THE YELLOW MEALWORM (TENEBRIO MOLITOR L.) IN ANIMAL NUTRITION: ADVANCES AND PROSPECTS FOR SUSTAINABLE LIVESTOCK PRODUCTION

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Abstract: Tenebrio molitor (TM) is one of the most promising insect species proposed as an alternative to conventional food and feed in the last years. This insect is interesting for its valuable nutritional profile, lower environmental impact compared to traditional livestock and its ability to valorise organic waste. Optimising production is essential for insects farmers and several factors including environmental parameters, rearing density, nutritional strategies are involved. These aspects are essential to maximize the growth and productivity of TM. For instance, supplementing the basal diet of TM larvae with additional sources of protein, carbohydrates and lipids proved to enhance nutritional composition and growth performance of larvae. Likewise, the right combination of temperature, relative humidity, and oxygen, strongly influences development time and reproductive success. Moreover, adopting high breeding density of beetles can increase the overall productivity, although it may reduce the productivity per female. This review also examines the processing methods of TM larvae and their inclusion, in various forms, in the diets of poultry, rabbits, and aquaculture species. The incorporation at proper level of TM larvae meal showed to supports growth performance, feed efficiency and quality of the final product without compromising animal health. However, further efforts are needed to make TM meal cost-competitive with conventional feed ingredients. Finally, the review examines one of the most abundant by-products of the TM industry, the frass, highlighting its potential applications as a sustainable fertilizer, its effects on common crops, and its possible use as biochar to support more sustainable agricultural practices and environmental management strategies.

Key words: insect protein, circular economy, optimisation, alternative feed ingredient, frass

Introduction

Over the last few years, insect farming has become a major topic of discussion regarding food and feed alternatives, as well as economic and environmental sustainability. Given the projected growth of the world population by 2050 (United Nations, 2019) and the decreasing availability and rising costs of conventional raw materials used in feed production, it is essential to identify new strategies for producing nutritious and sustainable food and feed that are viable from an environmental, economic, and social perspective. These strategies should focus on shortening supply chains, reducing waste, and minimizing environmental impact. Insects have been proposed as a viable protein alternative in human, livestock and fish (Van Huis et al., 2013). They are typically harvested from nature in tropical countries, but they can also be semi-domesticated to increase production or easily reared in confined industrial facilities ranging from small to industrial scale (Oonincx and Boer, 2012).

Among insect species, Tenebrio molitor (TM) was the first one approved by EFSA for human consumption. Commonly known as the yellow mealworm, TM is one of the most studied and produced insect species in Europe (Krzyzaniak et al., 2022). It belongs to the Tenebrionidae family (Coleoptera), and it is typically considered a pest of farinaceous materials, which constitute its primary food. The life cycle of the yellow mealworm comprises four distinct metamorphic stages: egg, larvae, pupa, and beetle. The entire cycle duration is influenced by environmental parameters such as temperature, humidity and population size lasting from 60 to 90 days under optimal conditions (Hardouin and Mahoux, 2003) up to one or two years under uncontrolled environment (Selaledi et al., 2019). Larvae are characterised by an elongated cylindrical shape, six legs behind the head and two short appendages at the abdomen's tips. During the growth phase, which lasts around 60 days under controlled conditions, the larvae undergo several moults until reaching adult size characterized by an average weight of 0.2 g and a length of 25-35 mm (Aguilar-Miranda et al., 2002). If not utilised for consumption or processed, larvae go against a period of latency and a transformation into pupae, a free-living creature of yellow colour and 1 cm long. The stage of pupae typically lasts about 6 days under farming conditions (Cotton, 1927), culminating in the emergence of beetles.

TM is particularly interesting due to a high environmental sustainability and the nutritive composition. From an environmental sustainability perspective, the life cycle assessment of TM can be compared to conventional protein sources such as meat, milk, fish meal, and soy due to the high energy consumption required to maintain optimal temperatures for rearing (van Huis and Oonincx, 2017). Nevertheless, considering direct emission levels, insects proved to be more efficient than conventional livestock, with 100 times lower total greenhouse gases

emission compared to the 18% of the livestock sector. TM exhibits significantly lower CO_2 equivalents per unit of edible proteins compared to broiler chickens and beef (Oonincx and Boer, 2012). As for emissions, the land use required for insect production is significantly lower compared to traditional raw materials used in feed formulations (Oonincx et al., 2010) and livestock farming. Producing 1 kg of edible protein from TM requires only 10% of the land needed for beef production (van Huis et al., 2013). Furthermore, insect farming uses significantly less water compared to traditional livestock (Dobermann et al., 2017; van Huis and Oonincx, 2017). Embracing the concept of circular economy, TM can be fed waste raw materials, effectively converting them into high-nutritional-value food through a bio-conversion process (van Huis, 2022). This represents a significant advantage in terms of valorisation, making it possible to use organic waste as a resource for producing valuable products rather than disposing of it.

