

The potential of black soldier fly (*Hermetia illucens* L.) larvae in chicken and swine nutrition. A review

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ABSTRACT. Raising insects for animal feed and human nutrition offers a valuable solution to many global problems. Recent regulatory developments have made the European food and feed market more receptive to insects. Concerns about prions causing transmissible spongiform encephalopathy previously limited the use of insects in livestock feed. However, the European Food Safety Authority found that the risk of bovine spongiform encephalopathy transmission from processed animal protein was lower than estimated. Consequently, the feed ban was repealed. Chickens and pigs, significant livestock species, require high protein and essential amino acids for growth. Alternative protein sources, such as insects, can improve production performance and yield economic benefits. Among insect species, black soldier fly (Hermetia illucens L.) is promising and well-tested option. Recent research suggests that black soldier fly larvae (BSFL) can partially replace traditional feed protein sources. This review highlights the potential of BSFL for chicken and pig feed, including improvements in growth, meat quality, metabolic and immune function, and overall animal welfare.

Introduction

In 2022, the United Nations published 27th edition of official population estimates and projections prepared by the Population Division of the Department of Economic and Social Affairs of the UN Secretariat (World Population Prospects, 2022). According to these projections, although the human population growth rate has fallen below 1% per year in 2020 (for the first time since 1950), global population growth is expected to continue. In 2022, the world population exceeded 8 billion people, an increase of 1 billion since 2010. The world's population could increase to about 8.5 billion in 2030 and 9.7 billion in 2050 (Röös et al., 2017; Fróna et al., 2019; Fraser, 2020; Sadigov, 2022). This creates the

need to feed millions people. The Food and Agriculture Organization (FAO, 2009) of the United Nations estimates that we currently produce enough food for 12 billion people, yet large quantities are wasted. Improper use of agricultural land potential, including too much land devoted to animal husbandry, is also a severe problem (Tolleson and Meiman, 2015; Salliou, 2023). Currently, pastures used for grazing and land used to grow crops for animal feed account for 77% of agricultural land, thus comprising nearly 30% of the total land surface of the planet (Rauw et al., 2023; Van Huis and Gasco, 2023). In addition, the International Feed Industry Federation (IFIF) predicts that animal protein consumption could double by 2050, which poses a challenge to the production of protein feed ingredients (Lu et al., 2022).

Therefore, alternative feed protein sources in the form of total or partial substitutes that would meet the specific nutritional needs of different livestock species are looked for (Nitharwal et al., 2022; Pexas et al., 2023; Thornton et al., 2023; Cherdthong, 2024). With this in mind, attention began to turn to edible insects as a potential feed ingredient (Wang and Shelomi, 2017; Nowakowski et al., 2021). In 2013, it was published a comprehensive report on the feasibility of using insects as a source of nutrients for humans and animals, indicating the validity of this direction in food and feed production (Van Huis et al., 2013). In subsequent years, regulatory changes were brought about, allowing the addition of animal protein (including insect protein) to animal feed. To date in the European Union (EU), seven insect species have already been approved to be used as feed ingredients for aquaculture organisms, pets, poultry, and pigs (Liceaga, 2021; Montanari et al., 2021; Lourenço et al., 2022). This is referred to in the Commission Regulation (EU) 2021/1372. This regulation amends Annex IV of Regulation (EC) No. 999/2001 and Annexes X, XIV, and XV of Regulation (EU) No. 142/2011. Insect species whose production has been authorized in the EU for this purpose are: black soldier fly (Hermetia Stratiomyidae), illucens) (Diptera: common housefly (Musca domestica) (Diptera: Muscidae), yellow mealworm (Tenebrio molitor) (Coleoptera: Tenebrionidae), lesser mealworm (Alphitobius diaperinus) (Coleoptera: Tenebrionidae), house cricket (Acheta domesticus) (Orthoptera: Gryllidae), banded cricket (Gryllodes sigillatus) (Orthoptera: Gryllidae) and field cricket (Gryllus assimilis) (Orthoptera: Gryllidae) (Lourenço et al., 2022). Moreover, numerous studies have investigated the contribution of insects as a feed ingredient in animal diets, focusing primarily on growth performance, microbiological and health consequences, and nutrient composition and utilization (Biasato et al., 2017; Nephale et al., 2024). Several insects have been tested as animal feed, with the most promising species being the black soldier fly (BSF), the yellow mealworm and the housefly. The BSF and yellow mealworm are also the most widely reared insect species today, and they are increasingly used as animal feed (Allegratti et al., 2018; Schiavone et al., 2018; Secci et al., 2018; Sogari et al., 2019; 2023; Astuti and Wiryawan, 2022; Van Huis and Gasco, 2023). Numerous studies have highlighted the possibility of incorporating BSF larvae (BSFL) primarily into chickens and weaning piglets diets as a partial or complete replacement for conventional

protein/fat sources. Moreover, using BSFL in livestock feeding yields positive results in animal health and performance, gut health aspects and product quality (Gasco et al., 2018; Sogari et al., 2019; Dörper et al., 2020; Flis et al., 2024).

