

Edible Bird's Nest, a Valuable Glycoprotein Source: Current Research Prospects and Challenges in Malaysia

(Sarang Burung Walit, Punca Glikoprotein Bernilai Tinggi: Prospek Penyelidikan dan Cabaran Semasa di Malaysia)

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Received: 6 January 2022/Accepted: 14 April 2022

ABSTRACT

Edible bird's nest (EBN) is a salivary secretion of swiftlets which consist of protein and carbohydrate rich glycoproteins. This natural ingredient is very valuable, nutritional and medically valuable. The EBN industry have grown rapidly and benefited the Malaysian economy, hence, it is viewed seriously and it is actively supported by the government. This review discusses the progress and development of EBN industry as well as the R&D activities and endeavours especially that which involves deriving peptides with biological activities from EBN and its by-product sources. Many studies have documented the therapeutic properties of EBN such as antiaging, antiviral, antioxidant, and antihypertensive. Studies have also been conducted to produce glycoprotein hydrolysates from EBN through enzymatic hydrolysis, and findings showed that these bioactive peptides increase solubility as well as antioxidant and antihypertensive activities. Enzymatic hydrolysis breaks long protein chains at specific sites and releases amino acids and small peptides with lower molecular weights. The EBN hydrolysates produced can improve bioactivity and overcome insolubility and low absorption of EBN prepared and consumed through traditional means. Further studies need to be carried out to optimise EBN glycoprotein hydrolysates production as well as maximising their bioavailability and efficacy in the human gastrointestinal system. In addition, EBN by-products produced during EBN cleaning process should be fully utilised to recover the high-value glycoproteins, while reducing pollution and wastage. By enhancing R&D activities of EBN, bioactive glycopeptides produced from EBN may become an important functional food ingredient for various uses and innovative value-added products in the future.

Keywords: Bioactive peptides; edible bird's nest; glycoprotein; hydrolysis; value-added product

ABSTRAK

Sarang burung walit (SBW) adalah rembesan air liur burung walit yang terdiri daripada glikoprotein yang kaya dengan protein dan karbohidrat. Bahan semula jadi ini sangat berharga, berkhasiat dan mempunyai nilai perubatan. Perusahaan industri SBW telah berkembang pesat dan menguntungkan ekonomi Malaysia, justeru, ia dipandang serius dan disokong secara aktif oleh kerajaan. Ulasan ini membincangkan tentang pengembangan dan pembangunan industri SBW serta aktiviti dan usaha R&D terutamanya yang melibatkan penghasilan peptida dengan aktiviti biologi daripada SBW dan produk sampingannya. Pelbagai kajian telah mendokumentasikan sifat terapeutik SBW seperti antipenuaan, antivirus, antioksidan dan antihipertensi. Banyak kajian juga telah dijalankan untuk menghasilkan hidrolisat glikoprotein daripada SBW melalui hidrolisis enzim dan hasil kajian mendapati bahawa peptida bioaktif ini meningkatkan kadar keterlarutan serta aktiviti antioksidan dan antihipertensi. Hidrolisis enzim memecahkan rantai protein yang panjang di tapak yang khusus dan membebaskan asid amino dan peptida kecil dengan berat molekul yang lebih rendah. Hidrolisat SBW yang terhasil dapat meningkatkan bioaktiviti dan mengatasi masalah ketidaklarutan dan penyerapan SBW yang rendah dalam kaedah penyediaan dan pengambilan SBW secara tradisi. Kajian selanjutnya harus dijalankan untuk mengoptimalkan

proses penghasilan hidrolisat glikoprotein SBW serta memaksimumkan bioketersediaan dan keberkesannya pada sistem gastrousus manusia. Selain itu, produk sampingan EBN yang terhasil sewaktu proses pembersihan harus dimanfaatkan sepenuhnya untuk memperoleh semula glikoprotein bernilai tinggi, sekaligus mengurangkan pencemaran dan pembaziran. Dalam usaha mempergiat aktiviti R&D SBW, glikopeptida bioaktif yang dihasilkan daripada SBW boleh menjadi bahan makanan berfungsi yang penting untuk pelbagai kegunaan dan produk nilai tambah yang inovatif pada masa hadapan.

Kata kunci: Glikoprotein; hidrolisis; peptida bioaktif; produk nilai tambah; sarang burung walit

INTRODUCTION

The Malaysian swiftlet industry plays an important role in contributing to the national economy. Swiftlets are unique creatures where their nests are made using their own saliva. Edible bird's nest (EBN) has been traded for hundreds of years between the Malay Archipelago and China since the 16th and 17th centuries. EBN harvesting in Malaysia traditionally began in the late 1800s in limestone caves in the Borneo Islands, such as Niah Cave and Mulu Cave in Sarawak, and Mandai Cave and Guamantong Cave in Sabah (Gausset 2002; Hobbs 2004; Ismail 1999; Lim & Cranbrook 2002).