Additionally, the nutritional composition of TM larvae closely resembles that of conventional meat products, positioning them as a promising alternative source of nutrients for both human and animal diets. TM larvae are particularly rich in protein, comprising about 50% of their dry matter (DM), and contain approximately 30% lipids, which are high in beneficial fatty acids FA such as oleic, linoleic, and palmitic acids (Kröncke et al., 2019; Trukhanova et al., 2022). TM larvae also supply most of the essential amino acids required daily and serve as an excellent source of minerals, except for calcium, though calcium levels can be enhanced through dietary modifications. Furthermore, they are a valuable source of vitamins, including vitamin B_{12} (Nowak et al., 2016).

Given these considerations, insect farming offers a promising solution to the growing demand for sustainable and efficient food and feed alternatives in the face of global population growth and environmental challenges. Their ability to thrive on waste materials further enhances their role in a circular economy, reducing competition for human food resources while valorising organic waste. Despite some challenges, such as the energy needed for rearing, the overall benefits of insect farming make it a compelling option for addressing food security, environmental sustainability, and economic viability.

Tenebrio molitor farming

Ensuring optimal environmental conditions is crucial in TM rearing to optimize and improve farm productivity. Temperatures commonly used in TM farming range from 25 to 28 °C (Koo et al., 2013; Kim et al., 2015). Within this range, the productive performances of both adults and larvae are maximized. Temperatures below 17 °C have been reported to inhibit embryonic development, while temperatures above 30 °C led to increased mortality mainly due to stress condition of the insects (Koo et al., 2013). Temperature also affects energy

assimilation efficiency, with higher values observed at temperatures ranging from 23.3 to 31 °C. Additionally, the content of proteins and lipids in TM varies according to temperatures (Bjørge et al., 2018). Closely related to temperature, relative humidity (RH) represents a source of water for insects influencing their adsorption capacity and several factors during the entire life cycle. Optimal RH values ranging between 60 and 75% have been reported in previous research (Manojlovic, 1987; Punzo and Mutchmor, 1980). A combination of optimal temperature and humidity is necessary for oviposition, embryological development, and eggs hatching. Specifically, dry conditions with about 12% RH can cause embryo death due to water loss from the eggs (Punzo and Mutchmor, 1980). RH also influences the number and length of instars, adult female activity, and larval growth rates. Larval growth is maximized at RH values of 90-100% and reduced at values around 30% (Hardouin and Mahoux, 2003). However, excessively high humidity levels can create an optimal environment for the growth of undesirable pests such as mites. Water intake plays a crucial role in TM rearing. Despite TM is able to absorb water from the humidity of the environment, as reported above, levels of RH higher than 60% can reduce time for larvae growth. Additionally, optimal growth is achieved with the provision of a water source. For instance, offering fresh carrots to larvae increased their survival rate and reduced development time (Oonincx et al., 2015). Similarly, the combination of vegetables with milk enhanced larval growth rates, as observed in the study of Lee et al. (2024).

Photoperiod also has a significant impact on the growth of TM larvae. Despite this species being negatively phototropic, longer light exposure during farming appears to be beneficial for larval growth. Specifically, shorter larval development times and longer pupal periods have been observed under conditions of 14 hours of light and 10 hours of dark. Furthermore, long-day conditions have been reported to increase the eclosion rate and the pupation time (Kim et al., 2015). However, a previous study reported a higher survival and growth rate of larvae in condition of constant darkness at 25 °C and 30 °C (Eberle et al., 2022).

Another environmental parameter influencing the TM rearing is the availability of oxygen. Specifically, low oxygen concentrations of about 10-15% inhibit growth rates, cause developmental abnormalities in mealworm populations and increase larval mortality (Greenberg and Ar, 1996; Loudon, 1988). Conversely, hyperoxia conditions lead to a lower number of instars and reduced final larval biomass compared to normoxia (Greenberg and Ar, 1996).

Among the factors influencing the productivity and the reproductive success of TM, the breeding density is one of the most significant (Morales-Ramos et al., 2012). High breeding densities of larvae affect the number and the duration of larval moults, resulting in fewer larval instars and smaller larvae (Morales-Ramos et al., 2012; Morales-Ramos and Rojas, 2015). Weaver and McFarlane (1990) observed a faster larval growth at a density of 0.33 larvae/cm² compared to

