% | | 2 times | | 1 active behaviours (walking, standing idle, ground pecking, foraging) / resting / walking and being active over time | |
| | | 10% | | 4 times | | most effective at promoting activity, \downarrow occurrence of hock burn and lameness / \downarrow resting / improved leg health / similar final weight as controls despite temporary period of \downarrow growth / walking and being | |
| ЛМК | 4-38 | 5% | DFI (DM) | once | Plate | active lasted longer \uparrow foraging / \uparrow birds activity/ preference for yml over bsfl / \downarrow frustration behaviours / no effect on plumage status, leg health or excreta corticosterone metabolites/ no effect on final bw or bwg / no effect on slaughter performance / no effect on haematological or serum parameters / no histopathological alterations / \uparrow spleen relative weight/ slight improvement in caecal microbiota / no negative effect | (Bellezza Oddon et al. 2021; Colombino et al. 2021; Biasato et al. 2022) |
| УМL | 6–18 | 10g/ bird/d | as fed basis | once | Scattered | on mucin composition or local immune response | (Pichova et al. |
| ΓW | 3-8 | ad libitum | I | once | In litter | fear-reducing effect, tonic immobility test or novel object test chicks had no access to feed, eating exclusively larvae, which caused | (Despins and |
| | 2–9 | | | | Feed and litter | distress and no weight gain feed consumption, 10 to 35 % of protein intake from larvae / no effect on hwo | Axtell 1995) |
| BSFL = black sc | oldier fly larva; Y | ML = yellow mealwo | irm larvae; LM = less | er mealworm; Df | FI = daily feed intake; DI | A = dry matter; FCR = feed conversion ratio; BW = body weight; BWG = body we | eight gain. |

Table 2. Live larvae inclusion in poultry diets: the insect species, bird species and bird age, dietary inclusion rate, provision frequency, and the method through which the larvae on the hirds the observed effects tooether with all indicated nresented are WPre

| ארור הורזי | | מובמורמ, וספרוורו או | | | | | | |
|--------------|--------------------|-----------------------|----------------------|------------------|------------|---------------|--|----------------------------|
| Insect sp. | Bird | Age (d) | Quantity | | Freq. | Method | Effects | Reference |
| BSFL | Laying hen | 126–210 | 10% | DFI (DM) | Once | Bowels | no 1 feed consumption / no influence on egg production, egg weight, shell thickness, gizzard weight, liver weight, bird fearfulness | (Tahamtani et al. 2021) |
| | | | 20% | | | | more than 25 % L in feed consumption / no influence on egg production, egg | |
| | | | Ad lik | bitum | | | weight, shell thickness, gizzard weight, ilver weight of bird reartuiness live larvae consumption approx. 52 % DFI (DM) / \downarrow feed consumption after | |
| | | | | | | | 21 weeks of age / fdaily BWG / no influence on egg production, egg weight, shell thickness, gizzard weight, liver weight or bird fearfulness / fabdominal | |
| RCEI | Turkey | 0_35 | 17 0% | DEI (frach) | Once | Dlata | Tat / eggs with paler yolks feather and skin damade at 1_5-week-and / drinking / forading at 3_5. | umedble// |
| | INING | | 0/71 | | | ומוכ | ↓ reaction and shirt damage at + week age / ↓ unitaring / ↓ rotaging at 3 weeks of age while ↑ foraging at 1 week of age / ↓ incidence of foot pad | and van |
| | | | | | | | lesions at 1–3 weeks of age / \downarrow ground pecking/ \uparrow daily BWG / \uparrow final BW / \downarrow | Niekerk |
| | | | | | | | FCR / \uparrow feed intake / no effect on litter quality or feather pecking / | 2019) |
| BSFL | Muscovy duck | 3–55 | 5% | DFI as fed | Once | Plate | No influence on growth performance / improvement of duck's welfare () excreta | (Gariglio et al. |
| YML | | | | basis | | | corticosterone metabolite) | 2023) |
| BSLF | Slow-growing | 28–81 | 10 % | DFI (DM) | Once | Plate | LBW might be improved / <pre> upstand GGT</pre> thus improved liver health / improved | (Bongiorno |
| | chicken | | | | | | FCR and average daily gain / improved immune system function, \uparrow leucocytes and monocytes. \uparrow relative weights of spleen and Burga of Fabricius / no | et al. 2022) |
| | | | | | | | negative effect on growth performance and blood traits | |
| BSLF | Local chicken | 469–546 | 10 % | DFI (DM) | 6 h/d | Dispenser | \downarrow feed intake / improved FCR / \downarrow feather damage / no effect on egg-shell | (Star et al. |
| | | | (12,5 g/ hen) | | | | strength, elasticity or Haugh unit / no effect on laying rate or egg-weight | 2020) |
| HFL | Local chicken | 14 d until sexual | 30–50 g/ bird/d | I | Once | I | improved DWG / improved clutch size, egg weight, number of eggs hatched and | (Dankwa et al. |
| | | maturity | | | | | chick weight | 2002) |
| BSFL = black | soldier fly larva; | YML = vellow mealwork | m larvae; HFL = hous | efly larvae; DFI | = daily fe | ed intake; DN | l = dry matter; FCR = feed conversion ratio; BW = body weight; BWG = body weight | gain. |

