Components of Silkworm Pupae

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Silkworm pupae are insects that are beneficial to human health, not only for their high nutritional value but, more importantly, for the variety of pharmacological functions they can perform when consumed.

Keywords: silkworm pupae ; composition ; functional food

1. Introduction

The silkworm is a lepidopteran insect. The life of a silkworm usually goes through five stages, lasting a total of about seven weeks. When the silkworm eggs hatch, they turn into newly hatched black and brown silkworms. After feeding and growing and shedding its shell five times, the silkworm becomes a mature silkworm, stops feeding, and starts to spit out a lot of silk in preparation for cocooning. This process takes 24–28 days. After 4 days of cocooning, the matured silkworm turns into a silkworm pupa. After about 2 weeks, the silkworm pupae turns into silkworm moths. The silkworm moths finish laying eggs within 3–5 days and die soon afterwards. Silkworm pupae are considered to be the harvest period for silkworm consumption as they are consumed as food in many regions due to their high nutritional value and various biomedical functions. **Figure 1** depicts the life cycle of the silkworm [1][2].

Figure 1. The life cycle of the silkworm.

Silkworm pupae are one of the main by-products of the silk industry and are mostly used as animal feed and fertiliser in South East Asia, for example in Japan, Korea, and India [3][4][5]. Silkworm pupae are also used as food insects, especially in China, where they have been eaten for over 2000 years [6]. There are many species of silkworm pupae; at present, the main commercial silkworm pupae used for research are *Bombyx mori*, *Antheraea pernyi*, *Antheraea yamamai*, *Samia ricini*, *Antheraea mylitta*, *Antheraea roylei*, and other species [4][7][8]. Additionally, the material composition and functional roles of these species are different. The biggest difference between the different species of silkworm pupae is the source of the silkworm's diet and the degree of domestication. For example, the mulberry silkworm, which eats mulberry leaves, is the silkworm that has been fully domesticated and is the most widely farmed [4]. Rich in proteins, oils, chitosan, vitamins, polyphenols, and other nutrients, silkworm pupae have long been used as an important source of high-quality proteins and lipids [9]. Silkworm pupae protein contains 18 amino acids and is rich enough in essential amino acids to meet the amino acid requirements of humans and is beneficial to human health [10][11]. Silkworm pupae oil contains a large number of unsaturated amino acids, especially Omega-3 fatty acids [12].

Silkworm pupae have long been used in a single way for direct consumption, for example as feed [5]. Gradually, the pupae have been further processed to extract nutrients and active ingredients and are used in food modification and pharmaceutical development [13]. Some researchers have used silkworm pupa powder as a protein enhancer in functional foods, and the addition of silkworm pupa powder enhances the flavour and taste of functional foods [13][14][15]. Examples are bread, yogurt, and food additives [7][13][16]. In addition, silkworm pupae can also be used in industry [17]. However, silkworm pupae are still not accepted by everyone because of the presence of allergens and unfriendly odours. We need to consider the safety and acceptability of silkworm pupae more thoroughly when using them [18]. In recent years, numerous studies have found that the active ingredients in silkworm pupae have various pharmacological functions, such as: anticancer, antioxidant, hepatoprotective, antibacterial, antiapoptotic, and immunomodulatory functions. This provides a broader prospect for the application of silkworm pupae. In the future, silkworm pupae will rapidly be developed for the health food and biomedical industries to meet the human demand for nutritious food and safe medicine [19][20].

2. Components of Silkworm Pupae

Silkworm pupae are rich in many nutrients. Protein, fat, and sugar are the most abundant substances, as well as minerals, vitamins, polyphenolic compounds, and many other nutrients [21][22].