0.03 larvae/cm², while larvae reared in isolation produced larger female pupae. Furthermore, in the study of Morales-Ramos and Rojas (2015), increasing the density from 0.44 larvae/cm² to 3.51 larvae/cm² reduced the growth, the feed consumption, and the feed efficiency of TM larvae. Concerning the breeding density of the beetles, high densities are typically linked to increased progeny production overall (Berggren et al., 2018; Deruvtter et al., 2019; Zim et al., 2022; Palumbo et al., 2024) (Figure 1). However, when examining progeny production per individual beetles, females raised at lower densities are significantly more prolific than those raised at higher densities (Morales-Ramos et al., 2012; Berggren et al., 2018; Deruytter et al., 2019; Frooninckx et al., 2022; Palumbo et al., 2024) (Figure 2). Indeed, high breeding density causes crowding stress in insects, which reduces the reproductive capacity of beetles along with an increased mortality due to cannibalism (Berggreen et al., 2018; Gerber, 1984; Morales-Ramos et al., 2012). Previous research has shown that all Tenebrionidae, including TM adults, exhibit cannibalistic behaviour (Kuriwada et al., 2009; Morales-Ramos et al., 2012). Specifically, under mass-rearing conditions, high densities lead to increased cannibalism of eggs, particularly those near the surface of the substrate (Kuriwada et al., 2009; Morales-Ramos et al., 2012). To prevent eggs cannibalism, separating the eggs from the beetles is a viable alternative. This can be achieved by using an oviposition grid, which allows the ovipositor to pass through the holes in the grid and lay eggs deep in the substrate. This way, the adults are unable to reach and eat the eggs. Indeed, as observed by Froonickx et al. (2022), the use of an oviposition grid resulted in a significantly higher yield of eggs.

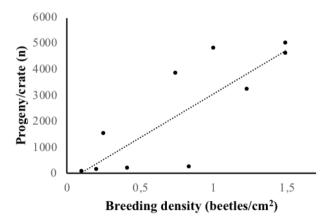


Figure 1. Linear regression of progeny produced per crate according to the beetles breeding density observed by Berggren et al. (2018), Deruytter et al. (2019) and Zim et al. (2022), modified.

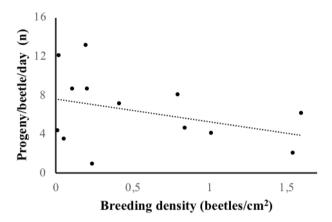


Figure 2. Linear regression of progeny produced daily per beetles according to the beetles breeding density observed by Morales-Ramos et al. (2012); Berggren et al. (2018), Deruytter et al. (2019) and Frooninckx et al. (2022), modified.

Tenebrio molitor feeding

In addition to environmental conditions, the productivity of TM can be influenced by several factors closely linked to farming techniques. One of the most significant is the choice of feeding substrate. Although TM can be raised exclusively on wheat bran, its diet can be supplemented with additional sources of protein, vitamins, minerals, and organic matter such as carrot, potato, and cabbage to provide essential nutrients. The quality of the feed directly affects oviposition: poor-quality diets can lead to fewer eggs laid and higher female mortality (Gerber and Sabourin, 1984). Van Broekhoven et al. (2015) found that incorporating 40% yeast-derived proteins into the diet of TM larvae shortened larval development time, reduced mortality, and increased weight gain and survival rates. However, this also affected the larvae's fatty acid (FA) profile, resulting in a comparable or slightly less favourable n-6/n-3 ratio than a wheat bran-only diet. Feed intake significantly influences various aspects of TM life cycle. For example, supplementing wheat bran with protein sources at a 1:4 ratio has been shown to shorten development time and improve feed conversion efficiency and fecundity. Similarly, adding carbohydrate sources at the same ratio enhances feed utilization, growth, development time, survival, and beetle fecundity (Morales-Ramos et al., 2013). Including protein and carbohydrate sources at a 1:4 ratio with wheat bran also significantly reduces the number of larval instars and the total development time (Morales-Ramos et al., 2010). Furthermore, diets containing 33-39% protein can reduce the time to pupation (Oonincx et al., 2015). Increasing dietary protein by supplementing with pea flour and rice flour has also been shown to raise the protein content of the larvae and to increase their levels of essential amino acids (Kröncke et al., 2023). Additionally, Rumbos et al. (2020) demonstrated that incorporating specific amylaceous commodities with protein contents ranging from 11% to 14%, as well as milk-based feed and layer hens feed with protein contents of 62% on average, significantly increased the larval biomass produced. However, feeding TM larvae legume flour led to a reduction in larval body weight, despite the substrate's high protein content. This suggests that while higher dietary protein generally promotes faster insect growth, as Rumbos et al. (2020) noted, the use of legume flours had an inhibitory effect, indicating that other dietary components also play a crucial role in TM larval development.

