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Research Paper

Valorization of organic waste through black soldier fly: On the way of a real circular bioeconomy process

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A B S T R A C T

The transition from a linear to a circular production system involves transforming waste into valuable resources. Insect-mediated bioconversion, particularly using black soldier fly (BSF) larvae, can offer a promising opportunity to convert the organic fraction of municipal solid waste (OFMSW) into protein-rich biomass. However, current regulatory restrictions do not allow the use of this substrate to obtain insect proteins for animal feed, prompting the exploration of other applications, such as the production of bioplastics. Here, we explored at laboratory scale an innovative and integrated circular supply chain which aims to valorize the OFMSW through BSF larvae for the production of biobased materials with high technological value. BSF larvae reared on this organic waste showed excellent growth performance and bioconversion rate of the substrate. The use of well-suited extraction methods allowed the isolation of high-purity lipids, proteins, and chitin fractions, which are building blocks to produce biobased materials. In particular, the protein fraction was used to develop biodegradable plastic films which showed potential for replacing traditional petroleum-based materials, with the possibility to be fully recycled back to amino acids. Socioeconomic analysis highlighted values generated along the entire supply chain, and life cycle assessment pointed out that lipid extraction was the most challenging step: implementation of more sustainable methods is thus needed to reduce the overall environmental impact of the proposed chain. In conclusion, this study represents a proof of concept gathering evidence to support the feasibility of an alternative supply chain that can promote circular economy while valorising organic waste.

1. Introduction

In recent years, various strategies are being considered worldwide to cope with environmental issues, including the implementation of a circular economy approach across sectors, aimed at halving carbon emissions within 2030 and achieving carbon neutrality by 2050 (Dantas

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et al., 2021). In this context, the disposal of the organic fraction of municipal solid waste (OFMSW) poses a significant challenge, reaching 900 million tonnes/year globally and expected to increase by 70 % within 2050 due to a rise in food waste (United Nations Environment Programme, 2021). The existing recovery systems of the OFMSW (i.e., anaerobic digestion and composting) are deemed unsatisfactory and somehow challenging as they depend on the chemical composition of the feedstock, offer minimal value, and raise environmental and public health issues (Paritosh et al., 2018), especially in developing countries (Adedara et al., 2023).

Among the recently proposed alternative strategies for recycling organic waste there is insect-mediated bioconversion. In particular, the black soldier fly (BSF), *Hermetia illucens*, is considered the most promising insect for valorising waste and by-products of the agri-food value chain (Athanassiou et al., 2024). In fact, thanks to their remarkable midgut plasticity (Bonelli et al., 2020; Bruno et al., 2024), BSF larvae can grow on a wide variety of organic leftovers and side streams, reducing the waste volume and transforming it into protein-rich biomass (Rehman et al., 2023). Numerous studies have been undertaken to characterize the growth performance of these larvae on different organic leftovers (Broeckx et al., 2021; Ceccotti et al., 2022), to define how the gut microbiota is shaped by the rearing substrate (Bruno et al, 2019a; Vandeweyer et al., 2023; De Filippis et al., 2024), and to evaluate the potential interference of contaminants in the substrate, such as heavy metals (Hu et al., 2023), pesticides (Meijer et al., 2021) and other chemicals (van der Fels-Klerx et al., 2020), demonstrating the safety and suitability of insect-mediated bioconversion for developing valuable circular supply chains. However, at least in Europe, the utilization of BSF meals in the feed sector is restricted by the current legislation which allows the use of insect proteins for animal feed production only if insects are reared on microbiologically and chemically safe substrates (European Commission, 2017). Therefore, while the use of other components derived from BSF larvae, such as lipids for the production of biodiesel and surfactants, does not pose particular limitations, alternative applications for proteins obtained from larvae grown on food waste must be explored.

A topic strictly related with waste management is plastic pollution. Indeed, the non-biodegradable nature of oil-based plastics has led to the catastrophic accumulation of a large amount of plastic waste and microplastics over recent decades, making plastic waste management one of today's most urgent environmental challenges (Shen et al., 2020; Ford et al., 2022). This issue is amplified by the widespread use of single-use plastics, especially in packaging, where products have a short life span between their production and disposal. In cases where plastic recycling is not feasible due to technical or economic constraints (Garcia and Robertson, 2017; Volk et al., 2021), governing institutions are thus increasingly promoting the use of renewable and biodegradable plastics. In this scenario, proteins are emerging as one of the most promising classes of biopolymers for the development of environmentally friendly bioplastics. The search for alternative sources of proteins (not in competition with the food and feed supply chains) and the generation of functional and biodegradable protein-based materials are thus two primary, leading-edge issues in research (Kamada et al., 2021; Shen et al., 2021; Peydayesh et al., 2021; Li et al., 2023; Peydayesh et al., 2023).

Few studies have pioneered the use of BSF proteins for producing bioplastics, but their performance was unsatisfactory, mainly due to their limited stability against chemical and physical agents, as well as the non-competitive costs of these derivatives, leaving large-scale application of such films substantially unexplored (Barbi et al., 2019; Barbi et al., 2021; Setti et al., 2020). Moreover, to the best of our knowledge, no studies have compared the production of bioplastics using proteins obtained from different developmental stages of BSF (i.e., larvae and pupae), which is one of the targets of the present work.

Here, we present an innovative and integrated circular supply chain that, starting from the biotransformation of the OFMSW through BSF larvae, leads to the targeted production of bioplastic films with high technological potential. In addition, a comprehensive assessment of the technological landscape, economic feasibility, and expected environmental impact of the whole production chain is presented.

2. Materials and methods

2.1. Insect-mediated bioconversion of the OFMSW

Insects were reared as reported in Pimentel et al. (2017). Batches of 300 six-day-old larvae were transferred to plastic containers (16 × 16 × 9 cm), fed with the experimental substrates, and kept in the dark at 27 ± 0.5 °C and 70 ± 5 % relative humidity (Bruno et al., 2019b). Two feeding substrates were used in this study: the OFMSW sampled from household waste (r-OFMSW, where "r" stands for "real") in five cities of Lombardy Region (Italy) and the surrogate-OFMSW (s-OFMSW), which was formulated to reproduce the OFMSW (see Bruno et al., 2024 for details) and selected to perform the experiments under reproducible and standardized conditions. Both substrates were minced before feeding the larvae. Insects were then collected at the appropriate developmental stage for the analyses (i.e., last instar larvae and 4-day-old pupae).

The growth performance of the larvae and the efficiency of the bioconversion of the r-OFMSW were evaluated through the following indexes: Relative Growth Rate (RGR), substrate reduction (D), Waste Reduction Index (WRI), Efficiency of Conversion of Ingested food (ECI), Nitrogen Conversion Efficiency (NCE), and Survival Rate (SR) (see Supplementary Table 1 for details). The rearing substrate, rearing residue (frass), and insects were frozen at −20 °C for 24 h and then dried at 60 °C overnight to determine the total content of dry matter for the calculation of the bioconversion indexes. All the experiments were conducted in triplicate.

The rearing substrate, larvae, and pupae were subjected to freezing and drying, as reported above, and finely minced. The samples were then analyzed by La-Chi laboratory (University of Padova, Italy) to determine their chemical composition. Crude protein, lipid and fibre, nitrogen-free extract, and ash content were determined according to AOAC International (Horwitz, 2000; Latimer, 2016). Crude protein content was determined considering nitrogen-to-protein conversion factor (Kp) of 6.25 for rearing substrate and 5.6 for insects (Janssen et al., 2017). Starch was determined by enzymatic digestion followed by glucose quantification by HPLC. Free glucose and fructose were quantified by HPLC, too. Chitin was calculated by using Van Soest acid detergent fibre method (Stelmock et al., 1985).

2.2. Extraction and quantification of components in protein and lipid fractions

2.2.1. Protein, lipid, and chitin extraction

After rearing on s-OFMSW and r-OFMSW, last instar larvae and 4-day-old pupae were sampled and used for lipids, proteins, and chitin extraction. The extraction procedure (Fig. 1) was carried out from five biological replicates for each condition and each analysis was repeated at least twice. For each replicate, the procedure used ≈20 g of dried biomass. Each sample was ground with an IKA A10 grinder for 2 min and immediately used for extraction.

Lipids were obtained by Soxhlet extraction (Caligiani et al., 2018) with petroleum ether (technical, boiling point 40–60 °C, Exacta + Optech Labcenter S.p.A., Modena, Italy) for 18 h. An alternative batch procedure for lipid extraction was also evaluated: briefly, grinded BSF biomass and petroleum ether were stirred at 1:2 w/v ratio for 1 h and then decanted for 5 min; the solvent containing fats was recovered by centrifugation at 2000 × g for 12 min (the procedure was repeated twice). Extracted lipids were stored at −20 °C under N_2 and unfrozen prior to the analysis.

Proteins were extracted from the lipid-free BSF-powder with a modified procedure from Smets et al. (2020). The defatted sample was dispersed in demineralized water (1:10 ratio w/v) under conditions of

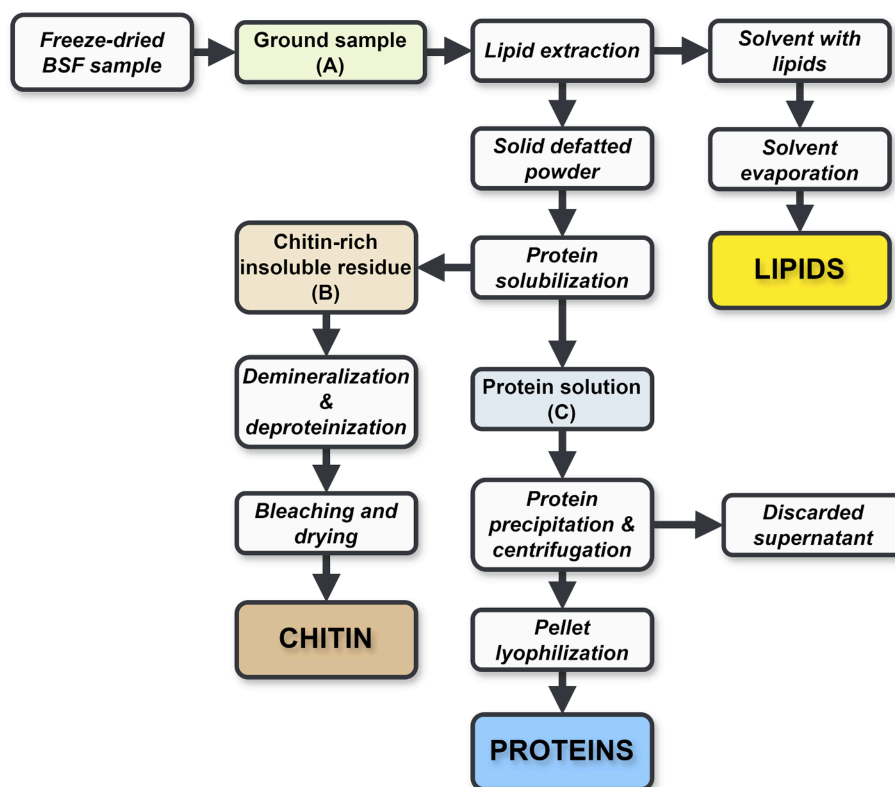


Fig. 1. Schematic representation of the biomolecule fractionation strategy set up in the present work. Intermediate compounds are indicated by A, B, and C. The final fractions extracted from BSF larvae and pupae are highlighted in brown, cyan, and yellow for chitin, proteins, and lipids, respectively. Processes are indicated in italics. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article).

maximal protein solubility (pH 11.0, adjusted with 1.0 M NaOH) and kept at 45 °C for 2.5 h. The suspension was centrifuged at 1840 × g for 1 min and filtered; the solid fraction containing chitin was stored at −80 °C and then lyophilized. The protein-containing supernatant was adjusted with 1 M HCl solution to pH 4 to induce protein precipitation. The solution was centrifuged at 1840 × g for 45 min and the pellet was frozen at −80 °C and lyophilized.

