

Earthworms: A Source of Protein

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Abstract: Earthworms are important invertebrates that have been widely used as food and traditional medicine sources for thousands of years. Not only have researchers proven that earthworms are rich in proteins and other nutrients, they have also taken a keener interest in their unique pharmaceutical properties. Recent research has successfully discovered some beneficial functional components of earthworms due to the rapid development of biological technologies in the past decades. Therefore, earthworms could be a novel dietary supplement for human consumption. This review aims to summarize the current research about nutritional and therapeutic values of earthworms; and present a matured earthworm-derived product from Bocom Pharmaceuticals (USA) Corp as an example of its incorporation into a dietary supplement.

Key words: Earthworm protein, earthworm powder, Bocom.

1. Introduction

Protein is an essential component of human diet; its quality can be evaluated following the World Health Organization (WHO) and Food and Agriculture Organization of the United Nations (FAO) guidelines. From the perspective of protein digestibility and amino acid score (AAS), animal proteins had better performance compared to proteins with plant origin [1]. In the “western diet”, people limit the number of animal species (livestock, poultry, fish, etc.) that they consume for energy and nutrients, but alternative food sources like invertebrates and insects are also considered nutritious and beneficial to human health [2, 3]. One such alternative food source is the earthworm. Earthworms, which occupy over 80% invertebrate biomass in terrestrial ecosystem, are the largest members of Oligochaetaor “segmented worms”. Not only do they play essential roles as “waste manager”, “fertility improver”, “soil manager” and “plant growth promoter” in our ecosystem, they also have a long history being used as food and medicinal products, which can be traced back to the

early 1340 A.D. [4, 5]. In the current market, a company called Bocom Pharmaceuticals (USA) Corp (“Bocom”) based in Southern California is dedicated to developing earthworm-derived products, and it has successfully registered earthworm protein as a New Dietary Ingredients (NDI) in FDI.

2. Food Sources

2.1 Earthworm as a Delicacy

Earthworms have been consumed and continue to be utilized in meals by people across the world. In China, earth worms have been consumed as a form of cuisine since ancient time. For example, P’ing-Chou k’o-t’an, a special dish from Hainan Island, is earthworm cooked with pieces of bamboo. Nowadays, some people in Fujian, Guangdong, and Taiwan still consume earthworm in their diet [6]. In Japan, making earthworm pie has been recorded as a custom [7]. A Dutch traveler observed Africans at the border between Transvaal and Botswana eating earthworms grilled on sticks. An indigenous earthworm named “kurekure”, known for its sweet and residual flavor, was recorded to be a tasty food material for Maori chiefs and offered as the last food for dying people [4]. Earthworms contribute significantly to the diet of

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people living in Amazon Basin as well. Early reports from mid-19th-century showed evidences about earthworm consumption in the Rio Negro area [8]. Smoked earthworms were sold three times the price in comparison with other delicacies, such as game, pork, chicken or smoked fish in Alto Orinoco markets of Venezuela. Traditionally, a dish contained earthworm and cassava was prepared for women after parturition among Yekuana, a tribe in Venezuela [9].

2.2 Nutritional Profile

Earthworms have also been well established by researchers as a great protein source for animal feeds in poultry and aquaculture practices [10-14]. Despite that earthworm consumption for humans is not as popular as ancient time, their superior nutrient levels are not deniable. Nutritional data of earthworms, specifically nutrient composition, essential amino acid (EAA) profiles, and mineral composition, were excerpted from literature and databases to compare with some commercially raised invertebrates and commonly consumed protein sources [3, 6, 9, 12, 15-19].