Including lipids in the diet of TM has also been tested and demonstrated to have beneficial effects at low concentrations, while high concentrations can be detrimental. This is because high lipid levels can cause substrate agglomeration, reduce aeration and subsequently cause respiration problems in mealworms (Alves et al., 2016). The FA content of the diet seems to strongly influence the lipid profile of TM. For instance, Melis et al. (2019) found that rearing mealworms on brewer's spent grain, which is rich in polyunsaturated fatty acids (PUFA), increased the levels of n-3 and n-6 PUFA in the larvae and improved feed conversion ratio (FCR). Similarly, Lawal et al. (2021) showed that the supplementation of TM diet with 10% chia seeds increased the *n-3* FA favourably reducing the n-6/n-3 ratio. In another study, Park et al. (2023) reported that including sesame cake in the TM diet enhanced the nutritional composition of the larvae. Regarding growth performance, the addition of 7.5% edible oils was found to slow larval growth (Rossi et al., 2022). In contrast, Ruschioni et al. (2020) found that including 25% olive pomace in the diet increased larval and pupal weights, improved survival rates, and extended development time. It also enhanced the protein and amino acid contents of the insects, without affecting their fatty acid profile. Furthermore, the inclusion of 5% and 10% camelina and linseed cake in the TM diet from four to nine weeks of age not only affected the FA profile of the larvae, reducing the n-6/n-3 ratio, but also improved their growth performance by increasing the final body weight and reducing the FCR (Palumbo et al., 2025b).

Tenebrio molitor harvesting and processing

As previously reported, under controlled conditions, the life cycle of TM ranges from 60 to 90 days. Typically, larvae reach the appropriate stage for processing at 8 to 9 weeks of age, at which point they are separated from residual feed and frass before undergoing further processing. Ensuring the quality and the safety of insects and insect-based products is a significant challenge in industrial

TM production. TM fresh larvae typically exhibit a high microbial load, predominantly composed of gram-positive bacteria, including faecal and total coliforms (Belluco et al., 2013). Particularly, previous research showed the possibility to isolate Enterobacteriaceae and spore forming bacteria, not belonging to pathogenic species, from TM fresh larvae (Klunder et al., 2012). Insects processing includes killing, microbial decontamination by heat, drying and grinding (van Huis and Tomberlin, 2017). Thermal treatments such as boiling, blanching, drying and other cooking methods are commonly used for microbial inactivation to ensure the safety of insects-based products (Yan et al., 2023). Indeed, boiling TM larvae for 5 minutes was found to be an efficient process to inactivate Enterobacteriaceae, whereas boiling followed by refrigeration from 5 to 7 °C resulted efficient against spore forming bacteria and spoilage (Klunder et al., 2012). Boiling larvae at temperature ranging from 70-150 °C not only affect the microbial load but also ensure the inactivation of enzymes responsible of the browning reaction caused by the oxidation of phenolic compounds (Wang et al., 2017; Cacchiarelli et al., 2022). In addition to boiling, freezing is a commonly used practice able to stop the microbial growth guaranteeing a gentle and quick death for the insects, thereby reducing their suffering (Lenaerts et al., 2018). Prolonged killing times, in fact, should be avoided to reduce the suffering of the insects and to prevent FAs oxidation, which can occur due to the stimulated metabolism of energy reserves (Larouche et al., 2019). The killing method is a crucial stage as it can also influence the proximate composition (Adámková et al., 2017; Leni et al., 2019; Caligiani et al., 2019) and the sensory traits of the final product (Farina, 2017; Palumbo et al., 2025a). For instance, in Hermetia illucens, a reduction in FA content (Caligiani et al., 2019) and in certain amino acids (Leni et al., 2019) was observed when freezing was used as the killing method, compared to blanching. Conversely, Adámková et al. (2017) found that freezing best preserved the fat content of TM larvae.

Along with the killing method, the drying process significantly influences the quality of the final product. The application of high temperatures for extended periods are responsible of a visual impairment of the larvae due to pronounced darkening and shrinkage (Purschke et al., 2018; Trukhanova et al., 2022). Furthermore, high temperatures promote the degradation of proteins and vitamins, as well as the oxidation of lipids, which deteriorate the flavour and affect both technological and functional properties of the product (Bußler et al., 2016; Kröncke et al., 2018; Baek et al., 2019). Among the drying processes involving high temperatures, oven drying, and microwave drying are the most commonly applied and tested. These processes are often proposed as alternatives to freeze-drying due to long operating times and high cost of the latter. Freeze-drying preserves the texture, the nutritive value, the aroma, and the colour of the product, promoting a sublimation of water and avoiding the application of high temperatures (Huang & Zhang, 2012). Compared to freeze drying, microwave drying has been associated

with a lower protein solubility (Kröncke et al., 2018). Additionally, microwave-treated larvae exhibited a reduction in vitamin B_{12} levels, and a darker colour compared to the fresh ones (Lenaerts et al., 2018). A dark brown colour was also observed in larvae dried using oven drying, along with significant shrinkage due to tissue collapse (Purschke et al., 2018). Thus, the optimal processing method is still not defined. However, a recent study reported that blanching at 100 °C for 180 s or freezing at -60 °C for 1h, followed by microwave drying at 103 °C for 15 min, preserved nutritional content, ensured microbiological safety, and was considered the preferred method for consumer acceptability (Palumbo et al., 2025a).