2.2.2. *Total carbohydrate content*

The carbohydrate content of the ground sample (Fig. 1, sample “A”) and of the final powder of protein extract (Fig. 1, sample “PROTEINS”) was measured with the phenol–sulfuric acid method (Sadasivam and Manickam, 2005), using glucose as standard. Before the analysis, chitin was removed from ground sample by solubilization (100 mg) with 10 mL of 3 N HCl and boiling for 3 h; the solution was neutralized with Na_2CO_3 at room temperature and 1 mL spinned at 16060 × g for 15 min. The protein extracts were dispersed in 0.1 M phosphate buffer (pH 11).

2.2.3. *Composition of lipid extracts*

The lipid extraction yield was determined as the weight ratio (% w/w) to the initial dried insect sample, after petroleum ether evaporation at 50 °C using a Laborota 4000 rotary evaporator (Heidolph, Schwabach, Germany). All samples were analyzed at UNITECH OMICs (University of Milano, Italy) using ExionLC™ AD system (SCIEX) connected to TripleTOF™ 6600 System (SCIEX) equipped with Turbo V™ Ion Source with ESI Probe. Following extraction with 2-propanol:acetonitrile (90:10, v:v), 0.1 % formic acid, and 10 mM ammonium acetate, samples were added with 10 µL of Internal Standard (IS, from Splash Lipidomix 330707-1EA, Avanti Polar Lipids). Each sample was analyzed twice in ESI positive and ESI negative modes. Data were analyzed with MS-DIAL software (ver. 4.24) integrated by the LipidBlast database (version 68).

2.2.4. *Composition of protein extracts*

The protein extraction yield was calculated as the weight ratio (% w/w) of the extracted dried proteins to the initial dried insect sample weight. The total protein content was determined by acid hydrolysis followed by quantification of the amino acid content through the ninhydrin reagent (Starcher, 2001), measuring absorbance at 575 nm.

The residual carbohydrate and DNA contents (% w/w) in protein extracts was assessed on each sample (10 mg) dispersed in 1 mL of 0.1 M phosphate buffer (pH 11). The carbohydrate content was measured as reported above. DNA content was determined by the DNA Quantitation kit (Bisbenzimide fluorescent assay, Merck KGaA, Darmstadt, Germany). Soluble protein content at different pH values was analyzed by the Bradford method (see below).

2.2.5. *Chitin and ash content*

Chitin was extracted through a formic acid/sodium hydroxide method (Hahn et al., 2022). In detail, 3 g of the defatted, deproteinized samples (Fig. 1, sample “B”) were added with 45 mL of 0.5 M formic acid (Merck, St. Louis, MO, USA) warmed at 40 °C and stirred at 200 rpm for 2 h for demineralization. Samples were centrifuged and the pellet was washed twice with Milli-Q water. After the addition of 30 mL of 1.25 M NaOH (for deproteinization), samples were heated up to 90 °C and stirred for 4 h, centrifuged, and washed again; then they were vacuum filtered and 150 mL boiling water were added to remove residual proteins and dyes. Samples were finally dispersed in 25 mL 4.5 % NaClO (v/v) and placed in a hybridization oven at 55 °C for 1 h. After the last spinning and washing step, the resulting white material was desiccated in the oven at 70 °C overnight. The extraction yield was calculated as the weight ratio (% w/w) of the dried and bleached chitin extract to the initial dried insect sample.

Ashes were determined according to UNI EN 14775. Ground samples (Fig. 1, sample “A”) were transferred into small ceramic crucibles and

dried overnight at 70 °C, incinerated using a muffle furnace at 550 °C for 8 h, dried into a silica desiccator, and the weight was determined gravimetrically. Ashes value was calculated as the weight ratio (% w/w) of the ashes to the initial dried insect sample.

2.3. Proteomic analysis and physical/chemical characterization of protein extracts

2.3.1. Proteomic analysis

Proteomic analysis was performed on the protein fraction recovered from larvae and pupae grown on s-OFMSW, separated by SDS-PAGE, followed by trypsin digestion and nLC-MS/MS analysis. Briefly, dried protein extracts were suspended in Milli-Q water up to a final concentration of 5 mg/mL and 1 M NaOH was added dropwise to reach a pH of 12. Samples were heated at 80 °C for 20 min. A 10 μL aliquot of this solution was mixed with an equal volume of Laemmli buffer 2X and loaded onto the gel for one-dimensional electrophoresis (Boreggio et al., 2022).

For nLC-MS/MS analysis, thin gel slices (1 mm each) were cut from SDS-PAGE lanes: each slice was treated with 0.02 μg/μL trypsin (in 25 mM Ambic) at 37 °C overnight, as detailed in Boreggio et al. (2022). MS data were analyzed by the Mascot search engine (Version 2.3.01), using the Proteome Discoverer software (Version 1.2.0 Thermo), and UniProtKB/SwissProt as protein database (Uniprot_Insecta_Reviewed, total sequences 10974, total residues 5363805). MS analysis was performed in triplicate, both for larvae and pupae samples, and the final identifications were obtained after validation.

2.3.2. Protein extracts solubility and secondary structure content

The solubility of protein samples at different pH was assessed using a modified version of the method by Mshayisa et al. (2022). Briefly, each sample (10 mg) was dispersed in 1 mL phosphate buffer and adjusted at different pH values, then incubated on a rotary shaker at 9 rpm for 30 min, at room temperature. Samples were centrifuged at 16060 × g for 20 min at 4 °C. Protein content of the supernatant was determined by the Bradford assay (Sigma-Aldrich, Saint Louis, MO, USA). Protein solubility was calculated as percentage (% w/w) of the total protein content. For UV–visible spectroscopy measurements, extracts solubilized at 0.2 mg/mL at the pH of maximum solubility were analyzed in the 240–800 nm range using a HP 8452A Diode Array spectrophotometer (Agilent Technologies, Santa Clara, CA, USA). Protein secondary structure in water was assessed through circular dichroism (CD), as reported in (Caldinelli et al., 2010), using a JASCO J-815 spectropolarimeter.

2.3.3. Protein charge (ζ-potential)

Protein ζ-potential was evaluated through electrophoretic light scattering (ELS) measurements. Protein extracts were dispersed at 1 mg/mL in Milli-Q water and brought to pH values ranging from 2 to 12 by adding small volumes of 1 N NaOH or HCl. Samples were then heated at 80 °C for 20 min. The analysis was performed in triplicate at room temperature (22 ± 2 °C) in DTS0012 type cuvettes by means of a Zetasizer Nano (Malvern Instruments, Malvern, UK). ζ-potential was automatically calculated from the electrophoretic mobility by means of the Hemholtz–Smoluchowski relation. 10 runs were performed for each measurement. All measurements were carried out in triplicate.

2.4. Production and characterization of bioplastics from insect proteins

2.4.1. Bioplastics production

Bioplastics production was carried out using proteins from larvae and pupae grown on either s-OFMSW or r-OFMSW (Fig. 1, sample "PROTEINS"), according to the protocol by Barbi et al. (2019) and Nuvoli et al. (2021) with slight modifications (Fig. 3A). Briefly, 250 mg of protein extracts were weighed and dispersed in 4.5 mL Milli-Q water. Extracts were solubilized by adding 0.5 mL of 1 N NaOH to a final pH of 11.5. The obtained protein suspensions (5 % w/v) were heated at 80 °C for 20 min and then brought to their initial volume by adding water; they were added with 125 mg of glycerol (50 % w/w on protein weight) and the resulting mixture mixed for 10 min under stirring at 400 rpm. The suspension was eventually cast onto silicone moulds: films were recovered after 24 h at 30 °C. Unless otherwise specified, films were placed in sealed plastic bags and stored in the dark at 20 °C, until further characterization.

2.4.2. Surface chemical groups of bioplastics

Surface chemical groups of bioplastics were investigated in ATR-FTIR mode using a Nicolet iS5 with KBr windows imaging system (Thermo Fisher Scientific, Waltham, MA, USA) equipped with a Vee-MAX III ATR (Pyke technologies, Madison, WI, USA), with a germanium crystal. Background and sample spectra were collected after > 20 min of N_2 purge. The IR absorption spectra were recorded in the 4000–600 cm^{-1} region based on 64 scans and a resolution of 4 cm^{-1}, with a beam incident angle of 55°.

2.4.3. Mechanical properties of bioplastics

For mechanical characterization, rectangular specimens (l = 25 mm, w = 5.45 mm, n = 3 for each formulation) were punched out from cast bioplastic films, and conditioned at 30 °C and 40 % relative humidity for 24 h. Tensile stress/strain curves were obtained under quasi static conditions using a Dynamic Mechanical Analyzer (MCR702, Anton Paar) equipped with tension clamps. A preload of 0.1 N was applied, and elongation rate was set at a 2 mm min^{-1}.