Table 1 shows the nutrient composition (dry-matter basis, DM) of earthworms and alternative food sources, including some commercial raised

invertebrates and conventional food. Protein usually contributes the highest concentration in nutrition. According to the listed data, protein contents of different earthworm species range between 61.9-72.9%, slightly higher than conventional protein sources like beef, pork, egg and salmon. Only chicken had a comparable protein percentage. Under the condition of industrial production, earthworm powder produced by Bocom contains more than 65% protein, even after a 6-month stability experiment (personal communication). Nutrient composition in invertebrates varied, likely due to differences in species, reproductive states, season, age/life stage, or sex [20]. Compared to selected invertebrates, earthworms contain relatively lower fiber content, which may be explained by the difference between the soft body (earthworm) and the hard exoskeleton (other invertebrates) [3]. In terms of fat content, selected invertebrate species vary greatly from 8.1% to 60%; chicken contains the lowest (30.3%) while pork has the highest (54.4%) among the common commodities. The crude fat level in earthworm is much lower than conventional food sources, and ω 3 polyunsaturated lipids in earthworms are quite high, similar to the lipid composition of some fish oils [21].

Table 1 Nutrient composition of earthworms and alternative food sources on dry weight basis (%DM).

| Source | Species | Preparation | Crude protein | Crude fat | Crude fiber | Ash | Energy (kcal/g) |
|-------------------|-------------------------------|--------------------|---------------|-----------|-------------|-----|-----------------|
| Earthworm | <i>Hyperiodriluseuryaulos</i> | Whole/raw/fasted | 63.0 | 5.9 | 1.9 | 8.9 | 3.5 |
| Earthworm | <i>Lunbricus terrestris</i> | Whole/raw/fasted | 64.0 | 9.8 | 0.6 | 3.7 | 4.3 |
| Earthworm | <i>Eisenia fetida</i> | Whole/raw/fasted | 61.9 | 11.1 | | 8.7 | |
| Earthworm | <i>Kuru</i> | Gut organs removed | 72.9 | 10.5 | | | |
| Earthworm | <i>Motto</i> | Gut organs removed | 64.4 | 6.6 | | | |
| Mealworm (larvae) | <i>Tenebrio molitor</i> | Whole/raw/fasted | 49.1 | 35.2 | 6.6 | 2.4 | 5.4 |
| Mealworm (adult) | <i>Tenebrio molitor</i> | Whole/raw/fasted | 65.3 | 16.1 | 20.4 | 3.3 | 3.8 |
| Waxworm | <i>Galleria mellonella</i> | Whole/raw/fasted | 34.0 | 60.0 | 8.2 | 1.5 | 6.6 |
| Silkworm | <i>Bombyx mori</i> | Whole/raw/fasted | 53.8 | 8.1 | 6.4 | 6.4 | 3.4 |
| Cricket (nymph) | <i>Acheta domesticus</i> | Whole/raw/fasted | 67.3 | 14.4 | 9.6 | 4.8 | 4.1 |
| Beef | | Ground/raw | 59.1 | 12.7 | | 5.2 | 6.0 |
| Pork | | Ground/raw | 43.4 | 21.2 | | 2.2 | 6.8 |
| Chicken | | Ground/raw | 65.2 | 8.1 | | 4.4 | 5.3 |
| Egg | | Whole/raw | 52.7 | 9.5 | | 4.4 | 6.0 |
| Salmon | <i>Atlantic</i> | Farmed/raw | 58.2 | 13.4 | | 3.2 | 5.9 |

Table 2 lists the EAA profiles of the selected protein sources. Essential amino acids are amino acids that cannot be synthesized by our body, i.e., they can only be obtained from our diets. Using hen egg protein as a reference, an EAA index called chemical score to protein score ratio (CSPS) was calculated according to Sogbesan & Ugwumba [18]:

CSPS (%) = (Total EAA of the sample / Total EAA of whole hen egg) × 100

All earthworm species presented were comparable with egg protein in terms of the total EAA amount. Especially for *Eisenia fetida*, *kuru* and *motto*, not only

do they have 30-40% higher EAA content when compared to eggs, their CSPS performance is better than other invertebrates and even some common food like beef and pork.