Before the use of TM larvae for protein extraction, lipid removal is required due to their high fat content. This step is essential not only to isolate the lipids for potential applications but also to enhance protein extraction, resulting in a more concentrated material with improved technological and functional properties (Choi et al., 2017). The solvents commonly employed for lipid extraction from TM larvae include ethanol, hexane-isopropanol mixture or hexane, all of which demonstrated high extraction yields (Bußler et al., 2016; Zhao et al., 2016; Purschke et al., 2017; Kim et al., 2020). However, an alternative and more sustainable method using supercritical CO₂ has shown an impressive extraction yield of 95% (Purschke et al., 2017). The protein extraction efficiency significantly influences the commercial production of food products rich in TM proteins. The chosen method should ensure a high extraction yield to meet large-scale production demands while preserving the functional properties of the proteins, resulting in a concentrate of protein that is as pure as possible (Zhao et al., 2016). The alkaline extraction followed by isoelectric precipitation is a commonly employed method for various substrates. In the case of TM protein extraction, this method resulted in an acceptable yield and a relatively high protein content of the extract (Bußler et al., 2016; Zhao et al., 2016; Kumar et al., 2019). Additionally, sonication has been observed as a further method that enhances protein extraction from edible insects (Choi et al., 2017).

Tenebrio molitor as feed

The nutritional composition of TM larvae closely resembles that of conventional meat products, positioning them as a promising alternative source of nutrients for both human and animal diets. TM have a high protein content, with crude protein comprising approximately 47-53% (DM), and a lipid content ranging 25-35% DM, depending on their diet and developmental stage. Their lipid profile includes a significant proportion of unsaturated fatty acids, notably oleic acid (C18:1) and linoleic acid (C18:2), as well as saturated FA such as palmitic acid (C16:0) (Kröncke et al., 2019; Trukhanova et al., 2022). They also supply most of the essential amino acids required daily and serve as an excellent source of

minerals, except for calcium, though calcium levels can be enhanced through dietary modifications. Additionally, TM larvae are a valuable source of vitamins, including vitamin B_{12} (Nowak et al., 2016). The excellent nutritive composition is emphasised by the ease of breeding and the reduced environmental impact of TM production compared to livestock (van Huis et al., 2013).

For these reasons, TM larvae are not only a promising alternative to conventional feed sources but also a viable substitute for traditional fish and soybean meals used in feed formulations (Makkar et al., 2014). Fish meal is an ingredient extensively used in aquaculture, pigs, and poultry nutrition obtained from small wild-caught marine fish usually not suitable for human consumption. However, due to several factor including an excessive exploitation of wild fish stocks, the availability of fish meal is decreasing, and costs are increasing (FAO, 2016b; van Huis and Oonincx, 2017).

Poultry

Several authors have investigated the inclusion of TM meal in broiler chickens' diets and found no detrimental effects on growth performance or carcass traits at inclusion levels of up to 10%. Notably, the inclusion of 0.3% full-fat TM meal in broiler diets increased feed intake (FI) and body weight gain (BWG), thereby reducing the FCR (Benzertiha et al., 2020). Similarly, Sedgh-Gooya et al. (2020) reported an increase in BWG with a 5% inclusion level of full-fat TM meal, while no significant effects were observed on FI and FCR. Andrade et al. (2023) observed that a 2% inclusion of full-fat TM meal enhanced FI and weight gain in broiler chickens and also modulated the innate immune response. Furthermore, Biasato et al. (2016) and Elahi et al. (2020) found that full-fat TM meal inclusion levels of 7.5% and 8% had no significant impact on growth performance or carcass traits. In contrast, replacing the broiler diet with 5% and 10% of whole dried TM larvae was reported to positively influence BWG, carcass yield, meat composition, and welfare traits (Vasilopoulos et al., 2023). Higher inclusion levels of full-fat TM meal have been associated with negative effects on the caecal microbiota of broiler chickens, particularly at 10% and 15% inclusion levels (Biasato et al., 2019). Additionally, Bovera et al. (2016) reported that a 29.7% inclusion of full-fat TM meal reduced ileal digestibility of DM, organic matter, and crude protein. Moreover, TM larvae can also be offered live as environmental enrichment for broilers; Bellezza Oddon et al. (2021) found that a 5% inclusion of live larvae had no negative impact on growth performance or health status. In laying hens, the effects of TM meal supplementation have shown some variability. Rahmawati et al. (2022) reported that dietary supplementation with 2% and 5% full-fat TM meal enhanced the n-3 content of eggs and increase egg weight, without significant effects on physical egg quality. However, Ait-Kaki et al. (2021) observed that a 5% inclusion level reduced both the final body weight of hens and egg weight, while shell breaking strength increased. Ko et al. (2020) found no significant effects on

egg production with full-fat TM meal inclusion levels ranging from 1% to 3%; however, 3% inclusion level led to a more intense yolk colour and an increase in histidine content. Expanding beyond chickens, the inclusion of live TM larvae has been tested as a nutritional enrichment in laying quails. Dalle Zotte et al. (2024) reported that an inclusion of 10% did not compromise growth performances, indicating potential applications across various poultry species.