2.5. Life cycle assessment

The LCA was carried out assessing bioplastics production from larvae and pupae with the goal to evaluate which source of protein was more effective in terms of (reduced) environmental impact of the system. Since the herein proposed value chain is not available on an industrial scale yet, primary data were collected for the analysis at laboratory scale, scaling up the electricity consumption. The functional unit (FU) of data collection referred to 1 kg of treated r-OFMSW, while results were presented per tonne of OFMSW potentially handled by a waste treatment facility. The analysis was carried out with SimaPro v.9.4 software and Ecoinvent 3.4 database. Impacts 2002+ was used as an impact assessment method. The system boundaries covered the processes from the OFMSW storage and its use for BSF larvae rearing to bioplastic production. The LCA did not consider the waste transport and the construction of treatment and valorization plants.

For the electricity mix the analysis referred to the Italian context while for the virgin materials to the European Union. The reference year was 2023, when the study was carried out. Details about the criteria and the complete life cycle inventory per FU are reported in Supplementary Text 1 and Supplementary Table 2, respectively.

2.6. Socioeconomic assessment

The socioeconomic analysis focused on three main frameworks: i) the innovation framework, with a patent landscape analysis (PLA); ii) the market opportunities framework, for the by-products and final products; and iii) the socioeconomic feasibility framework, aimed at identifying the elements of the Cost-Benefit Analysis (CBA).

PLA was conducted using Open data sources (Espacenet, Lens.org), in accordance with the WIPO guidelines (Trippe, 2015), to provide an overview of the main technological trends concerning the 3 main phases of the value chain: i) BSF-mediated bioconversion process of the OFMSW; ii) extraction of lipids, proteins, and chitin; and iii) preparation and characterization of protein-based bioplastics.

As to the two remaining frameworks, the analysis considered the process from the collection and management of the OFMSW (as the basis input for the larvae/pupae production line) to the choice of the main uses for insect-derived products and by-products, considering the

related economic sectors and final market opportunities. The CBA used these activities as the basis for the identification of the socioeconomic benefits that pile up along the production process, forming the so-called value chain.

If the process is evaluated in a circular bioeconomy framework, the final aim is the maximization of social welfare by weighting: i) externalities (i.e., positive or negative effects that the production has on the society) and ii) opportunity costs (i.e., benefits foregone by choosing a course of action different from what has been done before or from what might be done using the same materials, products or production processes) (OECD, 1993). Opportunity costs, that can be either positive or negative and are often related to externalities, are especially relevant from an environmental and social benefit perspective (Aleisa and Heijungs, 2022).

2.7. Statistical analysis

Statistical analysis was performed using GraphPad Prism version 7.00 (GraphPad software, La Jolla, SD, USA). Possible differences between bioconversion parameters, as well as between the solubility of protein derived from larvae and pupae, were checked by using the unpaired Student's *t*-test followed by Tukey's multiple-comparison *post hoc* test. Statistical differences between groups were considered significant at a p-value < 0.05.

3. Results and discussion

3.1. Bioconversion efficiency of the OFMSW

The s-OFMSW and r-OFMSW differed in terms of chemical composition (Table 1): the content of the main macronutrients (i.e., crude protein, lipid and fibre, and starch) was lower in r-OFMSW, while simple sugars, such as glucose and fructose, were more abundant.

The growth performance of the larvae and their efficiency in reducing and bioconverting the substrate are reported in Table 2. The maximum weight of the larvae on the two substrates was comparable, although three more days were necessary to insects reared on s-OFMSW to complete the larval period.

The difference in developmental time affected the RGR, which was higher in larvae reared on r-OFMSW. The comparable maximum weight reached by the larvae on substrates with differences in nutrient composition could be related to the different efficiency in the conversion of the ingested food. Actually, although larvae on r-OFMSW reduced the substrate to a lower extent (73.3 % compared to 83.5 % of s-OFMWS), their ECI was higher (24.1 % compared to 20.8 % of s-OFMSW). The higher conversion of r-OFMSW was likely due to an increase in the activity of digestive enzymes (i.e., proteases, amylases, and lipases), as already demonstrated for BSF larvae reared on the OFMSW (Bruno et al., 2024), although we cannot rule out a possible contribution of the microorganisms present in the substrate in its degradation (Bekker et al., 2021). Similarly, larvae grown on r-OFMSW converted nitrogen with a higher efficiency (50.4 %) compared to those on s-OFMSW (33.9 %), as shown by NCE. Finally, a considerable SR (higher than 90 %) was apparent for both diets (Table 2). However, despite variations in the bioconversion rate, all parameters were comparable to, or even better than, those reported in the literature for BSF-mediated bioconversion of this type of waste (Naser El Deen et al., 2023; Surendra et al., 2020), confirming the excellent growth and bioconversion performance of the larvae on both substrates. Moreover, despite the chemical composition of insects grown on the two rearing substrates showed slight differences in the protein, lipid, and chitin content (Supplementary Table 3), altogether these data demonstrate that the s-OFMSW well mimics the r-OFMSW.

Table 1
Chemical composition and moisture content of s-OFMSW and r-OFMSW. Values (g per 100 g of diet) are expressed on "as fed" and "dry matter" bases. The former is computed on the diet as fed to the larvae (taking into consideration the water content), the latter refers to a moisture-free basis. Values of s-OFMSW were obtained by Bruno et al. (2024).

	s-OFMSW		r-OFMSW	
Component	**As fed**	**Dry matter (%)**	**As fed**	**Dry matter (%)**
Crude proteins	7.9	22.1	3.0	14.6
Crude lipids	2.9	8.0	2.3	11.4
Crude fibre[a]	0.1	0.4	1.3	0.2
Nitrogen-free extract[b]	23.6	66.1	14.0	69.1
Ash	1.2	3.4	1.0	4.7
Starch	14.8	41.5	5.6	27.9
Glucose and fructose	2.7	7.4	2.9	14.5
Water content	64.3	–	78.4	–

[a] Includes most cellulose and insoluble lignin. [b]Includes sugars, organic acids, pectins, soluble lignin, hemicellulose, and a small percentage of cellulose.

Table 2
Developmental parameters and bioconversion efficiency of BSF larvae reared on s-OFMSW and r-OFMSW. The values are reported as mean ± SEM of at least 3 different experiments. Different letters indicate statistically significant differences between the two substrates (unpaired Student's t-test: $p < 0.05$). Values of s-OFMSW were obtained by Bruno et al. (2024). RGR: Relative Growth Rate; WRI: Waste Reduction Index; ECI: Efficiency of Conversion of Ingested Food; NCE: Nitrogen Conversion Efficiency; SR: Survival Rate.

Parameter	s-OFMSW	r-OFMSW
Larval period (days)	19.0 ± 0.6^a	16.3 ± 0.3^b
Maximum weight (mg)	260.4 ± 8.4^a	265.3 ± 5.8^a
RGR (mg mg^{-1} day^{-1})	0.07652 ± 0.00001^a	0.08927 ± 0.00002^b
Substrate reduction (%)	83.5 ± 1.6^a	73.3 ± 1.7^b
WRI (%)	6.4 ± 0.1^a	6.7 ± 0.2^a
ECI (%)	20.8 ± 0.5^a	24.1 ± 0.9^b
NCE (%)	33.9 ± 1.1^a	50.4 ± 1.3^b
SR (%)	91.1 ± 0.7^a	97.1 ± 1.7^b

3.2. Protein, lipid, and chitin extraction, and characterization of the different components

The extraction of the different biological components from larvae and pupae grown on s-OFMSW or r-OFMSW was carried out using methods aimed at maximising the purity of the protein fraction (Fig. 1, sample "PROTEINS").

For this reason, two alternative procedures for lipid extraction from samples at both developmental stages were compared: a two-step batch and a Soxhlet extraction procedure. Starting from 20 g of insect ground sample, the batch procedure required a total of 3 h and 80 mL of petroleum ether instead of 16 h ($\geq$ 16 cycles) and ~ 300 mL necessary for the Soxhlet procedure; a ~ 80 % purity degree was obtained with the batch procedure compared to Soxhlet. Notably, 70–80 % of petroleum ether was recovered. Since the obtainment of protein extracts with high purity required to minimize the amount of residual fats, the Soxhlet procedure was selected. It is worthy to note that for uses requiring protein samples at a lower degree of purity, the number of Soxhlet cycles can be decreased (e.g., after 6–8 cycles, $\geq$ 95 % of the total proteins are recovered with a 80 % purity) or the batch procedure can be taken into consideration.

Recovered lipids represented about ~ 42 % of the dry starting material for insects reared on s-OFMSW and ~ 30 % for those on r-OFMSW (Table 3), with no significant difference in lipid content between larvae and pupae reared on the same substrate. These values are higher than the average value (27 ± 11) reported from a meta-analysis of the current literature (Eriksen, 2022). Proteins were recovered by precipitation at pH of minimum solubility, as previously calculated (Smets et al., 2020): r-OFMSW larvae provided the highest amount of protein extract (16.4 %), which was 30 % more than all the other samples. A higher chitin content (~4.2 vs ~ 2.9 %) was observed in pupae compared to larvae

Table 3
Extraction yields of the main BSF components. The results are expressed as the percentage of mass ratio on the starting dried sample (20 g). Values represent the mean ± SD of five replicates.

Component	s-OFMSW		r-OFMSW	
	Larvae (%)	Pupae (%)	Larvae (%)	Pupae (%)
Lipids	42.6 ± 2.9	41.9 ± 1.6	30.3 ± 3.2	30.7 ± 2.2
Protein	12.9 ± 1.0	12.9 ± 2.1	16.4 ± 3.5	12.6 ± 1.9
Defatted sample[a]	23.5 ± 2.7	36.1 ± 3.4	33.9 ± 1.0	38.0 ± 5.4
Chitin	2.9 ± 0.2	4.2 ± 0.2	5.1 ± 0.2	4.6 ± 0.1
Ash	3.4 ± 0.1	2.7 ± 0.1	5.6 ± 0.1	6.8 ± 0.3
Soluble carbohydrates[b]	1.9 ± 0.2	1.7 ± 0.8	1.3 ± 0.1	0.55 ± 0.06
Total carbohydrates[b]	2.9 ± 0.1	3.2 ± 0.1	1.8 ± 0.2	0.66 ± 0.02

[a] The dried solid pellet after removal of the fraction of soluble proteins, used for chitin extraction.
[b] Carbohydrates were determined as glucose, therefore chitin and N-acetyl-glucosamine were not included.

reared on s-OFMSW, while similar values were measured for pupae and larvae reared on r-OFMSW (4.6–5.1 %) (Table 3). The use of deep eutectic solvents, alone or coupled to physical pre-treatments, was also investigated and resulted in a ~ 3-fold lower protein recovery (data not shown).