presents a new perspective on the potential utilisation of dehydrated larvae in poultry feeding practices. Indeed, in all other papers in which chicks were offered larvae in the live state, the birds were generally reported to consume the larvae without hesitation. An extreme case is the study by Despins and Axtell (1995), in which only LM larvae, and no other feedstuff, were offered to the chicks. The number of live larvae ingested by the chicks was so huge that they were visible through the skin of their distended crops. Unfortunately, the excessive ingestion of larvae also resulted in substantial distress to the chicks.

The modality of larvae distribution is another important aspect to consider, especially when the objective is to stimulate bird activity and foraging behaviours. For instance, whilst providing live YML or BSFL to birds just once a day stimulated the birds' interest in this food, rendering them temporarily more active, it did not result in any long-lasting foraging behaviour (Veldkamp and van Niekerk 2019; Biasato et al. 2022). So, instead of offering a transient stimulus of live larvae, it is of much greater benefit to supply the birds with larvae at multiple times throughout the day (Pichova et al. 2016). Indeed, the fact that a oneoff presentation of live larvae did not stimulate any substantial change in bird behaviour is not surprising, as described by Mench (2009). Thus, in addition to presenting an appropriate foraging substrate, the characteristics of its supply need to be such as to encourage the birds to peck and scratch at the ground in a more consistent manner, producing a grazing behaviour, and not the simple guick consumption of a daily ration. Live BSFL and YML are greatly appreciated by the birds, as demonstrated by the short time needed to empty the plates of larvae presented (Bellezza Oddon et al. 2021). This also implies the existence of some sort of competition between birds, otherwise some larvae would have been left over. By contrast, scattering the larvae in an appropriate loose litter provides the birds with the opportunity to search for them, stimulating the birds to accomplish all three components of foraging behaviour, namely ground pecking, ground scratching and grazing (Mench 2009; Pichova et al. 2016; Ipema, Gerrits, et al. 2020; Ipema, Bokkers, et al. 2020; Ipema et al. 2022). For example, dividing the daily supplementation with BSFL scattered into the litter into seven distinct applications increased bird activity and foraging behaviour throughout the day and over the entire production period (Ipema, Bokkers, et al. 2020). Certainly, from the bird's point of view, this modality of supplementation has the potential to substantially

improve bird welfare; on the other hand, the logistics of this form of supplementation might be very demanding upon the farmer (unless automated), whereas offering the larvae even only once a day, although not offering a prolonged stimulus, may still constitute a positive stimulus to improve the bird welfare. In this context, poultry equipment manufacturers should broaden their market reach by developing specialised automated dispensers designed for both live and dehydrated insect larvae.