Elementary analysis is presented in Table 3. Worms' mineral levels are generally higher than the conventional food. From the perspective of earthworms, though their amounts of minerals differ among species, they are especially richer in Ca and Fe compared to conventional meats. Calcium contents in *Motto* are comparable with fresh cheese and cows' milk [9]. Based on the mineral requirements set by WHO

Table 2 Essential amino acid (EAA) profile of earthworms and alternative food sources on dry weight basis (%DM).

| | Arg | His | Ile | Leu | Lys | Met | Phe | Thr | Trp | Val | Total EAA | CSPS (%) |
|-----------------------------|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----------|----------|
| <i>Lumbricusterresstris</i> | 3.7 | 1.4 | 2.3 | 4.6 | 4.0 | 1.2 | 2.3 | 2.9 | 0.5 | 2.6 | 25.5 | 1.0 |
| <i>Eisenia fetida</i> | 6.0 | 2.1 | 2.9 | 6.2 | 4.9 | 1.1 | 2.3 | 2.8 | - | 3.8 | 32.1 | 1.3 |
| <i>Kuru</i> | 6.0 | 1.9 | 3.4 | 6.2 | 5.4 | 1.7 | 3.0 | 3.4 | 0.8 | 3.4 | 35.2 | 1.4 |
| <i>Motto</i> | 5.6 | 1.6 | 3.0 | 5.5 | 5.0 | 1.4 | 2.7 | 3.0 | 0.9 | 3.2 | 31.9 | 1.3 |
| Mealworm (larvae) | 2.5 | 1.5 | 2.5 | 5.2 | 2.7 | 0.6 | 1.7 | 2.0 | 0.4 | 2.9 | 22.1 | 0.9 |
| Mealworm (adult) | 2.8 | 1.9 | 2.8 | 5.4 | 2.9 | 0.8 | 1.7 | 2.2 | 0.7 | 4.1 | 25.4 | 1.0 |
| Waxworm | 1.7 | 0.8 | 1.5 | 3.0 | 1.9 | 0.5 | 1.3 | 1.4 | 0.3 | 1.6 | 14.1 | 0.6 |
| Silkworm | 2.3 | 1.4 | 1.7 | 2.8 | 2.5 | 0.8 | 1.6 | 1.7 | 0.4 | 2.2 | 17.3 | 0.7 |
| Cricket (nymph) | 4.1 | 1.5 | 2.9 | 6.4 | 3.6 | 0.9 | 1.9 | 2.4 | 0.3 | 3.3 | 27.3 | 1.1 |
| Beef | 2.9 | 1.5 | 2.0 | 3.5 | 3.7 | 1.2 | 1.8 | 1.7 | 0.2 | 2.2 | 20.7 | 0.8 |
| Pork | 2.7 | 1.7 | 2.0 | 3.5 | 3.9 | 1.1 | 1.7 | 2.0 | 0.5 | 2.4 | 21.6 | 0.9 |
| Chicken | 4.2 | 2.0 | 3.0 | 5.1 | 5.6 | 1.7 | 2.6 | 2.7 | 0.5 | 3.1 | 30.5 | 1.2 |
| Egg | 3.2 | 1.2 | 2.5 | 4.3 | 3.4 | 1.5 | 2.6 | 2.2 | 0.8 | 3.1 | 24.8 | 1.0 |
| Salmon | 3.5 | 1.6 | 2.8 | 4.6 | 5.3 | 1.8 | 2.4 | 2.4 | 0.6 | 3.2 | 28.1 | 1.1 |

Table 3 Trace elements of earthworms and alternative sources on dry weight basis (mgkg⁻¹ DM).

| | Ca | Fe | I | Mg | Se | Zn |
|------------------------------|--------------------|--------------------|------------------|--------------------|-------------------|--------------------|
| <i>Lumbricus terresstris</i> | 2,707 | 307.3 ^b | 2.3 ^b | 829 | 2.4 ^b | 108 ^b |
| <i>Eisenia fetida</i> | 8,200 ^a | 495.3 ^b | | 1,000 | | 150.7 ^b |
| <i>Kuru</i> | 2,650 | 1,050 ^b | | 527 | 9.02 ^b | 149 ^b |
| <i>Motto</i> | 7,070 ^a | 2,990 ^b | | 792 | | 131 ^b |
| Mealworm (larvae) | 444 | 54.1 | 0.4 | 2,102 ^a | 0.7 ^b | 136 ^b |
| Mealworm (adult) | 636 | 60.1 | 0.6 | 1,669 | 0.4 ^a | 127 ^b |
| Waxworm | 586 | 50.4 | | 761 | 0.3 ^a | 61 ^a |
| Silkworm | 1,023 | 95.4 ^b | | 2,879 ^a | 0.8 ^b | 177 ^b |
| Cricket (nymph) | 1,201 | 92.6 | 1.2 ^b | 987 | 0.4 ^a | 297 ^b |
| Beef | 365 | 60.5 | | 578 | | 138.4 ^b |
| Pork | 360 | 22.6 | | 488 | | 56.5 ^a |
| Chicken | 224 | 30.6 | | 785 | | 54.9 ^a |
| Egg | 2,348 | 73.4 | | 503 | | 54.1 ^a |
| Salmon | 256 | 9.7 | | 769 | | 10.3 |