Rabbit

While the majority of research on TM as a feed ingredient has focused on poultry species, its potential applications extend to other livestock, including rabbits. In particular, incorporating 4% full fat TM meal into the diet of New Zealand White rabbits was found to increase final body weight (BW) and carcass yield (Kowalska et al., 2020). While the FA profile remained unchanged, a difference in amino acid content was observed, with higher levels of isoleucine and methionine in rabbit fed with TM meal. Similarly, in a subsequent study, Kowalska et al. (2021) reported that replacing 4% of soybean meal with full fat TM meal increased the rabbits' BW and muscle fat content, supporting the potential for partial replacement of soybean meal in rabbit diets. Additionally, the inclusion of 4% full fat TM meal was found to increase the apparent total tract digestibility of ether extract, acid detergent fibre, and acid detergent lignin, as well as to enhance the activity of caecal and colon bacterial N-acetyl-\beta-D-glucosaminidases (NAGases) (Strychalski et al., 2021). However, this inclusion level was associated with a reduction of enzyme activity and lower short-chain FA concentrations in the hindgut. The full replacement of soybean meal with 30 g/kg of TM meal has been shown to maintain rabbit performance without adverse effects (Volek et al., 2021). Furthermore, rabbits fed diets containing TM larvae meal exhibited reduced nitrogen losses in urine and decreased total nitrogen excretion. These findings were supported by a subsequent study, in which no adverse effects on growth performance or nitrogen output were observed (Volek et al., 2023).

Research on TM larvae fat inclusion in rabbit diets, primarily as a substitute for soybean oil, has also yielded promising results. Partial (50%) and total replacement of soybean oil with TM fat have been found to have no detrimental effects on growth performance, apparent digestibility, gut morphometric indices, or organ histopathology in rabbits. Moreover, this substitution did not negatively impact consumer acceptance of rabbit meat and, notably, reduced the meat's susceptibility to oxidation (Gasco et al., 2019).

Aquaculture

The inclusion of TM in aquaculture feed, either as a partial or complete replacement for fishmeal (FM), has been proposed as a promising strategy to enhance the sustainability of the aquafeed industry. Defatting TM meal is generally

preferred, as it facilitates the extrusion process, resulting in more stable pellets and reduced degradation (Rema et al., 2019).

The replacement of FM with 15% or 30% of full fat TM meal was evaluated in the diet of rainbow trout ($Oncorhynchus\ mykiss$), showing no adverse effects on growth performance, protein utilization, or physiological status. However, at the highest level of FM replacement, a significant reduction in n-3 FAs in the fillet was observed, along with a decreased viscerosomatic index and increased aerobic catabolism (Melenchón et al., 2020).

Studies on gilthead seabream (*Sparus aurata*) have shown promising results when partially replacing FM with TM larvae meal. Piccolo et al. (2014) reported that substituting 25% of FM with full-fat TM meal resulted in no significant negative effects on BWG or final BW. A subsequent study by the same research group (Piccolo et al., 2017) corroborated and extended these findings. They observed that 25% inclusion level of full-fat TM meal (corresponding to 35% of FM substitution on protein basis) led to statistically significant improvements in BWG, final BW, specific growth rate (SGR), and FCR.

Research on higher inclusion levels of TM meal in fish diets has yielded variable results across species. In rainbow trout, inclusion levels exceeding 25% of full-fat TM meal have been associated with adverse effects. FM replacement at 50%, 75%, and 100% with full-fat TM meal has resulted in significant reductions in BWG and final BW, coupled with increased FCR (Valipour et al., 2019). Additionally, a 50% inclusion of full-fat TM meal in replacement of FM has been shown to alter both total and free amino acid profiles in rainbow trout fillets, with notable reductions in free alanine, leucine, isoleucine, and lysine at the highest inclusion level (Iaconisi et al., 2019).

Referring to the European seabass (*Dicentrarchus labrax*), FM replacement of up to 80% with defatted TM meal has not compromised growth performance, nutrient utilization, flesh quality, or intestinal morphology (Basto et al., 2019). Even at 100% replacement of FM with defatted TM meal, growth performance parameters remained unaffected, although an increase in hepatosomatic index and alterations in FA profile were observed. At 80% replacement, protein and lipid digestibility remained unaltered along with growth performance (Basto et al., 2021).