The EBN-producing swiftlet species that are mainly traded worldwide are *Aerodramus fuciphafus* and *Aerodramus maximus*. They produce white and black EBN, respectively, and are available in Southeast Asian countries (Lim & Rahman 2005; Marcone 2005). The EBN industry is an example where the locals take advantage of the diverse bird species in Malaysia and subsequently grow into a commercial business (Mansor & Rahman 2013). Over time, EBN farming, processing and operations have emerged as a popular industry in Malaysia (Lim & Cranbrook 2002). Swiftlet farming has become a lucrative trade and the booming of the swiftlet industry has generated huge income to the population in Southeast Asian countries, especially in Indonesia and Malaysia (Thorburn 2014).

Over the last few decades, there are a lot of interest generated in the EBN industry and academia, where there were a lot of focus given into enhancing the industry through public-private collaboration, mainly mediated by the Malaysian government. However, the economic and policy data on EBN are somewhat fragmented. Aside from that, the harvested EBN have to go through tough cleaning process to remove impurities such as sand, feathers, and eggshells. During the process, a relatively huge amount of EBN by-products, which consist of these impurities that may have valuable glycoproteins attached to them, are usually discarded. Disposal of these waste material without further treatment is hazardous to the environment and also a huge economic loss, since the

by-products still comprises considerable amounts of glycoprotein and essential trace element that has potential applications. Besides that, almost all studies reported findings which were conducted on clean EBN, whereby the harvested bird's nest was processed to remove impurities. The properties of EBN processed by-products have not been studied and reported. Therefore, this paper reviews the economic value and governmental policy related to EBN, as well as discussing the topic on EBN processing by-products as a potential alternative source of valuable EBN glycopeptides.

PROSPECT AND DEVELOPMENT OF EDIBLE BIRD'S NEST INDUSTRY

The National Agro-Food Policy has identified the EBN industry as one of the seven high value agricultural industries (Ministry of Agriculture and Food Industries Malaysia 2011). The Malaysian government also specifically named the EBN industry as a high-impact agricultural sector activity under the new economic resource development plan to achieve the goal of making Malaysia a developed country with a high Gross National Income (GNI) (Mohd Syauqi et al. 2020). The industry has the potential to develop further and generate national revenue sources. Emphasis is given towards increasing the contribution of agriculture to Gross Domestic Product (GDP) and farmers' income, including producing high value products to meet the demands of specific markets and high-income consumers (Ministry of Agriculture and Food Industries Malaysia 2011).

EBN is considered as the country's new source of wealth in the field of agriculture and EBN's industrial enterprises are worth millions of Malaysian Ringgit (MYR). Malaysia is the world's third most important producer of EBN after Thailand and Indonesia (Mohd. Rashid 2016). Unprocessed EBN can be worth between RM 2,000/kg and RM 6,000/kg while cleaned nests are sold at prices between RM8,000/kg and RM20,000/kg. Malaysia's EBN products are in high demand by countries such as China, Hong Kong, and Taiwan. In 2019, total exports of EBN and its related products were

5,654 metric tonnes worth RM1.154 billion, while in 2020, total exports were 3,640 metric tonnes worth RM1.385 billion (Mohd Syauqi et al. 2020). As of April 2021, the industry's total exports have reached 962 metric tonnes with a value of RM714 million, indicating that exports of this commodity remain strong especially while the country is still facing the spread of the COVID-19 pandemic (Department of Veterinary Services Malaysia 2021).

The EBN industry in Malaysia is under the jurisdiction of the Malaysian Department of Veterinary Services (DVS) which is responsible for monitoring and coordinating all swiftlet breeding and harvesting activities, as well as controlling import and export of animal products including EBN. A total of 276 EBN processing plants and 2,516 EBN premises nationwide were registered with the DVS between 2014 and 2015, where the number of registered premises increased at an average rate of 75.7% per annum from 2009, with 146 processing plant premises (Mohd. Rashid 2016). As of June 2021, a total of 12,954 EBN premises have been registered with the DVS (Department of Veterinary Services Malaysia 2021). EBN has become a popular farming industry, and the entrepreneurial opportunity in it is very wide and guarantees a very lucrative return. As the contribution of the EBN industry to the national economy shows an increase, more and more people want to get involved with such business. By considering the appropriateness of the potential of this industry based on economic value, the government seeks to empower this industry through various supports and reinforcements (Mohd. Rashid 2016).