a: 33-67% mineral recommendation set by WHO for a male adult; b: 67-100% mineral recommendation set by WHO for a male adult.

and FAO, a male adult is recommended to intake 1,000 mg Ca, 14 mg Fe, 0.13 mg I, 260 mg Mg, 0.034 mg Se and 7 mg Zn per day [22]. Assuming 50 g (DM) earthworm consumption daily, a male adult would be able to fully meet his Fe and Se recommendations and cover the majority of the requirements for I and Zn. Mg intake from such amount of earthworm will not meet the recommendation, but Mg from other sources (same consumption) is not sufficient either. Therefore, around two spoons/servings of earthworm protein powder are believed to fulfill most daily dietary needs for a healthy male adult. In terms of the effect of extraction methods, Gunya et al. [23] concluded that the freeze drying process avoids losses of *Eisenia fetida* minerals in comparison with the oven-dry method, which might be explained by different processing temperatures. Sun & Jiang (2017) also reported rich vitamins in body fluids of *Eisenia fetida*.

3. Medicinal Values

Many ancient cultures, especially in the eastern hemisphere, have been utilizing earthworms for their medicinal benefits. Some herbalistic systems, Ayurveda (India), Traditional Chinese Medicine (China), Kampo (Japan), Traditional Korean Medicine (Korea), and Traditional Arabic and Islamic Medicine (TAIM), overlapped with regard to some practices using earthworms and their healing properties [4, 24]. From the perspective of contemporary medicine, development of one medicinal product usually starts from qualitative research until supported by quantitative evidence. Apart from the therapeutic properties of earthworms noted in traditional medicinal systems, they can be a great model for contemporary biological and pharmaceutical studies due to the lack of financial constraints and ethical restrictions. Therefore, increasing evidence-based investigations have revealed the therapeutic mechanisms of earthworms through bioprospecting as potential sources for medicinal usage [24].

3.1 Traditional Medicine

Earthworm is one of the most widely used material medica of animal origin in traditional medicinal systems [25]. According to studies from all over the world, earthworms have been reported to serve as galactagogues and treat several ailments including hypertension, smallpox, dental problems, headache, otitis, fever, asthma, hemorrhoids, arthritis, chronic cough, jaundice, hair loss, urinary impairments, wound healing, ulcers, influenza, and other symptoms and clinical presentations [26–28]. Some examples are listed below in this review.

In China, not only was earthworm listed in well-known ancient medicinal books, such as *Shennong Ben Cao Jing* or *Ben Cao Gang Mu*, it was also covered and described in contemporary publications like *Pharmacopoeia of People's Republic of China* [6, 29]. A clinical study showed that gyanyankan (a Chinese medicine mixture including earthworms) was effective in treating patients' mild hypertension [30, 31]. Pulverized mixture of earthworm and sandalwood can be a remedy for otitis with discharge [32]. Smallpox patients in Burma and Laos bathed in water soaked with earthworms and took coconut juice with roasted earthworm powder. It was believed that this practice reduced the mortality rate from 100% to 25% [33]. Similarly in India, earthworms were cooked with kingfish to feed children suffering from smallpox in the Nyishi tribe [34]. People from the area of Andhra Pradesh applied earthworm-camphor paste to the forehead to relieve headaches [35, 36]. Dried earthworms were used to cure wounds, sores, chronic boils, chronic cough and diphtheria whereas oil extracted from earthworms for hemiplegia, paralysis and muscular pain in Kerala [37]. Tribes in Tamil Nadu extracted earthworm mucus and gave it to children before meals for three days in order to alleviate asthma and prevent it from recurring [38]. Moreover, earthworms served as galactagogues to induce human lactation in areas including

Chhattisgarh, Jharkhand, Madhya Pradesh, West Bengal, and Orissa [40, 41]. Iranians even considered earthworms to be a kind of all-around wonder drug [33].