In detail, from 1 kg of s-OFMSW a total of 73.4 g of larvae were produced, from which 9.5 g of proteins, 31.3 g of lipids, and 2.1 g of chitin were isolated; under the same conditions, 48.7 g of pupae were produced, from which 6.3 g of proteins, 20.4 g of lipids, and 2.0 g of chitin were isolated.

3.2.1. Analysis of the lipid fraction

The ten most abundant lipids in larvae and pupae reared on s-OFMSW (the condition yielding the highest lipid accumulation) detected through ESI-MS are reported in Table 4. No free C12-C18 fatty acids (FA) were present in larvae while they are present in pupae where vaccenic acid, an ω-7 FA, was the most abundant component. On the contrary, saturated triacylglycerols (TGs) were the most represented components in lipids from larvae and their amount was higher than in pupae. The same trend was recorded for mono- and poly-unsaturated lipids. These data are in agreement with the physiological processes occurring during metamorphosis in holometabolous insects. Indeed, larvae need to save lipids as energy reserve to sustain metabolic functions during pupal stage, in which the insect does not eat and organs and tissue are extensively remodelled or even completely rebuilt (Rolff et al., 2019). As a consequence, the insect developmental stage affects lipid composition: TGs, which are lipids related to storage, are the main components in larvae, while rapidly metabolically usable fatty acids are mainly present in pupae. Although we initially envisioned that BSF lipids could primarily be used for biodiesel production, the analysis of the main components of the lipid fractions prompts us to focus on alternative high-value uses, including applications for the pharmaceutical and cosmetics sectors (Tettamanti and Bruno, 2024).

Table 4
Lipidomic analysis of larvae and pupae reared on s-OFMSW. The list of the ten most represented molecules is reported. Values are reported as a percentage of the total lipid content. FA: fatty acid, TG: triacylglycerol.

	Larvae	Pupae
Lipid	**% on total lipids**	
FA 12:0 Lauric acid Dodecanoic acid		5.38
FA 16:0 Palmitic acid Hexadecanoic acid		5.00
FA 18:2 Linoleic acid (ω 6)		8.49
FA 18:1 Vaccenic acid (ω 7)		13.35
TG 34:0|TG 10:0_12:0_12:0	4.41	3.18
TG 36:0|TG 10:0_12:0_14:0	13.83	11.38
TG 38:0|TG 12:0_12:0_14:0	8.59	6.09
TG 42:2|TG 12:0_12:0_18:2	3.40	3.64
TG 42:1|TG 12:0_12:0_18:1	3.57	3.23
TG 42:0|TG 12:0_14:0_16:0	3.54	
TG 44:1|TG 12:0_14:0_18:1	3.42	
TG 46:2|TG 12:0_16:0_18:2	4.57	2.60
TG 48:2|TG 12:0_18:1_18:1	3.10	
TG 48:1|TG 14:0_16:0_18:1	3.28	

3.2.2. Analysis of the protein fraction

The analysis of the purified protein fraction (Fig. 1, sample "PROTEINS" dissolved at pH 11) for both larvae and pupae indicated a protein purity > 70 % (Fig. 2A), with extracts from insects reared on r-OFMSW possessing a lower degree of homogeneity, especially for pupae (Supplementary Table 4). The presence of a residual content of carbohydrates in the protein extracts, between 2–6 %, was apparent, with a higher amount in larval extracts. When the insoluble components were separated by centrifugation prior to the analysis, the carbohydrate content of the larvae decreased, reaching a pattern similar to that of pupae (~3%). The fraction of soluble proteins at pH 11 was > 80 %. The larvae protein suspension showed higher absorbance intensity in the UV–visible spectra (Fig. 2B), being also visibly more turbid (Fig. 2C, left). After centrifugation, a higher amount of precipitate was visible for larval protein suspensions (Fig. 2C, right), and absorbance spectra of the supernatant for larval protein suspensions became comparable to those from pupae. No nucleic acids were detected (Supplementary Table 4).

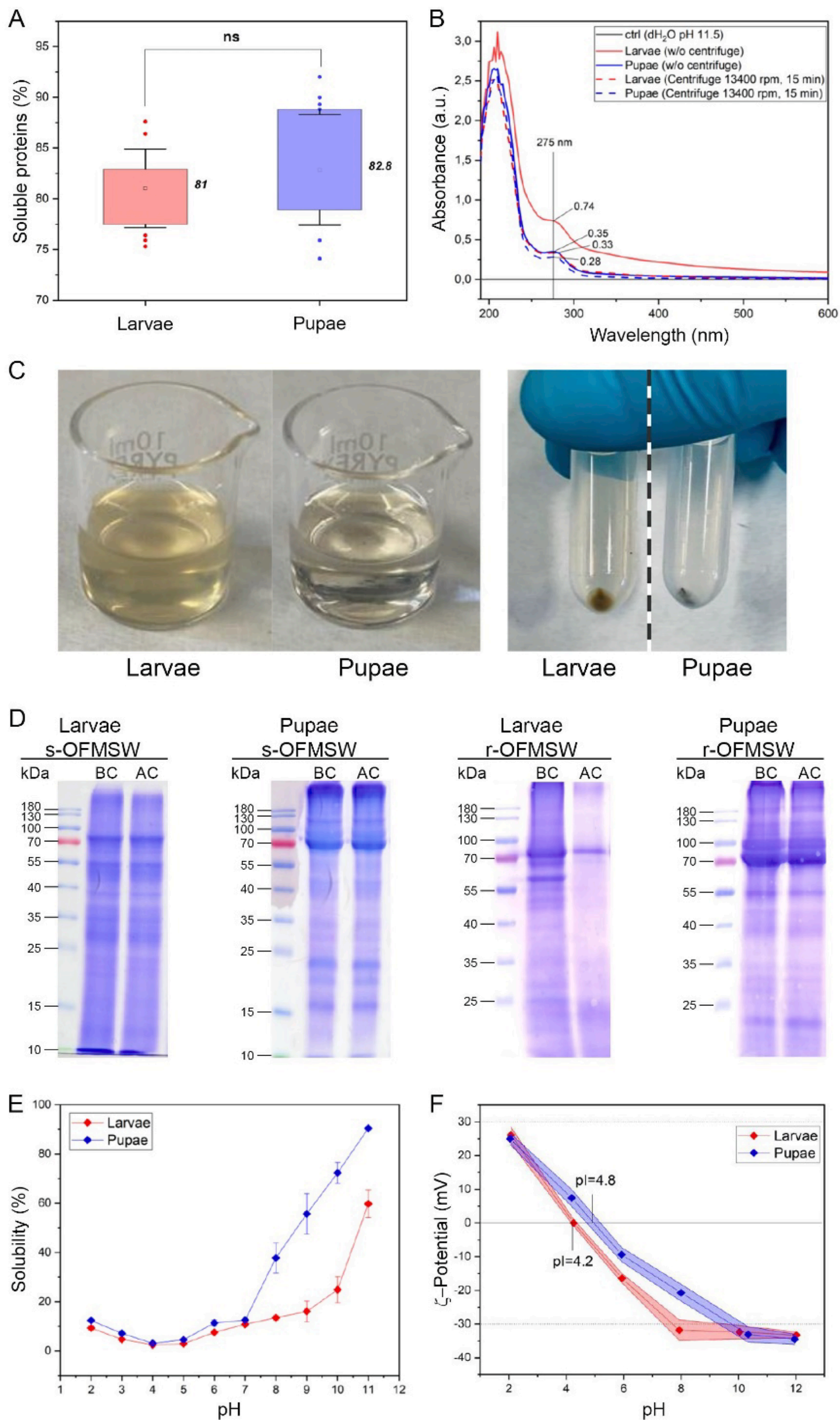
SDS-PAGE analysis showed marked differences in the band pattern between the two developmental stages, with proteins extracted from larvae more enriched in the 25 to 75 kDa region, and pupae extracts showing two main protein bands at ≈75 kDa and above 180 kDa, and three more bands in the 15–25 kDa region (Fig. 2D). Based on the quantification of NH_2 groups through TNBSA titration assay (Supplementary Table 5), longer polypeptides were present in pupal samples. CD spectra indicated that protein extracts from larvae and pupae were rich in proteins containing β-strands and turns as secondary structure elements (Supplementary Fig. 1), with differences between samples from insects reared on the two diets.

An insight on insect proteome by nLC-MS/MS analysis evidenced differences between protein extracts of larvae and pupae reared on s-OFMSW (Supplementary Tables 6 and 7). Among the most frequent validated proteins, a prevalence of structural and muscle proteins, belonging to actin, troponin, and tropomyosin families, was present in both samples, while a few enzymes (arginine kinase − for pupae only − and ATP synthase) were identified (Supplementary Table 8).

The solubility of proteins as a function of pH is reported in Fig. 2E: the solubility was lowest at pH 4 and was maximal at pH values ≥ 11 (in agreement with the fractionation method used, see Fig. 1). These values were supported by ζ-potential results (Fig. 2F). The strict correlation between particle precipitation stability and ζ-potential is indeed well described in the literature for a multitude of nanomaterials (Genovese and Lozano, 2001; Gharehbeglou et al., 2019; López-Zamora et al., 2018; Tabernero et al., 2017). In this case, an average isoelectric point of 4.2 and 4.8 was determined for larvae and pupae extracts, respectively, whilst the highest charge (absolute value) was found at alkaline pH. In particular, larvae extracts reached a ζ-potential plateau (−30 mV) at pH 8, whilst pupae showed a ζ-potential higher than 30 mV above pH 10.

3.3. Production and characterization of protein-based bioplastics

The bioplastics herein described were produced using a simple, scalable, water-based method, as outlined in Fig. 3A and detailed in Materials and methods. BSF larvae and pupae protein extracts were dispersed in alkaline water (0.1 M NaOH, pH 11.5) alongside a plasticizer (glycerol). Following the mixing process, yellowish to brownish, homogeneous, translucent and flexible free-standing films were generated by casting the solution onto polydimethylsiloxane substrates (Fig. 3B).



(caption on next page)

Fig. 2. Characterization of protein extracts. (A) Fraction of soluble proteins in larvae and pupae protein extracts, as determined by the Bradford assay. (B) Absorbance spectra of BSF larvae (red) and pupae (blue) protein extract suspensions before (continuous line) and after (dashed line) centrifugation at 13400 rpm for 15 min. (C) BSF larvae and pupae protein extracts water dispersed at pH 11.5 (left). Protein pellet after centrifugation at 13400 rpm for 15 min (right). (D) SDS-PAGE analysis of the resuspended protein extracts from larvae and pupae reared on r- and s-OFMSW. BC: before centrifugation; AC: after centrifugation. (E) Solubility of larvae (red) and pupae (blue) protein extract solutions at different pH values. (F) ζ-potential values for larvae (red) and pupae (blue) protein extract solutions at different pH. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article).