In order to ensure that the EBN industry flourish, planned development needs to be implemented, as outlined by the National Agro-Food Policy (2011-2020) (Ministry of Agriculture and Food Industries Malaysia 2011). Among them is focusing on R&D activities in the development of a competitive EBN industry. In the recent National Agro-Food Policy 2.0 (2021-2030), EBN has been specifically named as one of the High Value Commodities (HVC) (Ministry of Agriculture and Food Industries Malaysia 2021).

Such R&D activities are also in line with the government's policy, the Vision for Shared Prosperity 2030, under the Key Economic Growth Activities (KEGA), namely, Halal & Food Hub (KEGA 6), Malaysian Commodities 2.0 (KEGA 7) and Smart and High Value Agriculture (KEGA 13) (Ministry of Agriculture and Food Industries Malaysia 2021, 2011). This is also parallel to the *Entry Point Project* (EPP) implemented by the Department of Veterinary Services (DVS) under

the National Key Economic Areas (NKEA) whereby the EBN project is made to be one of the major projects with a high export target (Mohd. Rashid et al. 2013).

This proves that EBN is recognised as a 'high value agricultural commodity', where its growth should be prioritised and driven, as the EBN industry is expected to make the highest contribution compared to other EPP projects to the national economy by generating revenue to the country. The development and promotion of these premium-priced commodities will increase demand for quality and availability in local and export markets. Therefore, EBN's research and innovation development activities are very important in EBN's upstream and downstream industries to reduce production costs, increase productivity as well as produce and market high value products to consumers of all walks of life.

Overall, the Malaysian government has positioned the EBN industry as the main premium industry in Malaysia and devoted a large amount of funding to ensure this industry continues to thrive. The government, through its relevant agencies, constantly monitors the EBN industry and assists entrepreneurs in their efforts to strengthen the production and quality of EBN in Malaysia.

EDIBLE BIRD'S NEST AS A FUNCTIONAL FOOD WITH HEALTH BENEFITS

Keeping pace with changing times, and the occurrence of the COVID-19 pandemic has made people more aware about maintaining their health and taking nutritional supplementary products. This could be one of the reasons for the increasing demand for commodities such as EBN, especially among those with high incomes who can afford it. Because of its efficacy and ability to increase the quality of health, EBN is very suitable to be consumed to strengthen the body's immune system. A functional food is a food that provides advantages in addition to its basic nutritional value (Lim et al. 2021). EBN contain proteins and glycoproteins which are found abundantly and have the potential to provide nutritional and therapeutic properties (Nurul Nadiah et al. 2021). Thus, EBN is considered a functional food and is taken for consumption for the added health benefits it offers.

CHEMICAL COMPOSITION OF EDIBLE BIRD'S NEST

Proximate analysis by several studies is summarized in Table 1. The findings of previous studies show that the main components in EBN are carbohydrates and proteins, while lipid and ash are present in low amounts. Kathan and Weeks (1969) reported that the carbohydrate in EBN

comprise of sialic acid (9%), galactosamine (7.2%), glucosamine (5.3%), galactose (16.9%) and fucose (0.7%). Tung et al. (2008) reported similar composition of 10.8% sialic acid, 4.19% galactosamine, 5.29% glucosamine, 5.03% galactose, 0.44% fucose, 0.20% rhamnose and 0.75% manose. The presence of sialic acid in EBN was also reported in most of these studies.

In addition, amino acid analyses were also carried out and findings of essential and nonessential amino acids in EBN have been stated (Halimi et al. 2014; Kathan & Weeks 1969; Marcone 2005; Saengkrajang et al. 2013; Wang 1921b). All 18 amino acids were also

reported to be present in EBN (Ali et al. 2019) including the 9 essential amino acids required for tissue growth and restoration, namely phenylalanine, valine, threonine, histidine, tryptophan, isoleucine, methionine, lysine, and leucine (Daud et al. 2021a).

The different results in content and composition of EBN from the studies may be caused by several factors, such as geographical location, species and type of EBN as well as different extraction processes. The difference in the amount of saccharide and sialic acid content could be due to the different EBN hydrolysis processes, whether using acid or alkali.

TABLE 1. Summary of studies of the chemical composition of EBN from different location sources