BSF is a saprophytic insect that has attracted interest from the scientific community and industry in recent years. This concerns the BSFL ability to feed on a diverse substrate in the form of organic waste, rapid growth, and negligible environmental impact of breeding (Kaczor et al., 2023). BSFL can reduce organic matter by 42-56% by consuming it and accumulating it as protein (more than 40%) (Gobbi et al., 2013; Park et al., 2015; Salomone et al., 2017; Siddiqui et al., 2022). The potential of BSFL as an alternative feed ingredient is related to the possibility of controlling the process of their life cycle and, thus, their mass breeding (Sogari et al., 2019). The mass production of BSFL for feed purposes requires establishing routine quality control procedures since insect development is a dynamic process dependent on many biotic and abiotic factors. Such procedures should include, first and foremost, continuous monitoring of the health and productivity of adult insects in laying eggs and evaluation of the basic parameters of larval development and the physicochemical properties of the final products (Cickova et al., 2015; Lievens et al., 2021).

This review examines the potential of BSFL as feed for chickens and pigs, which are among the most important livestock species in global meat production. Both chickens and pigs have high demands for protein and essential amino acids during their growth periods. Therefore, enhancing the nutritional quality of their feed with alternative protein sources, such as BSFL, can effectively improve production performance and lead to increased economic benefits (Jin et al., 2016; Ao and Kim, 2019; Sogari et al., 2019; Ao et al., 2020; Hancz et al., 2024).

Legal framework for using insects as livestock feed (including BSF)

Raising insects to feed animals and nourish humans is attractive because it represents a potentially valuable solution to many world's problems today. Moreover, several important changes have happened at the regulatory level over the past few years, making the European food and feed market increasingly open to insects (Mina et al., 2023). Based on the European Food Safety Authority (EFSA, 2015) opinion dated October 8, 2015, certain insect

species were considered to meet safety conditions and carry no adverse effects on plant, animal or human health. This created the possibility of using insects as food and livestock feed. For many years, the use of insects in livestock feed has been considered risky due to suspicions about the presence of prions responsible for transmissible spongiform encephalopathies (TSEs), limiting their use only to mixtures intended for a narrow group of fur-bearing animals. The conclusions of a 2015 EFSA's opinion state that the prion risks associated with using unprocessed insects will be at the same or even lower levels as for other animal products as long as the insects are fed substrates that do not contain ruminant or humanderived material. In 2017, a list of processed animal protein derived from farmed insects, including BSF, was provided for the production of feed for livestock other than fur animals. At the regulatory level, the possibility of feeding processed insect protein to animals is a break from the EU's ban on the use of animal protein to feed livestock (the so-called feed ban), introduced in 2002 to prevent infectious bovine spongiform encephalopathy (BSE) (Regulation (EC) No 999/2001 of the European Parliament and of the Council of 22 May 2001 laying down rules for the prevention, control and eradication of certain BSEs). Regulation (EU) 2017/893, which amends Annexes I and IV to Regulation (EC) No. 999/2001 and Annexes X, XIV, and XV to Commission Regulation (EU) No. 142/2011, regards the provisions on processed animal protein. The regulation also specifies the substrates allowed as feed for insects. Furthermore, in the same year, Regulation (EU) 2017/1017 authorized live terrestrial invertebrates and dead terrestrial invertebrates with or without treatment as feed materials but not processed as described in Regulation (EC) No. 1069/2009 (Sogari et al., 2019; 2023; Lähteenmäki-Uutela et al., 2021; Renna et al., 2023).

According to a EFSA Scientific Opinion, it was assumed that the risk of transmission of the BSE prion agent posed by using processed animal protein in animal nutrition is four times lower than estimated in 2011 (Ricci et al., 2018). Therefore, in 2021, long-awaited changes were made to repeal the feed ban fully. As a result, the use of processed animal proteins in the feeding of livestock is now regulated by a new piece of legislation, namely, Commission Regulation (EU) 2021/1372 of 17 August 2021, amending Annex IV to Regulation (EC) No. 2001/999 of the European Parliament and the Council. Significantly, in the context of the topic at hand, the regulation expands the possibility of using

processed proteins from farmed insects in feeding pigs and poultry (Woodgate and Wilkinson, 2021; Renna et al., 2023).

Chemical composition of BSF

The chemical composition of insects varies from species to species; however, even within a single species, there are significant differences in this regard, depending on the developmental stage of the insect, the type of habitat, and the diet. Therefore, the nutritional value of BSF can be manipulated by interfering with the composition of waste streams for bioconversion/rearing substrate and by making sure that specific components are available in the stream (Van Huis et al., 2013; Sogari et al., 2023). In addition, to use BSFL as a feed ingredient, the chemical safety of the larvae must be guaranteed through the proper composition of the rearing substrate. That composition is a crucial safety factor because it bioaccumulates potentially hazardous compounds (such as toxic metals, pesticides, and mycotoxins) (Lievens et al., 2021).