In Africa, earthworm decoction was used to treat influenza; earthworms would be placed over the infected wound and held there with a cloth; ground earthworm paste was applied to the ear infection area [41]. Consuming earthworm powder with semi-liquid food was a practice in Nigeria for stomach ulcer [42]. Additionally, earthworm *Alma millsoni* was used to treat measles and bone problems and induced faster contractions for easier birth [43]. European folklore also recorded the medicinal values of earthworms for convulsion from epilepsy, arthritis, anodyne, diuretic, and diaphoretic usages [44, 45]. In Brazil, earthworms are used to treat schistosomiasis and lumps [46]. Local healers in Venezuela have used earthworms to treat malaria and anemia as well as giving them to women after childbirth [9]. Earthworm extracts from *Perionyx excavatus* could be an effective drug for rheumatic fever and rheumatic heart disease for Cherokee Native Americans in southern Kentucky [26].

3.2 Contemporary Researches

Years of evolutionary selection may result in development of some naturally occurring bioactive molecules with potential in medicinal usage. Despite the fact that earthworms have been living on our planet much longer than humans, it was not until the past decades that we began to raise awareness about their unique components from the biochemical point of view. Earthworms have been characterized for their fibrinolytic, anti-tumor, cytotoxic, antiviral, DNase, and anti-inflammatory activities [47].

3.2.1 Anti-fatigue and Immunologic Effects

Earlier animal studies have shown that mice taking insect-added feeds (ant, bee pollen, silkworm pupa, etc.) increased their swim endurance performance, and their selected biochemistry indexes improved significantly comparing to the control group without

fortification, proving the anti-fatigue effect of these insects [48]. The same procedure was adopted for studies about earthworm protein as well. Earthworm proteins and taurine composite significantly extended the loaded-swimming time of mice in high dose group ($1.5 \text{ g}\cdot\text{kg}^{-1}$ body weight after 30 days), reduced the serum urea nitrogen, and increased hepatic glycogen reserves in mice of middle dose group ($0.5 \text{ g}\cdot\text{kg}^{-1}$ body weight after 30 days) [49].

The immunologic function of earthworms may come from the production of several types of leukocytes and some secreted immune-protective molecules [50]. Significant increments of foot metatarsus thickened values and α indices in experiment groups where mice were fed with additional earthworms were observed and thus became evidences for earthworm immunologic functions [49]. An *in vitro* study concluded that earthworm injections were able to stimulate cell proliferation of splenocytes and increase the secretion of IL-2 (an important cytokine for T-cell proliferation) significantly, thus confirming the immunoregulatory activity [51]. Furthermore, active peptides (8,000-20,000 u) separated and purified from earthworms were found to enhance the phagocytic activity of mice macrophage, also suggesting their immuno-regulatory function [52]. An *in vivo* experiment on mice confirmed that earthworm peptide (3,000-20,000 Da) injection can enhance lymphocyte proliferation, phagocytic activity of macrophage, and the nitric oxide (NO) level in normal mice (0.1 and $0.5 \text{ mg}\cdot\text{mL}^{-1}$) and even in immunological suppression mice ($0.5 \text{ mg}\cdot\text{mL}^{-1}$) [53].