2.1. Silkworm Pupae Protein

Bombyx mori has a high protein content of 55.6% dry weight and is the most abundant dry matter in silkworm pupae [9].Biologically active peptides are peptides containing from several to several dozen amino acids, which have a variety of physiological functions [23]. These pupae proteins can be hydrolysed to produce a variety of biologically active peptides, which in turn can perform the pharmacological functions of silkworm pupae. The amino acid composition of the proteins is essentially the same in the different species of silkworm pupae, all consisting of 18 amino acids (except for *Eri* silkworm pupae). Of these, eight essential amino acids meet the requirements of the WHO/FAO/UNU recommendations. In addition, there are 10 non-essential amino acids that meet human requirements. Compared to hen eggs, pupae are higher in Phe and Pro [24]. Therefore, silkworm pupae are considered to be a high-quality source of protein and an important nutrient in silkworm pupae [25]. **Table 1** summarises the amino acid composition of the different varieties of silkworm pupae proteins.

Table 1. Amino acid composition of different varieties of silkworm pupae proteins [9][21][22][24].

Amino Acid (g/100 g of Protein)	*Bombyx mori*	*Eri* Silkworm Pupae	Mulberry Silkworm Pupae	*Antheraea pernyi*	Hen Egg
Asp	**9.1**	**9.89**	**10.9**	**6.41**	**8.92**
Thr	**3.9**	**4.75**	**5.4**	**4.64**	**4.47**
Ser	**3.7**	**5.25**	**4.7**	**4.64**	**6.72**
Glu	**9.5**	**12.9**	**14.9**	**12.74**	**12.13**
Gly	**3.6**	**4.94**	**4.6**	**4.42**	**3.02**
Ala	**3.9**	**6.05**	**5.5**	**6.26**	**5.03**
Cys	**1.4**	**0.53**	**1.4**	**1.5**	**1.90**
Val	**4.7**	**5.36**	**5.6**	**6.63**	**5.42**
Met	**3.4**	**2.31**	**4.6**	**1.47**	**2.81**
Ile	**3.4**	**4.42**	**5.7**	**7.95**	**4.88**
Leu	**6.2**	**6.63**	**8.3**	**3.24**	**8.11**
Tyr	**5.6**	**6.4**	**5.4**	**2.06**	**3.81**
Phe	**4.6**	**5.24**	**5.1**	**8.10**	**4.82**

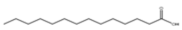
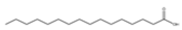
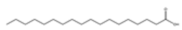
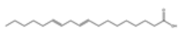
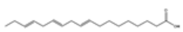
Amino Acid (g/100 g of Protein)	*Bombyx mori*	*Eri* Silkworm Pupae	Mulberry Silkworm Pupae	*Antheraea pernyi*	Hen Egg
Lys	**6.1**	**6.54**	**7.5**	**4.54**	**6.59**
His	**2.7**	**2.67**	**2.5**	**2.94**	**2.09**
Arg	**4.7**	**4.41**	**6.8**	**4.12**	**5.70**
Pro	**7.0**	**6.46**	**4.0**	**12.22**	**3.38**
Trp	**1.5**	**NA**	**0.9**	**4.05**	**1.72**

Values are expressed as g/100 g of protein. NA: Data not available.

2.2. Silkworm Pupae Oil

In silkworm pupae, the oil content is second only to protein. Of the four different species of silkworm pupae, *Eri* silkworm pupae has the highest oil content, at 26.2% [22]. The fatty acid composition of the different varieties of silkworm pupae oil is summarised in **Table 2**. As can be seen from the table, all the different silkworm pupae oils contain high levels of unsaturated fatty acids, with 77.71% in *Antheraea pernyi*. In addition to the fatty acids listed in the table, silkworm pupae also contain eicosapentaenoic acid and docosahexaenoic acid, which are Omega-3 fatty acids and have an important role in promoting human health [26]. Oil is an important nutrient, and silkworm pupae are not only rich in oils but also contain high levels of unsaturated fatty acids, especially polyunsaturated fatty acids, which have significant nutritional value as a source of edible oil [12].

Table 2. Fatty acid composition of different varieties of silkworm pupae oil [9][12][19][26][27][28].