Protein content and amino acids composition of the BSFL

BSFLs are rich in protein, ranging from about 30 to over 50 g per 100 g of dry matter (DM), regardless of the rearing substrate (Schiavone et al., 2017a; Liu et al., 2017; 2018; Yildirim-Aksoy et al., 2020; Tyshko et al., 2021; Huseynli et al., 2023; Diola et al., 2024). Commonly used substrates include organic waste (e.g., food scraps and fruit and vegetable peelings), animal manure (such as cow, chicken, and pig manure), crop residues, and food processing by-products. The studies also showed that differences in amino acid composition were minor regardless of the substrate rearing. An extensive study on the amino acid profile of BSFL depending on the type of the substrate (chicken feed, digest, vegetable waste, and restaurant waste) was conducted by Spranghers et al. (2017). In this study, the protein content was high regardless of the substrate (ca. 40-43 g/100 g DM). Lysine was determined at 2.3-2.6 g/100 g DM; all BSFL contained 1.5-1.7 g/100 g DM of threonine. The contents of methionine, isoleucine, and valine were 0.7-0.9 g/100 g DM, 1.7-1.9 g/100 g DM, 2.4–2.8 g/100 g DM respectively. Studies by other authors confirm the relatively low methionine content in BSFL (about 0.5-1 g/100 g), the higher isoleucine content (about 1.8-5.2 g/100 g), and the high levels of valine (about 2.8–6.8 g/100 g). In addition to valine, the most abundant essential

amino acids in BSFL are leucine (ranging from about 2.8-7.8 g/100 g) and lysine (ranging from about 2.3-8 g/100 g) (Schiavone et al., 2017a; Shumo et al., 2019; Rawski et al., 2020; Yildirim-Aksoy et al., 2020; Lu et al., 2022; Zulkifili et al., 2022; Miron et al., 2023). A comprehensive summary of research on the protein and amino acid content of BSFL is provided in a review paper by Lu et al. (2022). Against this background, the study by Shumo et al. (2019) is particularly noteworthy. The authors of this paper obtained noteworthy results, demonstrating a superior amino acid profile in BSFL compared to those found in previously cited studies. The levels of methionine (6.1–7.9%) significantly exceeded those found in BSFL grown on different organic waste streams. The levels of lysine (4.1–4.7%), isoleucine (1.6–1.8%), and leucine (3.0–3.7%) were within the accepted ranges. In this experiment, BSFL were reared on three different organic substrates: poultry manure, spent brewer's grain, and kitchen waste. The larvae were collected at the prepupal stage and, unlike the method used by Spranghers et al. (2017), were oven-dried at 60 °C for 48 h instead of being freeze-dried. In this study, the crude protein (CP) concentration in the dry weight of the feed was expressed using a nitrogenprotein conversion factor of 4.76, and it ranged from 33 to 41%. The authors note the importance of using different nitrogen-protein conversion factors. In insects, nitrogen can originate from both protein and non-protein sources, such as chitin (Jonas-Levi and Martinez, 2017; Kipkoech, 2023). Therefore, separating non-protein nitrogen from protein nitrogen was necessary to obtain an accurate crude protein content and avoid overestimates occurring with the standard coefficient of 6.25, which is usually applied (Jonas-Levi and Martinez, 2017). This issue is described in detail by Jonas-Levi and Martinez (2017). Additionally, Shumo et al. (2019) showed no significant effect of substrate on most of the essential amino acids important for animal nutrition, including methionine, lysine, isoleucine, and leucine. Considering other studies on the protein content of BSFL (regardless of the type of culture medium), it can be assumed that the average crude protein content is similar to that reported in this study. Significantly, the values shown for protein can be affected by the content of fat and fatty acids (there are currently two main types of BSFL in animal feed: skimmed and full-fat). In the case of full-fat BSFL, the protein content differences were insignificant, while after skimming, they varied over a wide range of ca. 22-65 g/100 g (Onsongo et al., 2018; Schiavone

et al., 2017a; Yildirim-Aksoy et al., 2020; Lu et al., 2022). Although defatted larvae are inherently a more concentrated source of protein, the observed differences may be related to the specific defatting methods used. Given the gaps in knowledge about the chemical composition of BSF at different stages of development, as well as the influence of various culture media and processing methods and their complex interrelationships, further in-depth research in this area is needed. Nevertheless, available literature suggests that BSFL can serve as a valuable source of protein and amino acids, the deficiency of which most often reduces feed's nutritional value.

Fat content and fatty acid composition of the BSFL

BSFL contain a large amount of fat compared to other insects. Ramos-Bueno et al. (2016) analyzed the fatty acid profiles of various insect species from various orders, specifically Diptera (Hermetia illucens and Lucilia sericata) at the larval stage, Orthoptera (Locusta migratoria, Acheta domestica, and Anacridium aegyptium) at the nymph stage, and Coleoptera (Tenebrio molitor and Zophobas morio) at the larval stage. This study found the highest percentage of fat (about 50% DM) in Z. morio and H. illucens species. Most literature indicates that the lipid content of BSFL ranges from about 20 to more than 40 g/100 g of DM (Spranghers et al., 2017; Shumo et al., 2019; Georgescu et al., 2022; Li et al., 2022; Saadoun et al., 2020; Huseynli et al., 2023). This value depends on the diet, farming conditions, and metamorphic stage. Modulation of these factors allows for the adjustment of the quantity and quality of lipids to achieve the desired nutritional composition of BSFL used as an ingredient in animal feed. Studies by various authors indicate that the lipid profile of BSFL, including the composition of fatty acids, depends on the content and composition of the substrates used for rearing the larvae. The study conducted by Saadoun et al. (2020), aimed at evaluating the content and profile of lipids extracted from BSF prepupae cultured on various media (including industrial by-products and by-products from retailers), demonstrated that the lipid profile of BSFL, including the proportion of saturated fatty acids (SFAs) and unsaturated fatty acids (UFAs), varied depending on the composition of the substrates. Of particular note is the extensive study by Ewald et al. (2020), which analyzed the proximate and fatty acid composition, larval growth, and feed conversion across 11 different diets, including organic waste such as mussels, bread, fish, and food waste. The authors suggest that the possibilities for modi-