In rainbow trout, complete replacement of FM with partially defatted TM meal has shown no significant adverse effects on gut and skin microbiota (Terova et al., 2021). High dietary inclusion of defatted TM meal, up to 100% replacement of FM, has been reported to increase lipid and energy content without significantly affecting blood gas pressure, electrolyte balance, or haematocrit (Sakhawat Hossain et al., 2025). These findings collectively suggest that the effects of TM meal inclusion are species-specific and dependent on the processing method of the meal (full-fat *vs* defatted). While high inclusion levels may be detrimental in some

species, others appear to tolerate substantial FM replacement with TM meal without compromising key physiological parameters.

The inclusion of TM meal has also been investigated in Pacific white shrimp (*Litopenaeus vannamei*) diets, yielding promising results, albeit with some variability. Replacing up to 30% of FM with TM meal led to increased BWG, feed efficiency, and hepatopancreas index, whereas higher inclusion levels were associated with reductions in these parameters. Additionally, higher inclusion levels were found to decrease cholesterol, triglycerides, and glucose levels (Sharifinia et al., 2023). Contrasting results were reported by Chung et al. (2015), where FM replacement at 50% and 100% resulted in increased BWG and SGR, along with improved FCR.

The substitution of fish oil (FO) with TM oil in the diet of Pacific white shrimp has demonstrated varying effects depending on the level of replacement. While complete substitution of FO with TM oil did not significantly alter shrimp performance parameters (Panini et al., 2017), partial replacement has yielded more favourable outcomes. Eom et al. (2023) reported that replacing 50% to 75% of FO with TM oil resulted in enhanced feed utilization efficiency. Moreover, their findings suggest that the optimal FO replacement level is dependent on the desired outcome: a 50% substitution was found to be optimal for growth performance, whereas a 25% replacement showed the greatest benefit for disease resistance against *Vibrio parahaemolyticus*.

Tenebrio molitor by-products – frass

Frass is the primary by-product of large-scale insect farming. Although it is often considered waste, frass possesses a nutrient profile rich in readily mineralisable macronutrients (NPK) and micronutrients, making it suitable for various applications (Houben et al., 2021). Typical frass composition includes approximately 2-5% nitrogen, 1.5-2.6% phosphorus, and 1.1-2.0% potassium and about 14% lipids (Poveda et al., 2019; Houben et al., 2021; Antoniadis et al., 2023; Palumbo et al., 2024; Amorim et al., 2024).

Frass predominantly comprises insect excrement, along with smaller amounts of shed exoskeletons and undigested or residual feed. Utilisation of this by-product can enhance the sustainability and profitability of insect farms. Numerous studies have examined the diverse applications of frass, particularly its potential as an organic fertiliser in agriculture due to its favourable NPK composition. Its chemical profile closely resembles that of poultry manure, with comparable plant growth rates and yields observed (Hénault-Ethier et al., 2023).

TM frass, in particular, has gained attention as a promising organic soil amendment rich in both macro- and micronutrients as well as plant growth-promoting microorganisms (PGPMs) (Fuertes-Mendizábal et al., 2023; Nogalska et

al., 2024). Research indicates that TM frass can match or even outperform synthetic fertilisers, especially when used in combination with mineral fertilisers. Application of TM frass at a concentration of 2% (v/v) to chard (*Beta vulgaris* var. cicla) has been shown to supply an NPK ratio of 3-1.5-2, along with essential micronutrients such as sulphur, calcium, magnesium, manganese, iron, and molybdenum. This treatment enhanced chlorophyll content, fresh weight, shoot length, and stem thickness (Poveda et al., 2019). The presence of calcium and magnesium in TM frass has been observed to mitigate the acidifying effects of urea when used in peat-based substrates (Nogalska et al., 2022).

TM frass, applied at 10 t/ha⁻¹ (NPK: 5-2-2) has been found to increase barley biomass (Houben et al., 2021). In wheat cultivation, soils treated with TM frass, alone or in combination with mineral fertilisers, exhibited improved root development, grain quality, and yields, achieving results equal to or better than those obtained with conventional synthetic fertilisers (Nyanzira et al., 2023). The beneficial effects of TM frass as an organic fertiliser on crop growth are closely linked to its application rate. In lettuce cultivation, frass application at a concentration of 1% (v/v) supplied sufficient macronutrients to support the crop's needs, resulting in increased aboveground biomass. However, when the application rate was raised to 2.5% (v/v), these benefits were reversed, leading to reduced plant growth, with the root system being particularly affected (Fuertes-Mendizábal et al., 2023).