3.2.2 Cardiovascular and Blood System Effects

A major cause of cardiovascular diseases is intravascular thrombosis, resulting from aggregation of fibrin in arteries. Fibrin is the main component of blood clots, which can be dissolved by fibrinolytic enzymes [50]. Studies involving several earthworm species have purified some fibrinolytic enzymes and clinically confirmed their positive effects. Mihara et al. [55] first isolated lumbrokinase with fibrinolytic and

thrombolytic activities from *L. rubellus*. Similar enzymes were found from other earthworm species as well [55, 56]. Lumbrokinase can effectively degrade fibrinogen, thereby reducing blood viscosity [57]. Wang et al. [58] reviewed that earthworm protease (EfP-III-1) partially activates prothrombin (cleavage at Arg274-Thr275) to produce thrombin, keeping the balance between fibrinolysis and fibrogenesis and decreasing severe bleeding complications in clinical usage, compared to other anticlotting drugs, such as tissue plasminogen activator (tPA) and urokinase. Furthermore, high stability and strong tolerance to organic solvents and high temperature of most earthworm fibrinolytic enzymes make them applicable as medicinal products. After lumbrokinase treatment for 17 patients with deep vein thrombosis, fibrinogen level decreased from $5.87 \pm 1.47 \text{ g}\cdot\text{L}^{-1}$ to $3.68 \pm 0.82 \text{ g}\cdot\text{L}^{-1}$; platelet aggregation rate dropped from $48.4 \pm 1.98\%$ to $32.6 \pm 0.65\%$, with statistical significance [59]. Ten (10) patients who had coronary artery disease and stable angina were treated with oral lumbrokinase for 30 consecutive days in addition to their standard medical therapy. Six (6) of 10 patients' anginal symptoms were ameliorated with no adverse reaction, including major or minor bleeding, was observed [60]. A trial with 200 diabetes mellitus in patients with microcirculatory impairment showed that using diamicon plus lumbrokinase capsules improved microcirculation without any major adverse reactions, which was significantly better than using diamicon alone [61]. Additionally, earthworm extracts revealed having anti-hypertensive, anticoagulant, and anti-hyperlipidemic activities in animal experiments [62-65].

3.2.3 Anti-microbial/Fungal Activity

Surviving against the exposure of various soil microorganisms, earthworms developed certain immune responses where anti-microbial peptides played an important role [66]. Recent studies have confirmed the anti-microbial and anti-fungal property of earthworm powder or coelomocytic fluid. For

example, dried *Lampito mauritii* and *Perionyx excavatus* earthworm powder showed strong antibacterial resistance against the *S. aureus*, *P. mirabilis*, and *P. aeruginosa* bacterial strains [67]. Anti-microbial peptides may trigger hemolysis, cytotoxicity, hydrolysis, agglutination, and opsonization to defend earthworms [66]. *Lumbricin I*, a proline-rich (15%) antimicrobial peptide with a molecular weight of 7,231 Da, was first isolated from *Lumbricus rubellus* by Cho et al. [68]. Following that, two more peptides, PP-1 (an isomer of *Lumbricin I*) and OEP 3, were identified from *Pheritima schiliensis* and *Eisenia fetida*, respectively [70]. Li et al. [70] found lumbricin-PG from skin secretions of earthworm *Pheretima guillelmi*. Another anti-microbial constituent lysenin (41 kD) can bind to the surface of nanoparticles and promote engulfment by earthworm phagocytes [71]; they can also oligomerize a dactaspore-forming toxin in the plasma membranes [72].

3.2.4 Anti-tumor Activity

A systematic review analyzed the anti-tumor potential of earthworm from databases like Google scholar and PubMed. Over 20 publications about earthworm-derived materials were studied for different cancers, including oral cancer, breast cancer, lung cancer, cervical cancer, colon cancer, others [73]. Mechanisms of earthworm anti-tumor property could be roughly divided into four theories. First of all, active components from earthworms may induce apoptosis-like cell death to mammalian tumor cell [71]. Secondly, the cytotoxicity effect of earthworms may be presented in cell growth inhibition and anti-proliferative activity [73, 74]. Cancer studies related to other insects, such as scorpion centipede, and leech, also suggested the same observation. Extracts from these insects had significant effects on proliferation inhibition of HepG2 cells [75]. The third possible mechanism might be through the enhancement of body immunologic function as mentioned above. A clinical research comparing

cancer patients taking Capsule 912 (a Chinese medicine containing earthworms) revealed that the amount of T3, T4 & T8 cell subsets for 27 patients who took the medicine increased significantly than before while the 25 patients from the control group remained unchanged [76]. Lastly, earthworm fibrinolytic enzyme and other active substances inhibit tumor growth or migration, possibly via matrix metalloproteinases 9 (MMP9) suppression and reduction of the micro-vessel density beside tumors. Mice injected by earthworm protein grew smaller tumors (nasopharyngeal carcinoma) and had less micro-vessel density around in comparison to those controlled mice [77, 78].