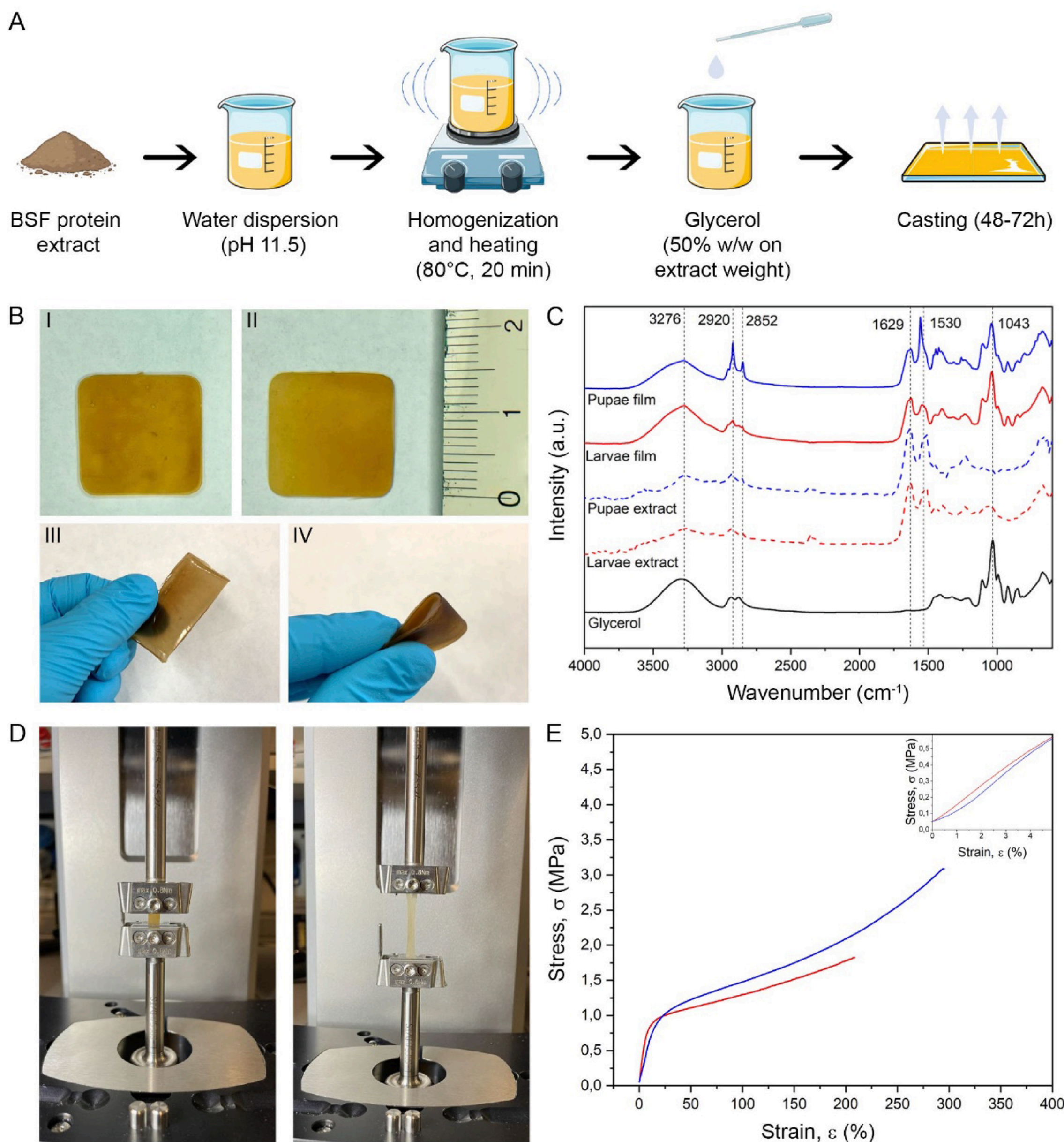


Fig. 3. Production and characterization of bioplastics from BSF larvae and pupae protein extracts. (A) Schematic process for the production of BSF protein-based bioplastics. (B) BSF protein-based bioplastics from larvae (I, III and IV) and pupae (II) extracts. (C) ATR-FTIR spectra of the protein extracts (larvae: red dashed line, pupae: blue dashed line), bioplastic films (larvae: red line, pupae: blue line) and glycerol (black line). (D) BSF pupae protein-derived film unloaded (left) and at maximum strain (right). (E) Stress–strain curves for bioplastics from BSF larvae (red line) and pupae (blue line) reared on the s-OFMSW. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article).

To investigate the presence of different chemical structures in films obtained from larvae and pupae, ATR-FTIR analysis was used (Fig. 3C). The analysis revealed, in both types of bioplastics, the protein characteristic Amide I (1700–1600 cm^{-1}) and Amide II (1600–1500 cm^{-1}) regions, while the strong signal at 1043 cm^{-1} was ascribed to –CO stretching of glycerol molecules (see Supplementary Text 2 for a broader outlook on the remaining modes and associated surface chemical groups). A focus on the Amide I region (Supplementary Fig. 2) revealed similar secondary structure profiles for the two groups of bioplastics, with a significant fraction of β-sheet (native (1690 and 1640 cm^{-1}) and aggregated (1624–1620 cm^{-1}) secondary structures (Supplementary Table 9), confirming the results gathered on the solubilized extracts (Supplementary Fig. 1).

Overall, these results suggest a similarity in the chemical profile of

proteins from extracts of larvae and pupae, as well as in the bioplastic films derived from them.

Mechanical tests showed that both materials had a rubber-like modulus and high deformation at break (Fig. 3D). However, films prepared from pupae protein extracts showed higher strain (ε_{break}) and stress at break (σ_{break}) compared to films from larvae (Fig. 3E), regardless of the rearing substrate (s-OFMSW or r-OFMSW) (Supplementary Fig. 3, Supplementary Table 10). We hypothesize that this different behaviour can be ascribed to the higher molecular mass of proteins from pupae extracts (Fig. 2D, Supplementary Table 5). Films' mechanical properties showed to be highly influenced by the conditioning method (Supplementary Fig. 4): for environments characterized by high relative humidity (e.g., 75 %), films displayed higher ε_{break} and lower σ_{break}. On the contrary, mimicking real conditions, after 8 days of exposure in open air (20 ± 2 °C, 50 ± 10 % R.H.) films displayed the highest values of σ_{break} and the lowest values of ε_{break}. Notably, when compared to BSF protein-based bioplastics reported in the literature (Supplementary Table 11), the pupae protein-based films herein produced displayed higher ε_{break} in each case (Supplementary Figure 5A), and comparable Young's modulus (E) and σ_{break} (Supplementary Figure 5B).

Regardless of the insect life stage, the obtained films possessed good chemical resistance and were insoluble in all tested organic solvents after 1 week (Supplementary Table 12). However, one of the major challenges associated with bioplastics (protein-based ones among them) is their inherently low water stability (Yashwant et al., 2023). In this regard, films dissolved in strong acidic (pH < 2) or alkaline (pH > 10) water solutions (Supplementary Text 2, Supplementary Figure 6, Supplementary Table 12). In neutral water, glycerol was rapidly released and high-water absorption was observed (Supplementary Figures 7 and 8), but films were macroscopically intact even after soaking for 6 days. Water uptake and volumetric variation after 24 h were higher for pupae bioplastic films, weight loss was higher for larvae films (44 % vs. 38 % in pupae) (Supplementary Table 13), and glycerol release was also faster in larvae-derived materials (25 % on dry film weight after 1 h, vs. 19 % for pupae) (Supplementary Table 14). For a detailed description of the methods behind swelling tests, quantification of water interaction parameters and glycerol release see Supplementary text 3, 4 and 5, respectively.

These results highlight the hydrophilic nature of BSF protein-based films. Notably, the macroscopic structure of each film was retained and none of them dissolved after prolonged immersion in water. This result suggests the formation of a robust protein network with water-stable interconnected structures. This network may derive from the formation of intermolecular β-sheets, as suggested by their high relative content (Supplementary Table 9). With these properties in mind, BSF protein-based bioplastics may be utilized in low-humidity contexts, as those found in printed and flexible electronics, where BSF proteins can be utilized as substrate materials or matrices for composites. Likewise, they can be used where long-term stability against environmental factors is not required, as in disposable packaging products, by exploiting the characteristic oxygen barrier properties of proteins (Purewal et al., 2024).

3.4. Life cycle assessment of the insect value chain

3.4.1. Normalization

Results of the normalization procedure are reported in Fig. 4A. The model suggested that non-renewable energy consumption, global warming potential (GWP), respiratory inorganics, and terrestrial ecotoxicity were mainly associated with the defatting procedure (Fig. 4B). This was due to energy consumption and depletion of fossil solvents like petroleum ether (modelled as Naphtha). Therefore, it seems that lipids chemical separation represents the most important process to be considered for mitigating environmental impacts.