Fatty Acids (Percentage of Fatty Acids)	Chemical Structure	*Bombyx mori*	*Eri* Silkworm Pupae	Mulberry Silkworm Pupae	*Antheraea pernyi*	Sunflower Oil
Myristic acid (C14:0)		**0.1**	**ND**	**0.18**	**NA**	**NA**
Palmitic acid (C16:0)		**24.2**	**26.98**	**23.18**	**17.25**	**5.6**
Palmitoleic acid (C16:1)		**1.7**	**1.82**	**1.07**	**1.14**	**NA**
Stearic acid (C18:0)		**4.5**	**4.73**	**4.69**	**2.23**	**2.2**
Oleic acid (C18:1)		**26.0**	**15.89**	**28.32**	**29.15**	**25.1**
Linoleic acid (C18:2)		**7.3**	**5.49**	**3.88**	**7.14**	**66.2**
α-Linolenic acid (C18:3)		**36.3**	**44.73**	**38.25**	**40.28**	**NA**
Saturated fatty acids	**—**	**28.8**	**31.71**	**28.05**	**19.48**	**7.8**
Monounsaturated fatty acids	**—**	**27.7**	**17.71**	**29.39**	**30.29**	**25.1**
Polyunsaturated fatty acids	**—**	**43.6**	**50.22**	**42.13**	**47.42**	**66.2**

Values are expressed as a percentage of fatty acids. NA: data not available.

2.3. Minerals

Minerals have an important role in living organisms. They are present in many forms in silkworm pupae. There are up to 25 different types of minerals in silkworm pupae, and these minerals may perform certain physiological functions in the organism [7][12]. **Table 3** lists the content of eight minerals in the three types of pupae, from which it can be seen that phosphorus, calcium, and magnesium are higher in the pupae. The type and content of minerals in pupae can vary depending on the type of pupa and the environment in which they have grown [24]. It is worth noting that the sodium-to-potassium (Na: K) ratio in silkworm pupae is very low, except for the minerals listed in the table. Na:K predicts the occurrence of non-communicable diseases, suggesting that consumption of silkworm pupae may reduce the likelihood of non-communicable diseases [29][30]. Non-communicable diseases include stroke, hypertension, cardiovascular disease, etc. [31][32]. Some pupae are also rich in selenium, which can be enriched in the pupae protein. Selenium-rich pupae play an important role in cancer prevention and defence against oxidative stress [33][34].

Table 3. Mineral composition of different species of silkworm pupae [21][22][24][35].

Minerals (mg/100 g Dry Weight)	*Bombyx mori*	*Eri* Silkworm Pupae	*Antheraea pernyi*
Phosphorus	474	584	272
Iron	26	24	4
Calcium	158	74.2	63
Zinc	23	7.24	3.57
Copper	0.15	1.75	0.73
Magnesium	207	178	154
Manganese	0.71	2.54	NA
Chromium	1.69	NA	9.84

Values are expressed as mg/100 g dry weight. NA: data not available.

2.4. Other Ingredients in Silkworm Pupae

In addition to the above ingredients, silkworm pupae contain many vitamins and are rich in them. For example, VA can reach 5 mg/g. The main vitamins in silkworm pupae include VA, VB1, VB2, VB3, VB5, VB7, VB9, VB12, VC, and VE [19][36]. Phospholipids and five tocopherols are also present in silkworm pupae. The five tocopherols are α-Tocopherol, β-tocopherol, γ-tocopherol, γ-tocotrienol, and σ-tocopherol [28]. Rare dimethyladenosine derivatives are also found in silkworm pupae [37]. Additionally, silkworm pupae contain polyphenols and flavonoids. Polyphenols and flavonoids were found in the pupae of the silkworm *Antheraea assamensis* at concentrations of 10 mg/g and 20 mg/g, respectively [35]. In native *Thai* mulberry silkworm pupae, the polyphenols mainly contain (+)-catechin, (−)-epicatechin, rutin, quercetin, myricetin, trans-resveratrol, luteolin, naringenin, and kaempferol [38]. The sugars in silkworm pupae can be divided into two main groups, chitosan and chitin, as well as isolated and purified polysaccharides, all of which are biologically active [39][40][41]. Chitosan and chitin from silkworm pupae are not cytotoxic, but have strong physiological activity, especially carboxymethyl chitosan [42][43]. All these substances had certain biofunctional activities, which contribute to the basis of the pharmacological functions of silkworm pupae.

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