TM frass can also be converted into biochar, offering promising applications in environmental remediation. Through pyrolysis, nutrients are stabilised, and a carbon-rich material is formed (Kumar et al., 2023; Thalassinos et al., 2023). Biochar derived from TM frass has been shown to function effectively as a bio-adsorbent for purifying wastewater and remediating contaminated soils (Yang et al., 2019). Moreover, TM frass biochar can aid in carbon sequestration supporting circular economy goals and reducing greenhouse gas emissions. TM frass biochar activated at various temperatures has been observed to capture 90% of CO₂ within 20 minutes, with biochar produced at 700 °C exhibiting the most favourable CO₂ storage and electrochemical behaviour (Wang et al., 2022).

Beyond these applications, recent research explored TM frass as bedding material for broiler chickens. Inclusion at 50-75% in replacement to wood shavings did not affect growth performance, while numerically reducing hock burn and footpad dermatitis incidence, and lowering microbial load, suggesting potential benefits for animal welfare (Tóth et al., 2025). A further study (Tóth et al., unpublished) with larger flocks is being conducted to confirm these preliminary results.

Conclusion

The optimisation of environmental parameters, including temperature, relative humidity, photoperiod, oxygen concentration, and rearing density, in conjunction with balanced nutritional strategies, is crucial for maximizing the growth and productivity of TM. While wheat bran serves as a suitable basal diet, supplementing with additional feedstuffs to the purpose of fulfilling the nutritional requirements in protein, carbohydrates, and lipids has been demonstrated to enhance larval development and nutritional composition. TM larvae represent a promising and nutrient-dense alternative to conventional feed ingredients across various livestock sectors, including poultry, rabbit, and aquaculture. When incorporated at appropriate inclusion levels, TM larvae meal supports growth performance and feed efficiency without compromising animal health. Furthermore, TM fat has shown potential as a viable substitute for traditional oils in feed formulations. However, the current limited commercial availability and relatively high production costs of TM larvae present significant challenges to their widespread adoption in livestock feed formulations. Competition with more economical conventional feed ingredients remains a substantial barrier to the largescale implementation of TM in livestock nutrition. The by-product of TM production, the frass, is demonstrating significant potential as organic fertiliser and soil amendment. Its application could contribute to sustainable agricultural practices and environmental management strategies, potentially offering additional value streams for TM producers. Collectively, these attributes position TM and its associated by-products as potential contributors to sustainable, circular food production systems. However, continued research efforts are essential to further refine rearing methodologies, optimise nutrient profiles, and facilitate the sustainable scaling of production.

Žuti brašnari (*Tenebrio molitor* L.) u ishrani životinja: napredak i izgledi za održivu stočarsku proizvodnju

Antonella Dalle Zotte, Bianca Palumbo

Rezime

Tenebrio molitor (TM) je jedna od najperspektivnijih vrsta insekata predloženih kao alternativa konvencionalnoj hrani i stočnoj hrani poslednjih godina. Ovaj insekt je zanimljiv zbog svog vrednog nutritivnog profila, manjeg uticaja na

životnu sredinu u poređenju sa tradicionalnim farmskim domaćim životinjama i svoje sposobnosti da valorizuje organski otpad. Optimizacija proizvodnje je neophodna za uzgajivače insekata i uključeno je nekoliko faktora, uključujući parametre životne sredine, gustinu uzgoja i strategije ishrane. Ovi aspekti su neophodni za maksimiziranje rasta i produktivnosti TM. Na primer, dopunjavanje bazalne ishrane larvi TM dodatnim izvorima proteina, uglienih hidrata i lipida pokazalo se da pobolišava nutritivni sastav i performanse rasta larvi. Isto tako, prava kombinacija temperature, relativne vlažnosti i kiseonika snažno utiče na vreme razvoja i reproduktivni uspeh. Štaviše, usvajanje visoke gustine razmnožavanja buba može povećati ukupnu produktivnost, iako može smanjiti produktivnost po ženki. Ovaj pregled takođe ispituje metode obrade larvi TM i njihovo uključivanje, u različitim oblicima, u ishranu živine, zečeva i vrsta akvakulture. Pokazalo se da uključivanje brašna larvi TM na odgovarajućem nivou podržava performanse rasta, efikasnost ishrane i kvalitet finalnog proizvoda bez ugrožavanja zdravlja životinja. Međutim, potrebni su dalji napori kako bi se sačma od transkripcionih sastojaka učinila konkurentnom po ceni u odnosu na konvencionalne sastojke hrane za životinje. Konačno, pregled ispituje jedan od najzastupljenijih nusproizvoda transkripcione industrije, insektni izmet, ističući njegove potencijalne primene kao održivog đubriva, njegove efekte na uobičajene useve i njegovu moguću upotrebu kao biouglja za podršku održivijem poljoprivrednim praksama i strategijama upravljanja životnom sredinom.

Ključne reči: proteini insekata, cirkularna ekonomija, optimizacija, alternativni sastojak hrane za životinje, insektni izmet.

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Conflict of interest

The authors declare no conflict of interest.

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