3.2.5 Anti-inflammatory and Anti-pyretic Activity

A recent study from Li et al. [79] further reported two novel analgesic and anti-inflammatory peptides purified and characterized from the coelomic fluid of earthworm (*Eisenia fetida*), VQ-5 and AQ-5. AQ-5 can inhibit the mitogen-activated protein kinase signaling pathway involved in analgesic and anti-inflammatory functions. Experiment from Omar et al. [80] showed that the anti-pyretic effect of earthworm ($100 \text{ mg}\cdot\text{kg}^{-1}$) was comparable to standard

drug paracetamol ($150 \text{ mg}\cdot\text{kg}^{-1}$). Since fever is the natural defense of the body, such suppression of the body temperature may come from the anti-inflammatory effects of earthworms.

3.2.6 Osteogenic Effects

G-90, a glycolipo protein extract from earthworm, appears to improve regeneration in bones and tissue. One study showed that earthworm extract given at an appropriate dose increases differentiation and proliferation of osteoblasts. Thus, future application includes bone tissue regeneration [81]. Another study concluded that wound healing was better treated with G-90 in mice [82].

4. Earthworm Powder and Marketing Concerns

4.1 Extraction of Earthworm Protein Powder

Fig. 1 presents the brief production process of Bocom earthworm protein powder. Bocom extraction uses simple materials (only earthworms and water) and different level of filtration technologies to obtain earthworm proteins. Fresh or dried earthworms will be selected and washed and soaked afterwards with

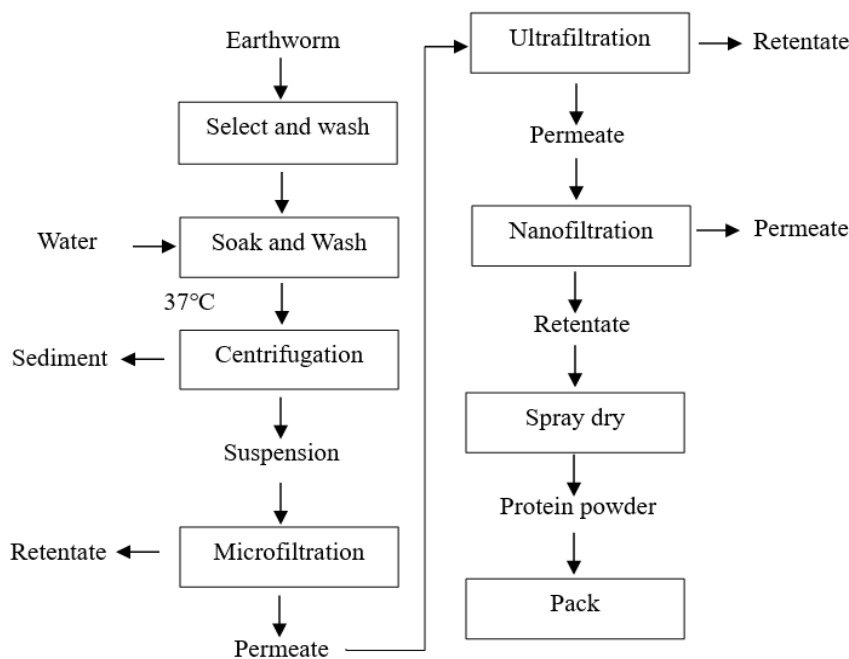


Fig. 1 Extraction of Bocom earthworm protein powder (personal communication).

drinking water for 5 hours. Such mixture will then be incubated at 37 °C and stirred in the tank for another 14 hours. Suspension from the mixture centrifugation will go through several filtrations (micro-/ultra-/nano-), retentate after nanofiltration will be spray-dried to produce protein powder. Such crude protein powder is already a great protein supplement (mix with beverages or food) to fulfill our daily nutrient requirements. Moreover, it can be used as an ingredient to produce composite products with other ingredients. For instance, FULI Capsule, another product from Bocom, is mainly made of earthworm protein, soy protein and taurine, aiming to enhance immunity and alleviate physical fatigue.