For this reason, the search for alternative green solvents and/or solutions that allow the recovery of chemical reagents and, therefore, their

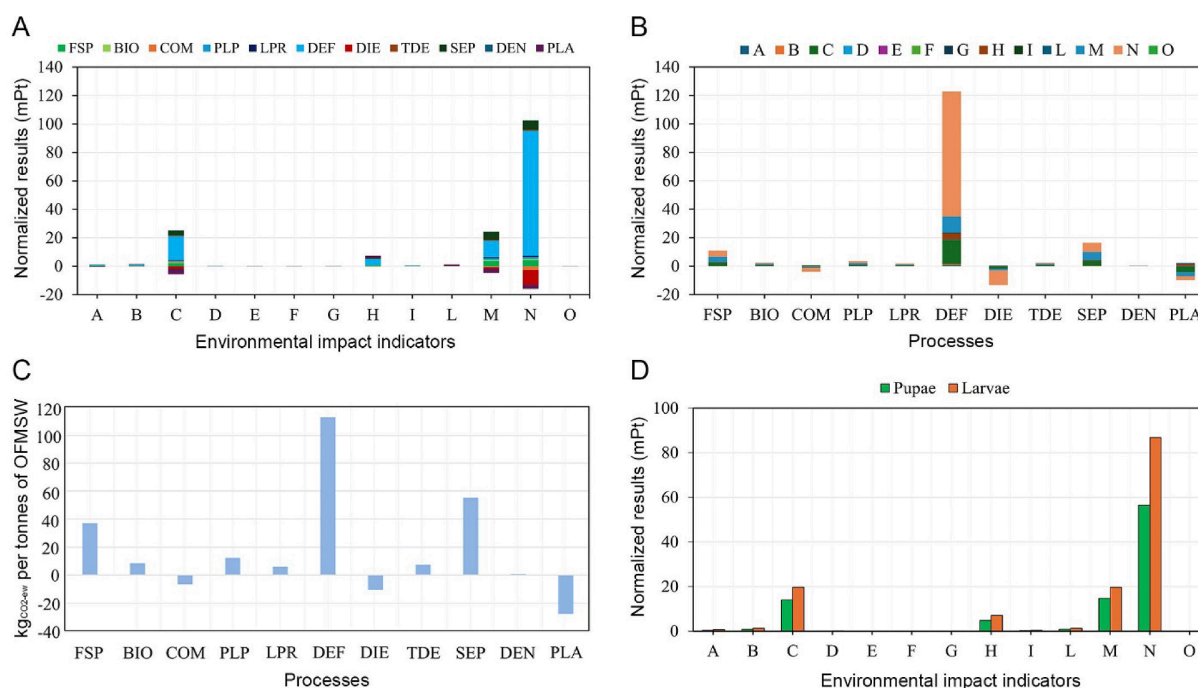


Fig. 4. Environmental profile of the OFMSW treatment by BSF larvae. Normalization of the results per (A) environmental impact indicator, and per (B) process. (C) GWP of the system and (D) comparison of the environmental impact indicators in scenarios with pupae or larvae. FSP: Feedstock pre-treatment; BIO: OFMSW bioconversion; COM: Composting; PLP: Insect growing and production; LPR: Larvae preparation for post treatment; DEF: Defatting; DIE: Biodiesel production; TDE: Treatment of defatted products; SEP: Protein separation and lyophilization; DEN: Protein denaturation; PLA: Bioplastics production. A: Carcinogens; B: Non-carcinogens; C: Respiratory inorganics; D: Ionizing radiation; E: Ozone layer depletion; F: Respiratory organics; G: Aquatic ecotoxicity; H: Terrestrial ecotoxicity; I: Terrestrial acid/nutri; L: Land occupation; M: Global warming; N: Non-renewable energy; O: Mineral extraction. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article).

possible reuse are needed for scaling up the application. One of the main limitations of this analysis is related to the scale-up of the inventory, necessary because the industrial scale of the value chain is not yet available. Scale-up factors were added to the inventory, but this procedure can be error-prone and should be carefully assessed in future studies.

3.4.2. Contribution analysis: Focus on the global warming potential (GWP)

Quantitative results related to the GWP are reported in Fig. 4C. On balance, the system potentially contributed to generate a net GWP of almost 195 $kgCO_{2\text{-eq}}$ per ton of OFMSW, including avoided impacts. Only the defatting procedure contributed to about 57 % of the total GWP due to petroleum ether consumption. Electricity consumption was the second parameter to be considered, which contributed to the impacts during the feedstock pre-treatment and protein lyophilization. The OFMSW treatment and larvae rearing contributed with about 57.7 $kgCO_{2\text{-eq}}$ per ton of OFMSW, in line with the literature, although affected by uncertainty (Salomone et al., 2017). This represented about 30 % of the total GWP. Therefore, protein extraction for bioplastics production seems to increase the impacts of the OFMSW treatment process, while avoided impacts due to biodiesel and bioplastics production does not appear to compensate for the comprehensive impacts. Compared to conventional treatment systems, OFMSW treatment with BSF larvae seems to be an interesting option. However, the production of bioplastics from larvae does not allow to increase the environmental benefits compared to fishmeal production (Mondello et al., 2017).

3.4.3. Comparison between larvae and pupae

Different environmental profiles were obtained considering the larvae or the pupae as the input biomass of the processes (Fig. 4D). The analysis showed that pupae allowed decreasing the environmental impacts mainly due to the smaller amount of dried powder and therefore of chemical reagents needed for bioplastic and biodiesel production. On average, the environmental impacts for pupae were reduced by about 32.4 % (from 137.4 mPt with larvae to 92.8 mPt with pupae).

3.5. Socioeconomic assessment of the insect value chain

Despite the emergence of a quite recent and growing patenting trend concerning innovations in waste bioconversion processes using BSF larvae, PLA provided evidence of a substantial technological freedom to operate, finding no significant patent infringement risk, especially for what concerns the phases (ii) and (iii) of the value chain (see Section 2.6) (Supplementary Figure 9).

As for the analysis of costs, unitary costs must be considered as experimental, based on a huge desk-based research activity (Supplementary Tables 15 and 16). Only variable costs were considered on the basis of output quantities obtained on a very small and experimental scale. However, according to Pahmeyer et al. (2022), when scaled-up, the productivity of the inputs increases significantly, because of scale economies, and fixed costs can be amortized in a few years. Moreover, as the quantity of produced biodiesel does not seem suitable for sale because of the small amount, the possibility of using it as an energy source within the production process could be investigated in further research. Finally, value-added chitins from BSF were produced, as an alternative to other available chitin sources (Soetemans et al., 2020).

Concerning market opportunities, the bioplastic films produced from BSF proteins revealed a high quality and higher performance than other protein-based plastic films (Nuvoli et al., 2021). The production process of proteins was the outcome of a controlled mix of biobased elements which have been tested (s-OFMSW and r-OFMSW) and of a process guaranteeing constant performance of the biofilm. In a scale-up perspective, since the production of bioplastics directly starting from a variety of categories of organic wastes shows a high variability because of the different organic and chemical qualities of each material (Otoni et al., 2021), this process valorizes OFMSW in a circular bioeconomy and ecological transition framework and the final high and constant quality biofilm is expected to have a high market value. One of the uses, for instance, might be in the electronic device sector as a basis for biodegradable printed circuit boards, other components and, possibly, 3D printing (Luoma et al., 2022).

As to opportunity costs, a first element is related to the use of OFMSW for rearing BSF larvae instead of alternative uses like composting, and a second one to the production of bioplastics from BSF larvae/pupae instead of alternative types of biomasses (e.g., crops). Composting requires that OFMSW must be pre-processed to remove any impurity (according to the Italian law on biomasses) (Decreto Legislativo 75, 2010) and then delivered to large composting facilities. This involves non-negligible pre-processing and transport costs that the present process reduces (Montresori, 2022). The process allows the reduction of purification costs: as larvae spontaneously separate indigestible waste from digestible one, OFMSW pre-processing costs are lower than for composting. Health and environmental benefits can also rise from the reduction of pathogens in the insect-processed proteins (Chia et al., 2020) compared to those reported in OFMSW and, therefore, in compost. Furthermore, processing facilities can be smaller than composting ones and might be located near the OFMSW collection sites (Bruni et al., 2020), with a reduction of transport costs and pollution externalities. As to the second opportunity cost concerning bioplastic production, the reduced use of biomasses from the agri-food sector (that, differently from OFMSW, can also serve for human nutrition) might have an important benefit for food security policies (Food Systems Countdown Initiative, 2023).

4. Conclusions

The present study represents a proof of concept of a circular supply chain that prioritizes waste reduction and resource recovery, based on the valorization of the OFMSW through BSF larvae. This approach leverages the larvae feeding capabilities to manage waste in a sustainable manner and produce valuable insect-based bioproducts for various industrial uses. BSF larvae fed on OFMSW yielded high-purity biomolecules, which can be utilized in the frame of circular (bio)economy processes, such as the production of biodiesel from lipids, bioplastics from proteins, and food additives from chitin. LCA and socioeconomic analyses revealed that this application offers benefits beyond traditional waste uses like composting, including the production of high-quality and high-performance materials that can replace oil-based ones, such as bioplastic films with enhanced properties for flexible electronic devices, or high oxygen barrier packaging.

In this regard, bioplastics herein presented possess unprecedented stretchability when compared to current BSF protein-based bioplastics (Barbi et al, 2019; Barbi et al, 2021; Nuvoli et al, 2021). By adding active ingredients, blending with other polymers, or performing post-treatments, common issues related to protein-based materials (i.e., mechanical performance and low water stability) can be addressed, and their stability and properties further improved and expanded.

Scale-up opportunities for the herein reported process are feasible in a medium-term perspective, supported by sustainable finance and transition finance tools, highlighting the potential of these production methods in ecological transitions.

CRediT authorship contribution statement

Daniele Bruno: Writing – review & editing, Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Marco Orlando:** Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Edoardo Testa:** Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Marco Carnevale Miino:** Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Giulia Pesaro:** Writing – original draft,

Methodology, Data curation, Conceptualization. **Matteo Miceli:** Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Loredano Pollegioni:** Writing – review & editing, Writing – original draft, Conceptualization. **Vincenzina Barbera:** Investigation, Formal analysis. **Elisa Fasoli:** Investigation, Formal analysis. **Lorenza Draghi:** Investigation, Formal analysis. **Alberto Pietro Damiano Baltrocchi:** Writing – original draft, Software. **Navarro Ferronato:** Writing – original draft, Software, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Raffaello Seri:** Writing – original draft, Conceptualization. **Elena Maggi:** Writing – original draft, Conceptualization. **Silvia Caccia:** Writing – original draft, Data curation. **Morena Casartelli:** Writing – review & editing, Writing – original draft, Supervision, Conceptualization. **Gianluca Molla:** Writing – review & editing, Writing – original draft, Supervision, Project administration, Funding acquisition, Conceptualization. **Maurizio Stefano Galimberti:** Writing – original draft, Supervision, Conceptualization. **Vincenzo Torretta:** Writing – original draft, Supervision, Conceptualization. **Andrea Vezzulli:** Writing – original draft, Conceptualization. **Gianluca Tettamanti:** Writing – review & editing, Writing – original draft, Supervision, Project administration, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Maurizio Stefano Galimberti, Vincenzina Barbera, Edoardo Testa, Elisa Fasoli, Gianluca Tettamanti, Daniele Bruno, Gianluca Molla, Marco Orlando, Loredano Pollegioni, and Morena Casartelli have patent BIO-NANOCOMPOSITE MATERIAL issued to 102022000019020. If there are other authors, they declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.wasman.2024.10.030.

Data availability

Data will be made available on request.

References

Adedara, M.L., Taiwo, R., Bork, H.-R., 2023. Municipal solid waste collection and coverage rates in Sub-Saharan African countries: A comprehensive systematic review and meta-analysis. Waste 1, 389–413. https://doi.org/10.3390/waste1020024.

Aleisa, E., Heijungs, R., 2022. Leveraging life cycle assessment to better promote the circular economy: A first step using the concept of opportunity cost. Sustainability 14, 3451. https://doi.org/10.3390/su14063451.