The quantities of live larvae offered to birds tested in the various studies range from 5 - 20% of the expected daily feed intake (Ipema, Bokkers, et al. 2020; Ipema, Gerrits, et al. 2020; Star et al. 2020; Bellezza Oddon et al. 2021; Colombino et al. 2021; Tahamtani et al. 2021; Biasato et al. 2022; Ipema et al. 2022; Bongiorno et al. 2022). The supplementation of feed with BSFL to laying hens at 20%, on an as fed basis, resulted in a decrease in feed consumption by 25%, and this quantity of BSFL supplementation did not influence egg production and quality (Tahamtani et al. 2021). Other studies used a dietary supplementation percentage of 5% YML or BSFL (Ipema, Bokkers, et al. 2020; Ipema, Gerrits, et al. 2020; Bellezza Oddon et al. 2021; Colombino et al. 2021; Biasato et al. 2022), 8% BSFL (Ipema et al. 2022), or 10% BSFL (Ipema, Gerrits, et al. 2020; Ipema, Bokkers, et al. 2020; Star et al. 2020; Tahamtani et al. 2021; Bongiorno et al. 2022). None of these dietary inclusion rates of live BSFL or YML had a negative effect on the birds' health or performance. Some of the positive effects observed included a slight improvement in the caecal microbiota, a lower plasma GGT concentration, reflecting a positive effect on liver health (Colombino et al. 2021), and higher monocyte and leucocyte concentrations, reflecting a positive effect on the immune system (Bongiorno et al. 2022). Furthermore, improvements were observed in the feed conversion ratio (Veldkamp and van Niekerk 2019; Star et al. 2020; Bellezza Oddon et al. 2021; Ipema et al. 2022; Bongiorno et al. 2022) and the final live body weight (Veldkamp and van Niekerk 2019; Ipema et al. 2022; Bongiorno et al. 2022). In Muscovy ducks, a 5% supplementation of live BSFL or YML showed an enhancement in the ducks' welfare, indicated by a decrease in excreta corticosterone levels and H/L ratio. (Gariglio et al. 2023). When the live BSFL were offered ad libitum plus their daily feed ration, the hens consumed only 52% of the daily feed intake (DM), and 69% of their protein intake was instead obtained from the larvae (Tahamtani et al. 2021). No effect was observed in the egg production or in the assessed egg parameters, but an increase in abdominal fat was observed. Thus, the high larvae consumption rate was also associated with a high overall level of fat consumption due to the relatively high fat content of the larvae. However, the protein consumed by these hens exceeded their actual dietary need for this nutrient (Tahamtani et al. 2021). Chicks receiving LM larvae ad libitum, scattered in their litter, in addition to their standard feed ration, evidenced a decrease in the feed consumption, their protein intake of 10% to 35% came from the larvae and no change in their body weight gain was observed (Despins and Axtell 1995). Although the empirical data on live larvae inclusion rates in the diet are still limited, 10% larvae in the diet is generally considered to be safe for poultry species. Higher amounts require further evaluation to establish whether they are advantageous for bird health and compatible with the economics of the poultry industry.

Excessive increases in broiler body weight are frequently associated with health problems in these birds - in their legs, in particular, due to the increased load placed upon them. By consequence, birds tend to become less active, walking and moving less in general and resting for greater lengths of times. Thus, scattering the live BSFL through the litter also helps to increase activity in these birds, and one would expect this to be associated with fewer leg problems (Ipema, Bokkers, et al. 2020; Ipema, Gerrits, et al. 2020; Ipema et al. 2022). Indeed, when feeds were supplemented with live BSFL, at 5% (Ipema, Gerrits, et al. 2020), 8% (Ipema et al. 2022), or 10% (Ipema, Gerrits, et al. 2020) of the daily feed intake, distributed by scattering in the litter four times a day, the incidence of leg problems was observed to decrease (Ipema, Gerrits, et al. 2020; Ipema, Bokkers, et al. 2020). Thus, the evidence appears to support the adoption of this practice as a means to improve leg health in broilers, though further studies are still needed to sustain this statement.

Feather pecking is another problem that poultry breeders and the industry in general must deal with. The causes of this stereotypic behaviour may vary. One regards the ease of access to feed. When feed is highly accessible, and birds are not required to search for their food, they have more time to get bored. Feather pecking presents one way in which the birds might fill this time. By contrast, if the bird is given the chance to forage, the likelihood that such a behaviour may occur is reduced (Savory 1978). Thus, scattering larvae in the litter could help avoid this stereotypic behaviour. To date, only the effect of offering batches of dietary live YML or BSFL, as attractive food items at which the birds can peck at, on feather pecking has been evaluated (Veldkamp and van Niekerk 2019; Biasato et al. 2022). In laying hens, a decrease in the feather damage was observed when BSFL were delivered through a special dispenser designed to spread their supply, at an adequate amount per hen, over a longer time (Star et al. 2020). In 4-5-week-old turkeys, larvae supply was associated with a reduction in aggressive back and tail pecking; however, it was not clear whether this reduction depended on the larvae supply itself, since this behaviour and the time of feeding did not coincide (Veldkamp and van Niekerk 2019). In broilers, no positive effect of larvae provision on feather pecking was observed (Biasato et al. 2022), consolidating the need for further studies in order to obtain a clearer picture of the influence of dietary insects on feather pecking behaviour (Table 3).