4.2 Safety Issues

Concerning the safety issue, Orozco et al. [83] proposed that earthworm proteins can be potential supplements in human and animal diets without risk of contamination by infectious agents or metals. Their experiment showed that there was no adverse physiological effect to substitute 30% daily protein of growing rabbits with earthworm meal (*Eisenia fetida* and *Lumbricus rubellus*). Earthworms often contain elevated levels of metal because of the assimilation of metals from their environment; however, fish and chicken fed with earthworms recorded no significant increases in heavy metals was found in their carcasses. According to Medina et al. [16], heavy metal levels were low in oven-dried earthworm meal, with 2.5 ppm Pb and 0.08 ppm Hg respectively (2 ppm Pb and 0.5 ppm Hg in tuna fish). In addition, they also did CEM174 human cell line test, and 96% viability was observed at 0.75 µg per well of earthworm protein, which revealed that *Eisenia fetida* proteins were not toxic at low concentrations. *In vivo* results revealed that there was no toxic reaction after continuously injecting earthworm extracts into the caudal vein and abdominal cavity of mice [84]. According to Bocom toxicological safety evaluation (personal communication), no effects were observed from rats

of the experimental group with maximum earthworm protein powder intake (8 g·kg⁻¹ body weight) after 90 days. Maternal toxicity, embryotoxicity, and teratogenic effects were not observed from healthy pregnant rats under the same feeding dose.

4.3 Acceptability

Long and slimy animals like earthworm may trigger people's "disgust" feeling [26]. Such negative concepts usually result in a natural aversion for earthworm consumption, which can be reinforced by the abundance of other available protein sources (livestock, fish, etc.). After all, culture plays a vital role in people's perception with regards to food preferences. In general, tropical and subtropical inhabitants have higher acceptability for earthworm eating. For the western society, although some terrestrial or marine invertebrates, such as snails and oyster, are considered as delicacies, consuming earthworm is not as popular [26]. Another obstacle for promoting earthworm products is its unpleasant smell. However, recent researches started to explore the possibilities of fortified earthworm products. Cayot et al. [85] found arepas (a maize-based pancake) fortified not more than 5.5% earthworm protein powder was satisfactory in terms of sensory. Bou-Maroun & Cayot [86] reported lipid oxidation to be the main origin of odor-active compounds (OAC) in earthworm powder. They eliminated 97.7% of the OAC and 93% of the volatile compounds arising from lipid oxidation through ultrasound delipidation. Though lipid depletion reduced the odor considerably, lipid depleted earthworm powder was not completely odor neutral. Some lipids still remained and may evolve during storage to produce off-flavors [87, 88]. Their further stability experiments concluded that temperature is the main parameter for the long-term stability of deamortized powder among storage conditions including light, gas (nitrogen or air storage), and temperature [88]. In order to enhance the acceptability of earthworm powder in food products,

flavoring and delipidation may be possible solutions. With no loss in acceptability, cocoa cookies can be fortified with 3.9% of *Eisenia fetida* proteins while cocoa/cinnamon cookies with 5.2%. Acceptability of unflavored cookies fortified with raw protein powder and deodorized powder were significantly different up to a protein substitution level of 5% [87].

4.4 Prospective

Nutritional and medicinal values of earthworms make them excellent natural and novel sources as dietary supplements. Developing different formulations (capsule, powder, liquid form, etc.) and flavors instead of selling whole earthworms may help to eliminate the negative perception of consuming worms, increase product acceptability, and thus expand the market. Furthermore, earthworm-derived products containing isolated or purified components with certain functions and activities may also be an appealing direction due to their health benefits.

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