Reference	Location source	Carbohydrate	Protein	Fat	Ash	Moisture	Sialic Acid
Kathan & Weeks (1969)	China		32.3%		20%		8.6%
Marcone (2005)	Malaysia and Indonesia	27.26%	62%	0.14%	2.10%	7.50%	
Tung et al. (2008)	Taipei						10.8%
Huda et al. (2008)	Penang and Indonesia	27-58%	24-49%	0.47-1.99%	2.75 - 7.53%	13.77-20.20%	
Norhayati et al. (2010)	Peninsular Malaysia		61.5 g/ 100 g				0.7-1.5%
Saengkrajang et al. (2013)	Thailand	25.4–31.4%	61.0-66.9%	0.4-1.3%	5.9-7.4%	17.8-24.3%	
Nurfatin et al. (2014)	Pahang and Terengganu	22.28% & 25.79%	58.55% & 55.48%	0.67% & 0.29%	2.60% & 2.57%	15.90% & 15.87%	
Hun et al. (2015)	Johor	46.47%	35.8%	1.5%	5.17%	11.27%	
Zainab et al. (2016)	Malaysia	11.3-12.9%	58.3- 63.9 %	0.05-0.09%		7.0-9.34%	
Hidayati Syamimi et al. (2018)	Malaysia	18.98 - 26.32%	54.29 - 60.59%	0.43 -1.19%	1.85 - 10.7%	12.28 - 16.62%	
Quek et al. (2018)	Malaysia	23.2 - 26.2 g/ 100 g	66.1 -68.4 g/ 100 g	0.02 -0.19 g/ 100 g	6.6 -9.9 g/ 100 g	16.9 - 19.4 g/ 100 g	7.2 - 13.6 g/ 100 g
Huang et al. (2019)	Malaysia and Indonesia	22% - 38%	44% - 54%				0.08 - 0.12 g/ 100 g
Tan et al. (2021)	Malaysia	27.97 - 31.68%	53.03 - 56.37%	0.1%	2.22 - 3.38%	10.8 - 14.04%	

EDIBLE BIRD'S NEST GLYCOPROTEIN

Glycoproteins are products of post-translational glycosylation of protein molecules and are important bioactive molecules because protein glycosylation adds a dimension of protein biological activity that is widely available in most organisms (Spiro 2002). EBN is a highly valuable glycoprotein (Kathan & Weeks 1969), as glycoprotein is rarely found in such large quantities. Previous studies have shown that EBN has carbohydrate and protein properties similar to salivary mucin (Wang 1921b).

The structure of the mucin glycoprotein of EBN salivary glands was found to consist of a polypeptide backbone and sugar side chains of N- and O-glycosylproteins which are natural sources of carbohydrate-rich material (Wieruszski et al. 1987). Mucin glycoprotein is a major and important component in EBN and has a very high molecular weight on the surface of epithelial cells and is densely glycosylated with O-linked glycans involved in various cell functions (Barchi Jr. 2013).

According to You et al. (2015), EBN glycoproteins are well known for their high sialic acid content. Sialic acid belongs to a family of nine-carbon acidic monosaccharides that occur naturally at the ends of the sugar chains attached to the surfaces of cells and soluble proteins (Wang & Brand-Miller 2003). Sialic acid, which has this carboxyl group, provides a negative charge to glycoproteins which contributes to the physiological effects on biological systems (Varki 2008). The main molecular species of sialic acid in EBN was found to be N-acetylneuramic acid (Houdret et al. 1975), which was also found in high concentrations in the human brain (Wang & Brand-Miller 2003).

Sialic acid is a high value monosaccharide that attracted much attention from researchers to detect and isolate it. Apart from the earliest studies that found sialic acid in EBN extracts (Howe et al. 1961, 1960; Kathan & Weeks 1969), there are other studies that have detected sialic acid such as the determination of N-acetylneuramic acid in sialic mucoids (Aminoff 1961), isolation N-acetylneuramic acid from EBN hydrolysates (Pozsgay et al. 1987), and enzymatic synthesis and release of N-acetylneuramic acid from submaxillary mucin (Comb & Roseman 1960, 1958). While these few past studies have reported methods to detect sialoglycoproteins in EBN, more studies for the isolation and determination of the structure and composition of glycoproteins in EBN should be carried out to elucidate and characterize sialoglycoprotein bioactivities in EBN. Similarly, there

should be additional studies that can help identify the potential biologic properties of sialoglycoproteins between EBN and diseases in human.

HEALTH BENEFITS OF EDIBLE BIRD'S NEST

Hobbs (2004) and Marcone (2005) stated that EBN is considered a luxurious and high-value food because of the public's perception of its efficacy and pharmacological characteristics as a food that increases bodily health and functions. Consuming EBN soup is said to be effective in reducing phlegm, relieving sore throats, reducing gastric problems, helping kidney function, increasing libido, beautifying skin, reducing asthma, relieving coughs, curing dry coughs, strengthening the immune system, accelerating recovery from diseases and surgery, and increasing energy, stamina, and strength (Hobbs 2004).

Much researches have been carried out to assess the therapeutic properties of EBN. It has been reported that EBN can prevent influenza virus outbreaks by neutralising the virus and inhibiting red blood cell aggregation (Guo et al. 2006). Bone strengthening properties of EBN has also been reported (Hou et al. 2021; Matsukawa et al. 2011). In addition, EBN was also found to possess wound healing activity in human skin keratinocytes and fibroblasts (Hwang et al. 2020). EBN also showed positive effects on cholesterol metabolism in HepG2 cells, thus demonstrating the potential for cardiovascular health (Akmal et al. 2020).