fying the fatty acid composition are limited. The calculated fatty acid production value (FAPV) does not support the conclusion that BSFL are capable of synthesizing polyunsaturated fatty acids (PUFAs). Rather, linoleic acid (C18:2), alpha-linolenic acid (ALA, C18:3), eicosapentaenoic acid (EPA, C20:5), and docosahexaenoic acid (DHA, C22:6) found in BSFL are most likely derived from the substrate. The increase in the percentage of EPA and DHA in BSFL was related to the higher fatty acid content in the diet. This is supported by a study by Rodriguez et al. (2022), which found that significant levels of EPA and DHA were recorded in BSFL reared on a high UFAs substrate (fish discards and coffee silverskin with Schizochytrium sp. algae). Similar studies that included algae such as the brown alga Ascophyllum nodosum (Liland et al., 2017) and microalgae Isochrysis sp. and Schizochytrium sp. (Truzzi et al., 2020) in the substrate for BSFL also showed an increase in EPA and DHA concentrations. Many other studies demonstrate that fisheries and aquaculture by-products as well as marine-based substrates significantly improve the lipid profile of BSFL, particularly the n-3 PUFAs profile (Eggink et al., 2022; Arena et al., 2023; Tirtawijaya et al., 2024). Another study by Georgescu et al. (2022) found that dietary enrichment with vegetable oils (linseed oil, hempseed oil, and rapeseed oil) improved the fatty acid profile for UFAs, especially in long-chain PUFAs from the n-3 family. Additionally, as the larvae develop, there can be a significant decrease in UFAs with a corresponding increase in SFAs, as shown in a study by Liu et al. (2017) that evaluated metabolic changes in BSF nutrient composition from egg to adult. BSFL can convert UFAs into SFAs, which are more stable and may be preferred as a form of energy storage. Therefore, the timing of larvae harvesting is crucial.

BSFL in broiler chickens and laying hens nutrition

The use of BSFL in chickens nutrition provides an opportunity to improve production efficiency, health, and animal welfare. BSFL have great potential as a chickens feed ingredient due to their high protein and fat content, which can enhance meat and egg quality by improving the lipid profile and increasing protein content. This makes them a potentially valuable substitute for traditional protein sources such as fish meal and soybeans (Zamri et al., 2023). BSFL is an attractive energy source because of its higher content of apparent

metabolizable energy (AME). The AME of BSFL was determined by Mahmoud et al. (2023) to be 19.1 MJ/kg DM, while the AME corrected for zero nitrogen retention (AME_N) was found to be 18.0 MJ/ kg DM. The AME value is much higher than that of SBM ranging 8.4-9.9 MJ/kg (Ravindran et al., 2014). Partially defatted BSFL is characterized by 16.25 and 14.87 MJ/kg DM of AME and AME_N, respectively, while for highly defatted meal these values for broiler chickens are 11.55 and 9.87 MJ/ kg DM (Schiavone et al., 2017b). Defatted BSFL meal offers a higher crude protein (CP) content and may be a better feed ingredient for chickens than soybean meal (SBM). Previous studies have shown that CP content ranges between 36-49%, with most literature indicating 40-45% (Makkar et al., 2014; Spranghers et al., 2017; Schiavone et al., 2017a,b; Dabbou et al., 2018). However, the CP content is significantly related to fat content. The skimming process increases the protein concentration in the final product, making the meal more abundant and potentially more valuable as a high-protein feed ingredient. A study evaluating the CP content of partially defatted and highly defatted BSFL meal found the content of this component to be approximately 55% and 65%, respectively (Schiavone et al., 2017b). In addition to CP content, indicators of poultry feed protein quality include amino acid profiles and ileal amino acid digestibility (Abd El-Hack et al., 2020). Chicken diets have traditionally relied heavily on plant-based protein sources. The main amino acids limiting the nutritional value of plant protein are lysine, methionine, and threonine. These amino acids are crucial for the desired growth of chickens and the maintenance of normal metabolic functions (Zulkifili et al., 2022). BSFL exhibits high levels of essential amino acids and an overall more favourable amino acid profile compared to SBM and most conventional plant protein sources (Tran et al., 2015; Spranghers et al., 2017; Abd El-Hack et al., 2020; Fatima et al., 2023). Lu et al. (2022) demonstrated a higher amino acid profile of BSFL relative to soybean meal, particularly in terms of leucine, lysine, and valine. Valine content was also higher compared to fish meal, while methionine and tryptophan were at similar levels to SBM but lower than in fish meal. BSFL also exhibited higher levels of essential amino acids such as alanine, proline, and tyrosine compared to soybean and fish meal (Tschirner and Simon, 2015; Lu et al., 2022). The levels of amino acids will vary depending on the conditions of