Very few studies have addressed the use of whole dehydrated larvae in poultry diets (Hwangbo et al. 2009; Traore et al. 2020a, 2020b; Ipema et al. 2022). Once again, scattering the larvae—specifically whole dehydrated BSFL-in the litter promoted bird activity more than when the larvae were provided in the regular feeder (Ipema et al. 2022). However, when dehydrated HF larvae were offered to guinea fowl in a 'cafeteria' test of food preference, they were not considered by the birds, which instead selected cereal grains as their preferred feed (Traore et al. 2020a). This preference for grains was not manifested in a local chicken breed (Traore et al. 2020b), showing that bird species must also be considered when considering the inclusion of whole dehydrated larvae in poultry diets. Regarding the quantity of dehydrated larvae with which to supplement the diet, more information is needed to identify the appropriate level. An inclusion rate of 8% of the total dietary dry matter was found to be beneficial for bird performance (Ipema et al. 2022), whereas up to 20% (on an as fed basis) was shown to improved live body weight and the feed conversion ratio in broilers (Hwangbo et al. 2009). The advantages that stand to be gained from the use of dehydrated larvae should also be considered. For instance, dehydrated larvae are easy to handle and no expensive and/or complex protocols need to be followed prior to offering the larvae to the birds, as are required in the case of live larvae (Table 4).

Other studies investigated dietary supplementation with defrosted BSFL in broilers and local chicken breeds; the quantities offered ranged from 2% to 30% (on an as fed basis) (Moula, Scippo, et al. 2018; Moula, Hornick, et al. 2018; Seyedalmoosavi et al. 2022). The birds expressed greater interest in feed supplemented with defrosted BSFL (Seyedalmoosavi et al. 2022), and

| nsect sp. Bird SSFL Broiler HF Broiler | Age (d) 0–35 1–35 | Quantity 8 % | | | | | |
|--|-------------------------|---------------------------------|-----------------|--------------------------|---------------------|---|--------------------------|
| SSFL Broiler HF Broiler | 0–35 1–35 | 8 % | | Freq. | Method | Effects | Reference |
| HF Broiler | 1–35 | | Tot. dietary DM | 4 times | Feeder Scattered | no f activity promoted / no {hock burns f activity / improved final BW, DM conversion ratio / initially, some birds consume no larvae | (lpema et al. 2022) |
| | | 5 % 10 % | as fed basis | Once | With feed | improved BW and FCR | (Hwangbo et al. 2009) |
| IF Local chicke | 126–168 | 20 % 8 am 12 noon | | Once cafeteria 30 min | Cafeteria | egg parameters not affected 1 maggot consumption / egg parameters not affected | (Traore et al. 2020a) |
| IF Guinea fowl | 308–357 | s pm 8 am 12 noon 4 pm | | Once cafeteria 30 min | Cafeteria | egg parameters not anected Preference for cereal grains over dehydrated maggots | (Traore et al. 2020b) |

| Table 4. | Defrosted b | olack soldie | r fly larva | e inclusion | in poultry | v diets: birc | l species, | bird age | , inclusion | rate, p | orovision | frequency, |
|----------|-------------|--------------|-------------|-------------|------------|---------------|------------|------------|-------------|---------|-----------|------------|
| and the | method thro | ugh which | the larvae | e were pres | ented are | all indicate | d, togeth | er with th | ne related | effects | on the b | ird. |