Sialic acid in EBN and is found to have many biological functions that are important for human health benefits, such as neuronal development (Khalid et al. 2019), antiviral activity against influenza viruses (Shi et al. 2017), prevention of hypertension (Wang et al. 2019), a strong immune regulator of cancer cells (Zhao et al. 2016), and skin whitening effect (Chan et al. 2015). Sialic acid can be used as an ingredient or functional supplement in many industries, especially the nutraceutical, pharmaceutical, and food industries (Wong et al. 2018). The study by Marni et al. (2014) also showed the presence of free sialic acid in EBN from Malaysia, which is very important for infant brain development (Colombo et al. 2003).

EBN is also being studied as an anti-aging agent to maintain good health and slow down the aging process and diseases (Chua et al. 2013; Hou et al. 2015; Yew et al. 2019). According to Careena et al. (2018), EBN significantly improves memory and confirms neuroprotective activity against neuroinflammatory inhibition and oxidative stress processes. Therefore, EBN can be a potential agent in the treatment of age-

related diseases, and sialic acid from EBN is useful as a health-promoting food that enhances the suppression of neurodegenerative diseases. Overall, these studies demonstrate the potential of EBN in improving the health conditions of consumers.

PRODUCTION OF PEPTIDE HYDROLYSATE FROM EDIBLE BIRD'S NEST

Traditionally, EBN is prepared for consumption by double boiling. However, most of the proteins, which account for as much as 50% of the dry weight of EBN, remain insoluble and provide a low protein extract rate of only ~5% (Wong et al. 2017). EBN does not dissolve easily by extracellular fluids because of its high charge density, high molecular weight protein and grouping of glycosylation sites (Dai et al. 2021). This results in low solubility and digestibility of EBN, and the anti-digestive properties of EBN may be related to the sugar chains attached to the ends of the peptide chains (Dai et al. 2021). Raw EBN which have not been processed contain large size and molecular weight proteins. Previous studies reported a wide range of molecular weight proteins in EBN (Guo et al. 2017; Utomo et al. 2014; Xu et al. 2019). Moreover, consuming double boiled EBN may not

adequately absorb enough of the nutrients into the body, and the exact goodness and properties of EBN might not be fully experienced.

The conversion of EBN glycoproteins to glycopeptides may help overcome this problem and increases the efficiency of their uptake. One of the ways this can be done is by hydrolysis treatment which can produce hydrolysate peptides with reduced molecular weight. Peptides can be hydrolysed by specific enzymes that can break down identified segments of the parent protein (Daliri et al. 2017) and release small peptides and free amino acid. Peptides with low molecular weights (<10 kDa) were found to be more effective as antioxidative and antihypertensive peptides compared to those with higher molecular weights (Daliri et al. 2017). Small bioactive peptides produced by enzymatic hydrolysis can increase the quality of biological function and activity of EBN (Gan et al. 2020), thus, help overcome absorption issues and give positive effects towards bodily functions and health.

HYDROLYSIS OF EDIBLE BIRD'S NEST

Enzymatic hydrolysis is a commonly used method for extracting glycoproteins in many studies, some of which are summarised in Table 2. Nurul Nadia et al. (2017)

TABLE 2. Summary of some studies on EBN hydrolysate produced through enzymatic hydrolysis

Reference	Analysis conducted		Raw EBN extract	EBN glycopeptide hydrolysate
Nurul Nadia et al. (2017)	Degree of hydrolysis		~14%	64-91%
	Molecular weight		51.3-123.0 kDa	<51.3 kDa
Nurfatin et al. (2016)	Degree of hydrolysis		7.6%	71.2-82.7%
	Peptide content		9.7%	62.02-86.68%
	Protein solubility		1.475%	81.9-104.1%
	Antihypertensive (ACE-i) activity		6.88%	86.24%
Etty Syarmila et al. (2014)	Degree of hydrolysis		~79%	86.5%
	Peptide content		27.7 mg/g	109.5 mg/g
	Protein solubility		25.5 mg/g	163.9 mg/g
Ramachandran et al. (2018)	Degree of hydrolysis		<70%	79.48 -82.33%
	Peptide content		59.48%	67.86-68.70%
	Antihypertensive (ACE-i) activity		<80%	80.22-86.97%
Ali et al. (2019)	Antioxidant activity	DPPH	35.0%	45.0%
		ABTS	81.5%	87.0%
Zulkifli et al. (2019)	Degree of hydrolysis		<12%	14.3-16.3%
	Protein solubility		~4.0 mg/g	313.50-607.5 mg/g
	Antioxidant activity	DPPH		10.4-29.9 %

reported that EBN hydrolysate produced by the hydrolysis of protease enzymes showed an increase in the degree of hydrolysis and low molecular weight yield compared to unhydrolysed EBN. Etty Syarmila et al. (2014) and Nurfatim et al. (2016) also reported a higher degree of hydrolysis, peptide content, and protein solubility activity after enzymatic hydrolysis treatment in EBN. EBN hydrolysate produced by the hydrolysis of the alcalase enzyme also has higher antioxidant activity than EBN crude extract, as reported by Ali et al. (2019) and Zulkifli et al. (2019).