larval rearing, substrate, and developmental stage. However, available literature suggests that BSFL meal can be primarily used as a replacement for soy products in chicken diets (Abd El-Hack et al., 2020; Zulkifili et al., 2022; Lu et al., 2022). Research has demonstrated that supplementing feed with BSFL meal can significantly enhance weight gain, nutrient assimilation, and overall health in both laying hens and broiler chickens. It is essential to consider the distinct nutritional requirements and production objectives of these two poultry groups when determining the optimal proportion of BSFL meal in their feed. According to available literature, recommended levels of BSFL inclusion in broiler diets, encompassing both BSFL meal and whole larvae, typically range from 5 to 25%, with an optimal range falling between 10 and 20% (Bovera et al., 2016; De Marco et al., 2015; Cullere et al., 2016; Schiavone et al., 2017a,b; Hartinger et al., 2021; Seyedalmoosavi et al., 2022). Hartinger et al. (2021) demonstrated the viability of partially replacing 15% of CP from SBM with BSFL meal in broiler diets without compromising performance or animal health. Various parameters were evaluated, including broiler performance, carcass traits, apparent ileal digestibility, intestinal morphology, and microbial metabolites. However, when replacing 30% of CP with SBM with BSFL meal, growth inhibition was observed, attributed by the authors to probable reduced digestibility in the ileum. Nevertheless, no changes in intestinal morphology or adverse effects of fermentation in the hindgut were noted. In a study by Seyedalmoosavi et al. (2022), which examined the impact of different levels of whole BSFL inclusion in broiler diets on nutrient utilization efficiency and growth performance, it was demonstrated that including up to 20% BSFLs in the diets did not adversely affect the evaluated parameters. However, it was observed that a higher proportion of BSFLs in broiler rations led to reduced protein utilization efficiency, attributed to lower total energy intake. Therefore, incorporating moderate levels of BSFLs can be advantageous in formulating broiler feeds, as it promotes optimal growth performance, feed conversion efficiency, and overall broiler health without detrimental effects.

Literature data suggests the feasibility of incorporating BSFL meal as a replacement for SBM in the diets of laying hens. Numerous studies show the impact of such an additive on egg production, egg quality and bird health. Optimal levels of BSFL content in the diet of laying hens typically range between 5 and 15% (Maurer et al., 2016; Bovera et al., 2018;

Secci et al., 2018; Chu et al., 2020). These feeding strategies are evaluated across the starter, middle, and late laying periods. Chu et al. (2020) investigated the effects of incorporating low levels of fullfat BSFL meal (0, 3, 6, and 9%) in the diets of layer chickens during the starter laying period (from 1 to 42 days of age). Their study assessed parameters such as growth performance, nutrient digestibility, plasma antioxidant ability, and gut health. It revealed that an increase in the content of full-fat BSFL meal resulted in a quadratic increase in final body weight and average daily gain, as well as in the digestibility of CP and ether extract. Furthermore, there was a linear increase in total superoxide dismutase and glutathione peroxidase activities in birds' plasma as well as in the ileal mucosal secretory immunoglobulin A. This indicates an enhanced antioxidant capacity and immune response of the ileal mucosa in laying chickens following the experiment. Consequently, it can be inferred that partially incorporating full-fat BSFL into the diet of chickens may prove beneficial during the starter period. Conversely, Marono et al. (2017) investigated the effects of BSFL meal as a complete substitute for SBM during the later laying period (from 24 to 45 weeks of age). In this study, two different isonitrogenous and isoenergetic diets were administered: hens in the control group were fed a corn-SBM-based diet, while in the other group SBM was entirely replaced with BSFL meal. Feed intake, number of eggs produced, and egg weight were recorded weekly throughout the experiment, and blood samples were collected at the end for further biochemical analyses. The authors observed a more favourable feed conversion ratio in hens fed BSFL; however, the lay percentage, feed intake, average egg weight, and egg mass were lower in this group. Hens fed BSFL produced a higher percentage of eggs in the small, medium, and extra-large classes, while those fed SBM produced a higher percentage of eggs in the large class. Consequently, substituting BSFL meal entirely for SBM may negatively impact feed intake and overall production performance of hens, despite the more favourable feed conversion ratio observed. These findings suggest the necessity of reducing the percentage of BSFL meal in laying hens' diets compared to that used in the study.