Athanassiou, C.G., Coudron, C.L., Deruytter, D., Rumbos, C.I., Gasco, L., Gai, F., Sandrock, C., De Smet, J., Tettamanti, G., Francis, A., Petrusan, J., Smetana, S., 2024. A decade of advances in black soldier fly research: from genomics to sustainability. J. Insects Food Feed. https://doi.org/10.1163/23524588-00001122.

Barbi, S., Messori, M., Manfredini, T., Pini, M., Montorsi, M., 2019. Rational design and characterization of bioplastics from *Hermetia illucens* prepupae proteins. Biopolymers 110, e23250.

Barbi, S., Macavei, L.I., Caligiani, A., Maistrello, L., Montorsi, M., 2021. From food processing leftovers to bioplastic: A design of experiments approach in a circular economy perspective. Waste Biomass. Valor. 12, 5121–5130. https://doi.org/10.1007/s12649-021-01376-3.

Bekker, N.S., Heidelbach, S., Vestergaard, S.Z., Nielsen, M.E., Riisgaard-Jensen, M., Zeuner, E.J., Bahrndorff, S., Eriksen, N.T., 2021. Impact of substrate moisture content on growth and metabolic performance of black soldier fly larvae. Waste Manag. 127, 73–79. https://doi.org/10.1016/j.wasman.2021.04.028.

Bonelli, M., Bruno, D., Brilli, M., Gianfranceschi, N., Tian, L., Tettamanti, G., Caccia, S., Casartelli, M., 2020. Black soldier fly larvae adapt to different food substrates through morphological and functional responses of the midgut. Int. J. Mol. Sci. 21, 4955. https://doi.org/10.3390/ijms21144955.

Boreggio, M., Rosini, E., Gambarotti, C., Pollegioni, L., Fasoli, E., 2022. Unveiling the bio-corona fingerprinting of potential anticancer carbon nanotubes coupled with D-amino acid oxidase. Mol. Biotechnol. 64, 1164–1176. https://doi.org/10.1007/s12033-022-00488-y.

Broeckx, L., Frooninckx, L., Berrens, S., Noyens, I., Goossens, G., Verheyen, G., Wuyts, A., Van Miert, S., 2021. Growth of black soldier fly larvae reared on organic side-streams. Sustainability 13, 12953. https://doi.org/10.3390/su132312953.

Bruni, C., Akyol, Ç., Cipolletta, G., Eusebi, A.L., Caniani, D., Masi, S., Colón, J., Fatone, F., 2020. Decentralized community composting: Past, present and future aspects of Italy. Sustainability 12, 3319. https://doi.org/10.3390/su12083319.

Bruno, D., Bonelli, M., Cadamuro, A.G., Reguzzoni, M., Grimaldi, A., Casartelli, M., Tettamanti, G., 2019a. The digestive system of the adult *Hermetia illucens* (Diptera: Stratiomyidae): Morphological features and functional properties. Cell Tissue Res. 378, 221–238. https://doi.org/10.1007/s00441-019-03025-7.

Bruno, D., Bonelli, M., De Filippis, F., Di Lelio, I., Tettamanti, G., Casartelli, M., Ercolini, D., Caccia, S., 2019b. The intestinal microbiota of *Hermetia illucens* larvae is affected by diet and shows a diverse composition in the different midgut regions. Appl. Environ. Microbiol. 85, e01864–e10918. https://doi.org/10.1128/AEM.01864-18.

Bruno, D., Bonelli, M., Valoroso, M.C., Roma, D., Montali, A., Pellegrino, M.G., Marzari, M., Caccia, S., Tettamanti, G., Casartelli, M., 2024. Black soldier fly larvae efficiently bioconvert the organic fraction of municipal solid waste thanks to their midgut functional plasticity. J. Insects Food Feed. https://doi.org/10.1163/23524588-00001193.

Caldinelli, L., Molla, G., Bracci, L., Lelli, B., Pileri, S., Cappelletti, P., Sacchi, S., Pollegioni, L., 2010. Effect of ligand binding on human D-amino acid oxidase: implications for the development of new drugs for schizophrenia treatment. Protein Sci. 19, 1500–1512. https://doi.org/10.1002/pro.429.

Caligiani, A., Marseglia, A., Leni, G., Baldassarre, S., Maistrello, L., Dossena, A., Sforza, S., 2018. Composition of black soldier fly prepupae and systematic approaches for extraction and fractionation of proteins, lipids and chitin. Int. Food Res. 105, 812–820. https://doi.org/10.1016/j.foodres.2017.12.012.

Ceccotti, C., Bruno, D., Tettamanti, G., Branduardi, P., Bertacchi, S., Labra, M., Rimoldi, S., Terova, G., 2022. New value from food and industrial wastes – Bioaccumulation of omega-3 fatty acids from an oleaginous microbial biomass paired with a brewery by-product using black soldier fly (*Hermetia illucens*) larvae. Waste Manag. 143, 95–104. https://doi.org/10.1016/j.wasman.2022.02.029.

Chia, S.Y., Tanga, C.M., Osuga, I.M., Cheseto, X., Ekesi, S., Dicke, M., van Loon, J.J.A., 2020. Nutritional composition of black soldier fly larvae feeding on agro-industrial by-products. Entomol. Exp. Appl. 168, 472–481. https://doi.org/10.1111/eea.12940.

Commission, E., 2017. Commission regulation (EU) 2017/893 of 24 May 2017 amending Annexes I and IV to regulation (EC) no 999/2001 of the European Parliament and of the Council and Annexes X, XIV and XV to commission regulation (EU) no 142/2011 as regards the provisions on processed animal protein. Off. J. Eur. Union L138, 92–116. http://data.europa.eu/eli/reg/2017/893/oj.

Dantas, T.E.T., de-Souza, E.D., Destro, I.R., Hammes, G., Rodriguez, C.M.T., Soares, S.R., 2021. How the combination of circular economy and industry 4.0 can contribute towards achieving the sustainable development goals. Sustain. Prod. Consump. 26, 213–227. https://doi.org/10.1016/j.spc.2020.10.005.

Decreto Legislativo 75, 2010. Riordino e revisione della disciplina in materia di fertilizzanti, a norma dell'articolo 13 della legge 7 luglio 2009, n. 88. Gazzetta Ufficiale, 121. https://www.parlamento.it/parlam/leggi/deleghe/10075dl.htm.

De Filippis, F., Sequino, G., Bruno, D., Bonelli, M., Nolasco, A., Esposito, F., Cirillo, T., Tettamanti, G., Ercolini, D., Casartelli, M., Caccia, S., 2024. A comprehensive analysis of coffee silverskin bioconversion by Hermetia illucens larvae. J. Insect Food Feed. https://doi.org/10.1163/23524588-00001241.

Eriksen, N.T., 2022. Dynamic modelling of feed assimilation, growth, lipid accumulation, and CO2 production in black soldier fly larvae. PLoS ONEne 17, e0276605.

Ford, H.V., Jones, N.H., Davies, A.J., Godley, B.J., Jambeck, J.R., Napper, I.E., Suckling, C.C., Williams, G.J., Woodall, L.C., Koldewey, H.J., 2022. The fundamental links between climate change and marine plastic pollution. Sci. Total Environ. 806, 150392. https://doi.org/10.1016/j.scitotenv.2021.150392.

Garcia, J.M., Robertson, M.L., 2017. The future of plastics recycling. Science 358, 870–872. https://doi.org/10.1126/science.aaq0324.

Genovese, D.B., Lozano, J.E., 2001. The effect of hydrocolloids on the stability and viscosity of cloudy apple juices. Food Hydrocoll. 15, 1–7. https://doi.org/10.1016/S0268-005X(00)00053-9.

Gharehbeglou, P., Jafari, S.M., Hamishekar, H., Homayouni, A., Mirzaei, H., 2019. Pectin-whey protein complexes vs. small molecule surfactants for stabilization of double nano-emulsions as novel bioactive delivery systems. J. Food Eng. 245, 139–148. https://doi.org/10.1016/j.jfoodeng.2018.10.016.

Hahn, T., Tafi, E., von Seggern, N., Falabella, P., Salvia, R., Thomä, J., Febel, E., Fijalkowska, M., Schmitt, E., Stegbauer, L., Zibek, S., 2022. Purification of chitin from pupal exuviae of the black soldier fly. Waste Biomass Valor. 13, 1993–2008. https://doi.org/10.1007/s12649-021-01645-1.