Besides enzymatic hydrolysis, there are also other methods conducted by different studies to obtain protein hydrolysates, either from EBN or other sources. They include methods by physical means such as microwaving (Lee et al. 2016), sonicating (Jin et al. 2021; Mintah et al. 2019) and chemical methods such as acidic hydrolysis (Kong et al. 2016). The solubility, type, and biological activity or bioactive properties of peptides and the amount of sialic acid content may be affected by different hydrolysis and extraction methods.

Overall, the conversion of EBN glycoproteins to glycopeptides through a suitable hydrolysis method proves that the resulting bioactive peptides have an impact on bioactivities, such as antioxidants and antihypertensives, thereby increasing the solubility, function, and ingestion of EBN for widespread use in various industries (Daud et al. 2021b; Yan et al. 2021). Thus, hydrolysis methods other than enzymes or a combination of methods to obtain EBN glycopeptides should be studied as well. Such studies could show the mechanisms underlying the effectiveness of the bioactive properties of EBN and introduce new methods of enhancing its properties. The positive results of the investigations have the potential to be advanced to the stage of clinical studies and production of products or functional foods that have pharmacological effect on human health.

BIOACTIVE PEPTIDES OF EDIBLE BIRD'S NEST

Bioactive peptides are part of a specific protein chain that is inactive in the parent protein sequence (Sarmadi & Ismail 2010) and is formed by amino acids held together by covalent bonds, or also known as amide or peptide bonds (Sánchez & Vázquez 2017) which usually consist of 3-20 amino acid (Manikkam et al. 2016). When these inactive bioactive peptides are liberated by enzymatic hydrolysis, they will be able to carry out various physiological functions and thus improve health (Möller et al. 2008).

Research on the development of peptides derived through enzymatic proteolysis having high nutritional characteristics, functional biological activity, as well as great solubility, breakdown, and biocompatibility is ongoing and promising (Yao et al. 2021). Bioactive peptides derived from food protein hydrolysis have shown functional and bioactivities potential. For example, bioactive peptides exhibiting antihypertensive activity as inhibitors of the angiotensin-converting enzyme I (ACE-i) have been obtained and isolated from the hydrolysis of protein sources such as meat, fish, and beans (Jang & Lee 2005; Lee et al. 2010; Sitanggang et al. 2021; Wang et al. 2013).

Since EBN is also known to be a food source of protein, bioactive peptides from enzyme-treated EBN exhibit antioxidant activity (Ghassem et al. 2017; Nurul Nadia et al. 2015), antihypertensive activity (Nurfatin et al. 2016; Ramachandran et al. 2018), and prebiotic activity (Babji et al. 2018). Increase in the function and nutrition of EBN, especially in the aspect of bioavailability of EBN glycopeptide have also been identified (Amin et al. 2019; Azmi et al. 2021; Gan et al. 2020; Noor et al. 2018; Yan et al. 2021).

Henceforth, it is proven that enzymatic hydrolysis treatment can produce EBN peptide hydrolysate and improve functional characteristics in EBN, which has the potential as a source of natural bioactive ingredients that can be used in healthy food formulations to reduce the effects of the oxidation process on the body (Etty Syarmila 2019) and to increase beneficial microbes in the gastrointestinal system (Babji et al. 2018). Thus, this functional ingredient of EBN can be applied in food products such as ready-to-drink products, prebiotic drinks, yogurt and ice cream enriched with EBN hydrolysate. Consequently, there is potential for more research on the enhancement of EBN bioactive hydrolysates that can be easily digested to maximize their delivery, absorption and effectiveness to target organ systems of the body.

PROCESSING OF EDIBLE BIRD'S NEST

The EBN industry faces challenges from various levels and requires the best approaches to overcome them. Among the challenges faced are increasing competitiveness, enforcement of laws and regulatory compliance systems, quality control and assurance, safety and authenticity, as well as issues regarding pollution during processing and optimisation of industrial waste (Mohd. Rashid 2016).