Research on the utilization of BSFL in chicken nutrition has primarily concentrated on the nutritional benefits of larvae, particularly their high-quality animal protein content with a favourable amino acid profile and their beneficial fat content with a favourable fatty acid profile. These components play

a crucial role in supporting proper growth, development, and the quality of eggs and meat. Feeding BSFL to chickens can improve the fatty acid profile of the meat, particularly n-3 and n-6 PUFAs (Makokha et al., 2023). It may also increase mediumchain fatty acids (MCFAs), such as lauric and myristic acids, which in turn could help lower cholesterol levels. MCFAs have been shown to increase the expression of genes involved in cholesterol elimination (CYP7A1) (Aprianto et al., 2023). In addition to these nutritional aspects, it's noteworthy to consider the presence of bioactive peptides in BSFL. These peptides have been found to enhance chickens' resistance to infection, potentially offering an opportunity to reduce the reliance on antibiotics in the poultry industry. The antimicrobial properties of BSFL are attributed to their ability to thrive in harsh environments regulated by bacteria and fungi. The development of larvae under such conditions is facilitated by the production of antimicrobial substances, enabling them to thrive in an environment with a diverse microbiome. Antimicrobial activity is demonstrated, among other factors, by small cysteine-rich cationic proteins known as defensins, found in insect foraging cells (Park et al., 2014; 2015; Xia et al., 2021; Zhang et al., 2022). The antimicrobial properties of BSFL are also demonstrated by the lauric acid it contains, which can additionally enhance energy metabolism by being easily converted to energy in the liver (Suryati et al., 2023). MCFAs are an effective energy source, as they are rapidly and efficiently oxidized in the liver (Dabbou et al., 2021; Aprianto et al., 2023). Additionally, the observed improvement in the health of test animals may be attributed to the presence of chitin, a component of the insect body (Pedrazzani et al., 2024). Chitin serves as a dietary fibre with prebiotic properties, exerting a beneficial influence on the development of the digestive system. It stimulates intestinal peristalsis and fosters the growth of normal intestinal microbiota. A healthy intestinal microflora reduces the penetration of pathogens and their toxins across the intestinal barrier, thereby reducing the burden on the immune system. Lowering the production of pro-inflammatory cytokines helps prevent chronic inflammation and promotes a balanced immune response. In the glandular stomach and intestines of chickens, chitin can be broken down into chitosan and chito-oligosaccharides by acid chitinase (Tabata et al., 2017). Chitin that reaches the large intestine can be utilized by microorganisms as a fermentable substrate (Yan et al., 2023). The fermentation process produces short-chain fatty

acids, which have anti-inflammatory properties. As a result, this contributes to improved intestinal health, enhanced nutrient absorption, and bolstered immune system function, as confirmed by the findings of the cited studies.

In efforts to enhance the welfare of hens, studies have explored the practice of feeding them live BSFL, aiming to promote foraging behaviour and mitigate undesirable behaviours such as feather pecking. Indeed, reducing feather pecking poses a significant challenge in improving the welfare of laying hens. Several studies have demonstrated an improved plumage condition in response to feeding live BSFL to hens (Star et al., 2020; Tahamanti et al., 2021). Furthermore, these studies have indicated that the facilitation of natural foraging behaviour does not negatively impact feed utilization, weight gain, or egg parameters (Star et al., 2020; Tahamanti et al., 2021). A study by Tahamtani et al. (2021) assessed the effect of incorporating live BSFL (at varying levels: 0, 10, 20, and *ad libitum*) into the diets of laying hens (aged 18 to 30 weeks), revealing notable differences between hens provided free access to BSFL and those in other experimental groups. Hens provided with BSFL ad libitum consumed less commercial laying concentrate feed. Additionally, birds in this group exhibited greater weight gain compared to hens in all other feeding regimens. This study was the first to offer varying amounts of live BSFL larvae, including an ad libitum portion, to laying hens. A similar study assessing the preference of broilers for whole BSFL over traditional protein feed (SBM) was conducted by Seyedalmoosavi et al., 2022. In this study, the larvae eating time and eating rates of broilers suggested a strong preference for BSFL over regular feed.

Hence, this area of research holds significant potential for application and warrants further exploration. It is imperative to undertake efforts to optimize the utilization of BSFL larvae as a feed additive in hen nutrition and establish an optimal framework and guidelines for breeders. There is undeniable potential for BSFLs as a feed ingredient for both laying hens and broilers, owing to their nutrient content, functional properties, and practical applicability in this type of animal production.

Use of BSFL in pigs' nutrition

BSFLs are a valuable and balanced source of protein and energy in pigs nutrition. The metabolizable energy content of full-fat BSFL for growing pigs was determined to be 19.1 MJ/kg DM, while that of defatted product 14.2 MJ/kg DM.