| Bird | Age (d) | Quantity | | Freq. | Method | Effects | Reference |
|---------------|---------|----------|--------------|---------------|----------------|---|----------------------------------|
| Broilers | 0–42 | 10% | DFI (as fed) | Once morning | Plate | Stimulate bird interest in feed / no adverse effects on growth performance or FCR | (Seyedalmoosavi et al. 2022) |
| | | 20% | | | | Stimulate bird interest in feed / no adverse effects on growth performance or FCR | |
| | | 30% | | | | Stimulate bird interest in feed / ↓ total DM intake, ↓ protein utilisation efficiency, some effects on liver functioning | |
| Broilers | 40–53 | 8% | wet weight | Ad libitum | With feed mash | No influence on BW, BWG, FCR / no influence in the prevalence of most bacterial families in the caeca of birds | (Moula, Hornick, et al. 2018) |
| Local chicken | 30–80 | 2% | DFI (DM) | Not specified | Not specified | Improved BWG gain / no influence on FCR, internal organ weights and meat protein content | (Moula, Scippo, et al. 2018) |

DFI = daily feed intake; DM = dry matter; FCR = feed conversion ratio; BWG = body weight gain.

while no effects on the feed conversion ratio were observed with inclusion rates up to 20% (Moula, Scippo, et al. 2018; Moula, Hornick, et al. 2018; Seyedalmoosavi et al. 2022), 30% inclusion led to a decrease in protein utilisation efficiency and might likely affect liver function (Seyedalmoosavi et al. 2022). Thus, the data available to date appear to be favourable of including defrosted BSFL in poultry diets at rates up to 20%, but further research is needed to sustain these findings. However, the practical challenges associated with maintaining the cold-chain preservation during larvae storage, as well as the administration of a defrosted product that could be damp and gelatinous, do not appear to support the promotion of this practice.

Future perspectives

The use of live, dehydrated or defrosted insect larvae as a dietary supplement in poultry farming is still in its infancy. So, to help this practice become applied more widely, the supply chain of insects to the poultry industry needs to be guaranteed. Insect production companies need to be able to offer affordable and gualitatively standardised live, dehydrated or defrosted insects (Murta 2021). Optimised processing conditions for drying or freezing whole larvae of the different insect species need to be identified to guarantee high quality and safe products (Kröncke et al. 2018; Melgar-Lalanne et al. 2019; Wessels et al. 2020). Thus, further studies are needed to evaluate the effects of such processing on the nutritional value and safety of larvae. The systems for transporting the different processed larvae also need to be standardised to guarantee the integrity and/or freshness of the product. Finally, the methods of distributing the larvae to the birds need to be customised to assure larvae availability throughout the day, and by consequence the various benefits this brings to the birds. Thus, the transfer of new technologies from research entities and companies to the poultry industry will be essential to achieve these goals (Murta 2021).

Conclusions

A common objective of the studies performed to date was to improve bird welfare by inducing foraging behaviour and stimulating greater activity through the dietary use of live, dehydrated or defrosted whole larvae. Larvae scattered in the litter and at multiple times throughout the day was more effective in stimulating these behaviours than providing larvae in batches once daily. For this reason, the larvae distribution system is key for guaranteeing the prolonged availability of larvae to the birds; moreover, to encourage the take-up and continuation of the practice, the system must be easy for farmers and poultry companies to implement and maintain.

Ten percent of the daily feed intake can be safely substituted with live larvae in poultry diets, inducing positive effects on bird activity, foraging behaviour, performance and health status. Further studies are needed to establish the inclusion rates and modalities of distribution that are most beneficial for bird health whilst being economically feasible and sustainable for the breeder.

The advantages of the live larvae reside in their attractiveness to birds, eliciting innate behaviours

which may improve the birds' welfare. Furthermore, live insects can provide the bird with fresh, unaltered nutrients. Maintaining and distributing live larvae can be a labour-intensive and costly process, particularly in production systems that involve very large flocks. Dehydrated larvae, on the other hand, are more manageable and do not necessitate complex protocols before being provided to the birds. The adoption of dehydrated larvae may offer several advantages, including more efficient storage and transportation, as well as enhanced biosecurity under farm conditions. However, the utilisation of dehydrated larvae is still in its early stages and requires further research to gain a better understanding of administration methods and acceptance among various poultry species.

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Ethical approval

Animal Care and Use Committee approval was not obtained for this study because the data were obtained from published scientific articles.

Disclosure statement

No potential conflicts of interest are reported by the authors.

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Data availability statement

The data presented in this article are freely available being obtained from cited published articles.

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