Beliefs about the efficacy yet difficulty of obtaining EBN raw materials makes it a food commodity of an exotic value that is high in demand and high in market

prices. In addition, the high demand for EBN is caused by public awareness about its nature and function as a food that improves health. However, it also shows that EBN is a premium food item that is expensive, not easily accessible, and cannot be afforded by everyone. The very high price of EBN is not only contributed by the dangerous methods of obtaining the nest, but also the high processing cost, due to the difficult and tedious cleaning procedure (Tagliacozzo & Chang 2011).

According to Azmi et al. (2021), in the early 70's, the EBN cleaning methods that was carried out in the industry was secret information. However, as bird nest houses became more and more popular in the 80's, EBN cleaning became an industrial necessity. Initially, an effective cleaning method without considering nutrient changes in EBN was carried out by trial and error method due to lack of R&D activities.

Yeo et al. (2021) stated that unclean EBN harvested from swiftlet caves or bird houses do not go through any cleaning process and are directly sorted, dried, graded, trimmed, weighed and packed, while clean EBN are raw EBN that have gone through cleaning processes of sorting, soaking, picking of feathers and impurities, moulding, drying, grading, and packing (Department of Standards Malaysia 2010). The cleaning stage determines the various grades of EBN which also determines the market price of EBN. EBN is assessed based on dry mass, size, colour, dirtiness, and number of feathers through physical appearance (Lee et al. 2021).

In terms on regulations and standard operating procedures in Malaysia, the processed EBN needs to conform to the Food Regulations 1985 (Regulation 178A), as well as the standard specified in Malaysian Standard on Edible-Birdnest (EBN) – Specification (MS 2334:2011). Aside from that, the Food Safety and Quality Division, Ministry of Health Malaysia has put forward a Standard Operating Procedure (SOP) on the Control of the Safety of Raw Edible Bird's Nest along the Food Supply Chain; and the Standard Operating Procedure on the Control of Nitrite Level in Edible Bird's Nest. All these regulations, standards and SOPs are in place to ensure the quality and safety of the processed EBN, rendering it fit for human consumption.

CLEANING PROCESS OF EDIBLE BIRD'S NEST

One of the problems identified and is of research interest regarding the cleaning process in the EBN industry is the generation of by-products during the EBN cleaning process. Before raw EBN can be used and commercialised,

the unclean harvested EBN must be cleaned to remove impurities such as sand, eggshells, bird's feathers, and other foreign materials. The major concern and procedures during cleaning that will contribute to the quality of EBN are the use of chemicals such as hydrogen peroxide; practice of hygiene condition to reduce contamination and growth of bacteria and fungi; the reduction of the amount of nitrates and nitrites; the removal of all other contaminant and impurities; the preservation with minimal degradation of nutrients.

The EBN cleaning process is a laborious one, and the process produces large amount of by-products that are usually just thrown away. The EBN by-products, which consist mainly of bird's feathers, still contain the valuable glycoproteins and important elements attached to it, and these should not be wasted. Its disposal without further treatment results in huge economic loss and may also cause environmental pollution. This EBN by-product is of great interest to study and to be used as a high value functional ingredient for various applications.

EDIBLE BIRD'S NESTS BY-PRODUCT

There are many studies and investigations that have been carried out on clean EBN. However, studies on by-products of EBN (Figure 1) are very limited. There are several studies conducted on the feathers of the swiftlet bird, among them the method of genomic DNA extraction (Wang et al. 2013) and keratin (Badrulzaman et al. 2021). Another study was about EBN by-products composition to add value to the formulation of facial products (Hamzah et al. 2016) and EBN by-products properties after going through enzymatic hydrolysis methods (Ling et al. 2020; Ng et al. 2020). Ling et al. (2020) and Ng et al. (2020) both reported that EBN hydrolysates obtained from EBN by-products have the same physicochemical properties as clean EBN and have high protein, amino acid and N-acetylneuramic acid contents and exhibit antioxidant activity.

Besides studies on the feathers of the swiftlets, there are studies reported on various treatments on feather waste of other birds, such as chicken and duck, which are a by-product of animal husbandry and agriculture (Ben Hamad Bouhamed et al. 2020; Cheong et al. 2018; Maciel et al. 2017). Feather waste has a high concentration of keratin proteins and amino acids, making it a possible resource for recovering valuable products (Cheong et al. 2017). They can be converted into bioactive protein hydrolysate with the potential to be useful in human nutrition and health (Callegaro et al. 2019, 2018). Feather