The net energy content approximated 14.5 and 11 MJ/kg DM, respectively (Crosbie et al., 2020). Scientific studies confirm their beneficial effects on growth, health, and feed acceptance in pigs (Sogari et al., 2019; Mlček et al., 2021; Ipema et al., 2021a,b; 2022; Van Heugten et al., 2022; Phaengphairee et al., 2023). The use of BSFL as a feed additive has been primarily evaluated for its impact on growth performance, meat quality, protein and amino acid digestibility, and gastrointestinal health and function (Tan et al., 2020; Mlček et al., 2021; Zhu et al., 2022). These studies were conducted mainly to assess the potential of BSFL to replace traditional protein sources such as SBM and fish meal in the diets of weaned piglets (Sogari et al., 2019; Mlček et al., 2021; Zhu et al., 2022). Zhu et al. (2022) observed significant improvements in meat quality and growth performance when fish meal was replaced with 4 and 8% BSFL in the diets of growing pigs. In this case, BSFL had an effect on increasing average daily gain and backfat thickness, as well as increasing intramuscular fat content. Intramuscular fat concentration positively affects the tenderness and flavour of pork, which are highly valued aspects of meat quality. Based on these findings, the authors suggest that BSFL can effectively replace fish meal in growing pigs nutrition. The same addition of 4 and 8% full-fat BSFL to the feed of weaned piglets was used by Spranghers et al. (2018). Additionally, the authors of this work used an addition of 5.4% defatted BSFL (compared to a control diet containing soybean as a source of protein and fat). There were no differences in daily gain, feed intake, and feedto-gain ratio between the experimental groups. Apparent total tract digestibility of feed with BSFL supplementation was not significantly different from that of the control feed (protein digestibility in the range of 77–78% in each group). In contrast, ileal digestibility was lower for the diet containing 8% full-fat BSFL and higher for the other feeding regimens (4% full-fat and defatted) compared to the control diet. This study confirms the possibility of partially replacing soy-containing feed with BSFL (in a limited amount) without negatively affecting production performance.

The beneficial properties of BSFL can be further enhanced by the addition of other feed ingredients, inducing a synergistic effect. Although the number of such studies is still limited, this area requires further research and offers an opportunity to develop many new solutions. An interesting study in this regard was conducted by Szczepanik et al. (2022), where weaned piglets were fed diets enriched with full-fat

BSFL (2.5 and 5%) and astaxanthin, a compound with strong antioxidant and anti-inflammatory properties, which can also be a valuable addition to pig diets. Supplementation of BSFL with astaxanthin had no negative effect on feed intake, feed utilization, body weight gain, or organ weights of pigs in all feeding regimens. The addition of 2.5% BSFL and astaxanthin (used in mixed feeds separately or together) can reduce the susceptibility of pork fat to oxidation and improve its shelf life. BSFL exhibits antioxidant activity due to its favourable fatty acid profile, particularly the long-chain PUFAs from the n-3 series. When included in pig diets, BSFL can also lead to pig fat with improved oxidative stability (Lu et al., 2022). Using both agents (separately and in combination) at a lower level of BSFL (2.5%) does not adversely affect biochemical and haematological blood parameters. However, increasing the addition of BSFL to 5% (without astaxanthin) may negatively affect some haematological parameters, possibly due to the presence of lauric acid (Szczepanik et al., 2022).

BSFLs can provide valuable and balanced protein, which enhances growth performance and helps reduce reliance on traditional, less balanced protein sources such as soy or fish meal. An important aspect of using BSFLs is their ability to improve animal health parameters, thereby increasing production efficiency. BSFLs support the health of pigs' digestive tracts through bioactive compounds such as chitin and chitosan. These compounds have prebiotic properties that promote a healthy intestinal microbiota, leading to improved digestion and nutrient absorption. Enhanced intestinal health results in better overall pig condition and a reduced risk of disease. Additionally, chitosan has antibacterial and antiviral properties that help protect the gut from pathogens and lower the risk of infection (Khayrova et al., 2019; Lu et al., 2022). Yu et al. (2019) examined the effects of BSFL on colonic microbiota and bacterial metabolite production in fattening pigs. They found that including 4 and 8% BSFL in the diet increased the abundance of beneficial bacteria such as Lactobacillus, Roseburia, and Clostridium XIVa, while decreasing the abundance of Streptococcus (at 4%). At the 8% inclusion level, an increase in Clostridium XIVa was observed. The addition of 4% BSFL increased the total concentrations of short-chain fatty acids, butyrate, and isobutyrate in the colon, while reducing protein fermentation products such as total amines, cadaverine, tryptamine, phenol, p-cresol, and skatole. At an 8% inclusion level, BSFL further increased butyrate concentration and decreased that of phenol,