Horwitz, W., 2000. AOAC International official methods of analysis of AOAC International. The association of official analytical chemists, Gaithersburg, MD, USA. Methods 925.10, 65.17, 974.24, 992.16.
Hu, C., Yang, L., Wang, H., Xiao, X., Wang, Z., Gong, X., Liu, X., Li, W., 2023. Analysis of heavy metals in the conversion of lake sediment and restaurant waste by black soldier fly (*Hermetia illucens*). Front. Bioeng. Biotechnol. 11, 1163057. https://doi.org/10.3389/fbioe.2023.1163057.
Food Systems Countdown Initiative, 2023. The food systems countdown report 2023: The state of food systems worldwide, New York: Columbia University; Ithaca: Cornell University; Rome: Food and Agriculture Organization of the United Nations (FAO); Geneva: Global Alliance for Improved Nutrition (GAIN). Doi: 10.36072/fsci2023.
Janssen, R.H., Vincken, J.-P., van den Broek, L.A.M., Fogliano, V., Lakemond, C.M.M., 2017. Nitrogen-to-Protein conversion factors for three edible insects: *Tenebrio molitor*, *Alphitobius diaperinus*, and *Hermetia illucens*. J. Agric. Food Chem. 65, 2275–2278. https://doi.org/10.1021/acs.jafc.7b00471.
Kamada, A., Rodriguez-Garcia, M., Ruggeri, F.S., Shen, Y., Levin, A., Knowles, T.P.J., 2021. Controlled self-assembly of plant proteins into high-performance multifunctional nanostructured films. Nat. Commun. 12, 3529. https://doi.org/10.1038/s41467-021-23813-6.
Latimer, G.W., 2016. AOAC International official methods of analysis of AOAC International. The association of official analytical chemists, Gaithersburg, MD, USA, p. 3172.
Li, T., Zhou, J., Peydayesh, M., Yao, Y., Bagnani, M., Kutzli, I., Chen, Z., Wang, L., Mezzenga, R., 2023. Plant protein amyloid fibrils for multifunctional sustainable materials. Adv. Sustainable Syst. 7, 2200414. https://doi.org/10.1002/adsu.202200414.
López-Zamora, L., Martínez-Martínez, H.N., González-Calderón, J.A., 2018. Improvement of the colloidal stability of titanium dioxide particles in water through silicon based coupling agent. Mater. Chem. Phys. 217, 285–290. https://doi.org/10.1016/j.matchemphys.2018.06.063.
Luoma, E., Välimäki, M., Ollila, J., Heikkinen, K., Immonen, K., 2022. Bio-based polymeric substrates for printed hybrid electronics. Polymers (basel) 14, 1863. https://doi.org/10.3390/polym14091863.
Meijer, N., de Rijk, T., van Loon, J.J.A., Zoet, L., van der Fels-Klerx, H.J., 2021. Effects of insecticides on mortality, growth and bioaccumulation in black soldier fly (*Hermetia illucens*) larvae. PLoS One 16, e0249362.
Mondello, G., Salomone, R., Ioppolo, G., Saija, G., Sparacia, S., Lucchetti, M.C., 2017. Comparative LCA of alternative scenarios for waste treatment: The case of food waste production by the mass-retail sector. Sustainability 9, 827. https://doi.org/10.3390/su9050827.
Montresori, G., 2022. Un nuovo approccio regolatorio. Arera e Comuni: Qualità e costi all'utenza nella gestione dei rifiuti urbani. AcerQuality, Allegato redazionale al numero 3/2022 di ACER. https://www.compost.it/wp-content/uploads/2022/06/2022.03_acer_AQ.pdf.
Mshayisa, V.V., Van Wyk, J., Zozo, B., 2022. Nutritional, techno-functional and structural properties of black soldier fly (*Hermetia illucens*) larvae flours and protein concentrates. Foods 11, 724. https://doi.org/10.3390/foods11050724.
Naser El Deen, S., van Rozen, K., Elissen, H., van Wikselaar, P., Fodor, I., van der Weide, R., Hoek-van den Hil, E.F., Rezaei Far, A., Veldkamp, T., 2023. Bioconversion of different waste streams of animal and vegetal origin and manure by black soldier fly larvae *Hermetia illucens* L. (diptera: Stratiomyidae). Insects 14, 204. https://doi.org/10.3390/insects14020204.
Nuvoli, D., Montevecchi, G., Lovato, F., Masino, F., Van Der Borght, M., Messori, M., Antonelli, A., 2021. Protein films from black soldier fly (*Hermetia illucens*, Diptera: Stratiomyidae) prepupae: Effect of protein solubility and mild crosslinking. J. Sci. Food. Agric. 101, 4506–4513. https://doi.org/10.1002/jsfa.11091.
OECD - Organisation for Economic Co-operation and Development, Centre for Co-operation with European Economies in Transition, 1993. Glossary of industrial organisation economics and competition Law, OECD, ISBN 9264137939. https://search.oecd.org/daf/competition/mergers/2376087.pdf.
Otoni, C.G., Azeredo, H.M.C., Mattos, B.D., Beaumont, M., Correa, D.S., Rojas, O.J., 2021. The food-materials nexus: Next generation bioplastics and advanced materials from agri-food residues. Adv. Mater. 33, e2102520.
Pahmeyer, M.J., Siddiqui, S.A., Pleissner, D., Gołaszewski, J., Heinz, V., Smetana, S., 2022. An automated, modular system for organic waste utilization using *Hermetia illucens* larvae: Design, sustainability, and economics. J. Clean. Prod. 379, 134727. https://doi.org/10.1016/j.jclepro.2022.134727.
Paritosh, K., Yadav, M., Mathur, S., Balan, V., Liao, W., Pareek, N., Vivekanand, V., 2018. Organic fraction of municipal solid waste: Overview of treatment methodologies to enhance anaerobic biodegradability. Front. Energy Res. 6, 75. https://doi.org/10.3389/fenrg.2018.00075.
Peydayesh, M., Bagnani, M., Mezzenga, R., 2021. Sustainable bioplastics from amyloid fibril-biodegradable polymer blends. ACS Sustainable Chem. Eng. 9, 11916–11926. https://doi.org/10.1021/acssuschemeng.1c03937.
Peydayesh, M., Bagnani, M., Soon, W.L., Mezzenga, R., 2023. Turning food protein waste into sustainable technologies. Chem. Rev. 123, 2112–2154. https://doi.org/10.1021/acs.chemrev.2c00236.
Pimentel, A.C., Montali, A., Bruno, D., Tettamanti, G., 2017. Metabolic adjustment of the larval fat body in *Hermetia illucens* to dietary conditions. J. Asia. Pac. Entomol. 20, 1307–1313. https://doi.org/10.1016/j.aspen.2017.09.017.
Purewal, S.S., Kaur, A., Bangar, S.P., Singh, P., Singh, H., 2024. Protein-based films and coatings: An innovative approach. Coatings 14, 32. https://doi.org/10.3390/coatings14010032.
Rehman, K.U., Hollah, C., Wiesotzki, K., Rehman, R.U., Rehman, A.U., Zhang, J., Zheng, L., Nienaber, T., Heinz, V., Aganovic, K., 2023. Black soldier fly, *Hermetia illucens* as a potential innovative and environmentally friendly tool for organic waste management: A mini-review. Waste Manag. Res. 41, 81–97. https://doi.org/10.1177/0734242X221105441.
Rolff, J., Johnston, P.R., Reynolds, S., 2019. Complete metamorphosis of insects. Phil. Trans. r. Soc. B 374, 20190063. https://doi.org/10.1098/rstb.2019.0063.
Sadasivam, S., Manickam, A., 2005. Biochemical methods, 2nd Edition. New age International, New Delhi.
Salomone, R., Saija, G., Mondello, G., Giannetto, A., Fasulo, S., Savastano, D., 2017. Environmental impact of food waste bioconversion by insects: Application of Life Cycle Assessment to process using *Hermetia illucens*. J. Clean. Prod. 140, 890–905. https://doi.org/10.1016/j.jclepro.2016.06.154.
Setti, L., Francia, E., Pulvirenti, A., De Leo, R., Martinelli, S., Maistrello, L., Macavei, L.I., Montorsi, M., Barbi, S., Ronga, D., 2020. Bioplastic film from black soldier fly prepupae proteins used as mulch: Preliminary results. Agron. 10, 933. https://doi.org/10.3390/agronomy10070933.
Shen, M., Huang, W., Chen, M., Song, B., Zeng, G., Zhang, Y., 2020. (Micro)plastic crisis: Un-ignorable contribution to global greenhouse gas emissions and climate change. J. Clean. Prod. 254, 120138. https://doi.org/10.1016/j.jclepro.2020.120138.
Shen, Y., Levin, A., Kamada, A., Toprakcioglu, Z., Rodriguez-Garcia, M., Xu, Y., Knowles, T.P.J., 2021. From protein building blocks to functional materials. ACS Nano 15, 5819–5837. https://doi.org/10.1021/acsnano.0c08510.
Smets, R., Verbinnen, B., Van De Voorde, I., Aerts, G., Claes, J., Van Der Borght, M., 2020. Sequential extraction and characterisation of lipids, proteins, and chitin from black soldier fly (*Hermetia illucens*) larvae, prepupae, and pupae. Waste Biomass Valor. 11, 6455–6466. https://doi.org/10.1007/s12649-019-00924-2.
Soetemans, L., Uyttebroek, M., Bastiaens, L., 2020. Characteristics of chitin extracted from black soldier fly in different life stages. Int. J. Biol. Macromol. 165, 3206–3214. https://doi.org/10.1016/j.ijbiomac.2020.11.041.
Starcher, B., 2001. A ninhydrin-based assay to quantitate the total protein content of tissue samples. Anal. Biochem. 292, 125–129. https://doi.org/10.1006/abio.2001.5050.
Stelmock, R.L., Husby, F.M., Brundage, A.L., 1985. Application of Van Soest acid detergent fiber method for analysis of shellfish chitin. J. Dairy Sci. 68, 1502–1506. https://doi.org/10.3168/jds.S0022-0302(85)80989-9.
Surendra, K.C., Tomberlin, J.K., van Huis, A., Cammack, J.A., Heckmann, L.-H.-L., Khanal, S.K., 2020. Rethinking organic wastes bioconversion: Evaluating the potential of the black soldier fly (Hermetia illucens (L.)) (Diptera: Stratiomyidae) (BSF). Waste Manag. 117, 58–80. https://doi.org/10.1016/j.wasman.2020.07.050.
Tabernero, A., González-Garcinuño, Á., Sánchez-Álvarez, J.M., Galán, M.A., Del Valle, E. M.M., 2017. Development of a nanoparticle system based on a fructose polymer: Stability and drug release studies. Carbohydr. Polym. 160, 26–33. https://doi.org/10.1016/j.carbpol.2016.12.025.
Tettamanti, G., Bruno, D., 2024. Black soldier fly larvae should be considered beyond their use as feedstuff. J. Insects Food Feed 10, 1–7. https://doi.org/10.1163/23524588-241001ED.
Trippe, A., 2015. Guidelines for preparing patent landscape reports. World intellectual property organization, Geneva, 131 pp. Doi: 10.34667/tind.28858.
United Nations Environment Programme, 2021. Food waste index report 2021. https://www.unep.org/resources/report/unep-food-waste-index-report-2021.
van der Fels-Klerx, H.J., Meijer, N., Nijkamp, M.M., Schmitt, E., van Loon, J.J.A., 2020. Chemical food safety of using former foodstuffs for rearing black soldier fly larvae (*Hermetia illucens*) for feed and food use. J. Insects Food Feed 6, 475–488. https://doi.org/10.3920/JIFF2020.0024.
Vandeweyer, D., Bruno, D., Bonelli, M., IJdema, F., Lievens, B., Crauwels, S., Casartelli, M., Tettamanti, G., De Smet, J., 2023. Bacterial biota composition in gut regions of black soldier fly larvae reared on industrial residual streams: Revealing community dynamics along its intestinal tract. Front. Microbiol. 14, 1276187. https://doi.org/10.3389/fmicb.2023.1276187.
Volk, R., Stallkamp, C., Steins, J.J., Yogish, S.P., Müller, R.C., Stapf, D., Schultmann, F., 2021. Techno-economic assessment and comparison of different plastic recycling pathways: A German case study. J. Ind. Ecol. 25, 1103–1376. https://doi.org/10.1111/jiec.13145.
Yashwant, A.S., Kashyap, P., Goksen, G., 2023. Recent advances in the improvement of protein-based edible films through non-thermal and thermal techniques. Food Biosci. 55, 103032. https://doi.org/10.1016/j.fbio.2023.103032.