p-cresol, and skatole. The 4% BSFL supplementation also decreased the expression of TLR-4 and pro-inflammatory cytokines (interferon-γ) while increasing the expression of anti-inflammatory cytokines (interleukin-10) and intestinal barrier genes encoding (zonula occludens 1, occludin, and mucin 1). The increased expression of colonic mucosal genes was correlated with changes in the composition of the intestinal microbiota and the metabolite profiles of these bacteria, indicating improved immune homeostasis of the intestinal mucosa. Dietary supplementation with full-fat BSFL, both alone and in combination with multi-probiotics, as a substitute for dietary antibiotics was also studied in weaning piglets. This supplementation showed improved growth performance, beneficial effects on intestinal health, and enhanced antioxidant capacity (Phaengphairee et al., 2023). BSFLs improved nutrient digestibility and increased serum levels of immunoglobulin A and glutathione peroxidase, while also reducing levels of pro-inflammatory cytokines. Additionally, pigs fed BSFL had longer duodenal villi, a higher villus heightto-crypt depth ratio, and shorter crypt depth. In pigs fed BSFL supplemented with 0.1% multiprobiotics (Bacillus subtilis, B. licheniformis, Saccharomyces cerevisiae), caecal pH was lower, and the number of Lactobacillus spp. in the faeces increased while Escherichia coli decreased. This indicates that BSFL has great potential to replace antibiotics in weaned piglets (Phaengphairee et al., 2023). The antagonistic activity of BSFL against E. coli was demonstrated in microbiological studies using BSFL extracts by Park et al. (2014). This is particularly relevant to swine farming, as E. coli bacteria play an important role both as part of the normal intestinal microbiota and as a potential pathogen in weaned piglets (Kim et al., 2022). Some strains of *E. coli*, primarily enterotoxigenic E. coli (ETEC), cause diarrhoea in weaned piglets, posing a serious breeding problem (Boeckman et al., 2022). ETEC-induced diarrhoea can lead to dehydration, weakness, and consequently increased mortality. Therefore, BSFLs can be an effective tool to control and prevent infections caused by pathogenic E. coli strains. The antimicrobial properties of BSFLs are largely due to their favourable fatty acid profile, particularly the high content of lauric acid (C12:0) (Zeng et al., 2022; Hong and Kim, 2022; Suryati et al., 2023; Middelkoop et al., 2024). In the animal body, lauric acid is converted to monolaurin, a glyceride that exhibits antiviral, antibacterial, and antiprotozoal properties (Matsue et al., 2019; Nitbani et al., 2022; Abd El Ghany et al., 2024). The increase in Lactobacillus spp. and decrease in E. coli demon-

strated by Phaengphairee et al. (2023) might be related to the premise proposed in the literature, according to which monolaurin, unlike antibiotics, can act antagonistically towards pathogenic microbiota, while probiotic bacteria remain unaffected (Finke, 2013; Ushakova, 2016; Lee et al., 2020; Borrelli et al., 2021). However, not all studies in this regard are conclusive, especially with regard to in vitro studies. Admittedly, the antagonistic activity of BSFL against E. coli was demonstrated by Park et al. (2014) in microbiological studies using BSFL extracts. However, there are also in vitro studies in which no significant antimicrobial activity against E. coli was reported, even with a high amount of C12:0 in BSFL fatty extracts (Spranghers et al., 2018). Therefore, further in-depth research in this area is needed.

BSFL can serve not only a nutritional function but also enrich the pigs' environment and, as described for broilers, contribute to their welfare. The use of whole live BSFL allows pigs not only to eat but also to sniff, manipulate, and destroy the larvae, fulfilling their exploratory needs. Exploratory behaviours are species-specific (Mkwanazi et al., 2019; Ipema et al., 2022). Ipema et al. (2021a, 2021b) showed a high motivation of pigs to consume live BSFL and a clear preference for BSFL over other feeds and enrichment products. The consequences of incorporating BSFL into the diet of pigs have attracted considerable interest, and the potential effectiveness of using live BSFL as environmental enrichment for these animals is also worth noting. Relatively little attention has been paid to this issue to date, and the results of recently published studies are promising (Ipema et al., 2021a,b; 2022).

The above information applies primarily to weaned piglets. This is a critical period for piglets in commercial breeding, where diet significantly impacts health (mainly gastrointestinal health and function, and intestinal microbiota), growth, and nutrient absorption efficiency. Piglets after weaning are particularly sensitive to dietary changes, making them an ideal model for research on new protein sources. As a result, there is less literature available on feeding BSFL to pigs in later stages of growth (growing-finishing pigs, gilts, and sows), and this is an area that requires further research. Research on the use of BSFL in feeding these animals later in life primarily concerns fattening pigs and considers parameters such as weight gain, carcass composition, and overall production performance. An interesting research direction is the potential use of whole live larvae as environmental enrichment and for stress reduction.

Conclusions

In summary, existing research suggests that black soldier fly larvae (BSFL) can serve as a beneficial feed additive in the diets of chicken and pigs. They offer a valuable alternative to traditional vegetable-based protein sources, primarily due to their high essential amino acid content and favourable nutrient profile. Many studies cited in this paper indicate that incorporating BSFLs can effectively replace soybeans and fish meal in feed formulations. This substitution holds practical implications for enhancing chickens and swine growth, improving meat quality, and enhancing metabolic function and immunity in these animals. Despite the considerable potential of BSFLs in livestock nutrition, further research and technological advancements are necessary to optimize their practical application. Priority should be given to developing standardized solutions and procedures for BSFL production, considering variations in larval chemical composition and nutritional value based on developmental stage, habitat type, and diet. Furthermore, manipulating the nutritional value of BSFLs by adjusting the composition of the rearing substrate according to the nutritional requirements of the target livestock is feasible. It's crucial to ensure the chemical safety of BSFLs by selecting substrates that minimize the risk of bioaccumulation of potentially hazardous compounds. The recent regulatory changes, particularly the repeal of the feed ban, have opened up possibilities for utilizing BSFL as a feed additive in livestock practice. Consequently, the use of processed proteins derived from farmed insects in pig and chicken feed is now permissible and promoted.

Conflict of interest

The Authors declare that there is no conflict of interest.

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