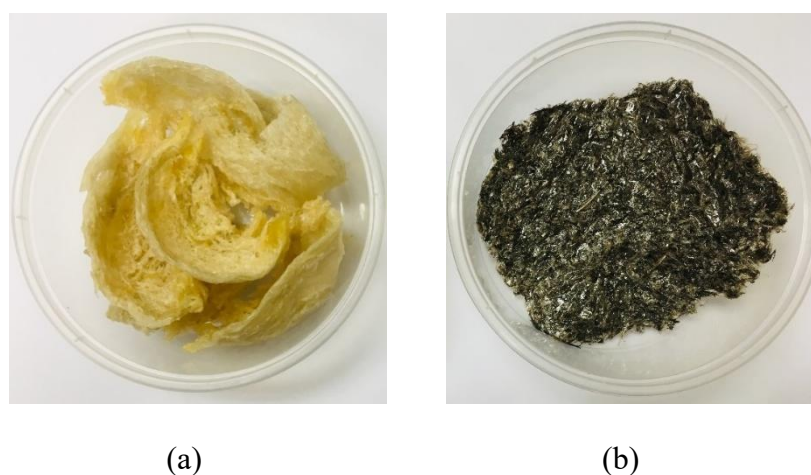


FIGURE 1. (a) Processed clean EBN and (b) EBN by-product

protein hydrolysates can be used as bio-organic fertilizer for agriculture (Bhari et al. 2021) and can become a future source of alternative food and value-added products (Chaturvedi et al. 2021).

The studies carried out on the bird feathers waste can be used as a guide and a benchmark for more studies to be carried out on the by-products of EBN, especially on the waste of the feathers of swiftlet birds, which make up the lot of EBN by-product. Consequently, the management of agricultural or farming by-products requires a more practical approach of converting or recycling waste materials into value-added secondary products. Through this approach, the by-products can be fully utilised and the agricultural and farming industry can reduce pollution and move towards the practice of zero waste.

The recovery of functional products from EBN industrial waste, which is of lower cost and is more competitive in terms of price and can be enjoyed by the community is one of the efforts in line with EBN industry strategy which is included in research activities. This could benefit the EBN industry as a solution to reduce the amount of waste products, at the same time the quality of the recovered compounds can be maintained and applied in potentially functional food products.

CONCLUSION

In this review, we have discussed the prospects of the EBN industry for development and economical advantage in Malaysia. With efforts to empower the industry by the government, the EBN industry has the potential to contribute to the national income. Following the current

trend, the focus on R&D activities is very important for the Malaysian EBN industry to compete and stay ahead as the leading EBN supplier and producer on the world stage. The encouragement to conduct more studies and research is due to the recognition of the nutrients and nutrition of EBN to the level of health as well as its effects on the human body. However, more studies should be done to optimize the production of glycopeptide hydrolysates from EBN and to unravel the potential interactions between EBN bioactive peptides and other components within EBN. In addition, hydrolysis processes and other approaches to increase the bioactivity and bioavailability of EBN bioactive peptides as well as their absorption and changes in the digestive system should also be studied. By taking full advantage of the use of EBN, including its by-products, bioactive peptides derived from EBN can be valuable functional food ingredients for a variety of uses and applications in the food, health, pharmaceutical, and nutraceutical industries.

ACKNOWLEDGEMENTS

This review is supported by the Research Excellence Consortium fund (KKP/2020/UKM-UKM/5/1) (JPT (BKPI) 1000/016/018/25 (21)) provided by the Ministry of Higher Education, Malaysia. The authors would like to acknowledge the Innovation Centre for Confectionery Technology (MANIS) and the Department of Food Sciences, Faculty of Science and Technology, Universiti Kebangsaan Malaysia, which provided all the facilities and resources for this review.

REFERENCES

- Akmal, M.N., Intan-Shameha, A.R., Mansor, R., Ideris, A. & Rahman, A. 2020. High-dose edible bird's nest extract (EBN) upregulates ldl-r via suppression of *HMGCR* gene expression in HepG2 cell lines. *Sains Malaysiana* 49(10): 2433-2442.
- Ali, A.A.M., Hidayati Syamimi, M.N., Chong, P.K., Abdul Salam, B. & Lim, S.J. 2019. Comparison of amino acids profile and antioxidant activities between edible bird nest and chicken egg. *Malaysian Applied Biology* 48(2): 63-66.
- Amin, A.M., Din, K. & Kee Chow, H. 2019. Optimization of enzymatic hydrolysis condition of edible bird's nest using protamex to obtain maximum degree of hydrolysis. *Asian Journal of Agriculture and Biology* 7(1): 1-9.
- Aminoff, D. 1961. Methods for the quantitative estimation of N-acetylneuraminic acid and their application to hydrolysates of sialomucoids. *Biochemical Journal* 81(2): 384-392.
- Azmi, N.A., Lee, T.H., Lee, C.H., Hamdan, N. & Cheng, K.K. 2021. Differentiation unclean and cleaned edible bird's nest using multivariate analysis of amino acid composition data. *Pertanika Journal of Science and Technology* 29(1): 2174-2020.
- Babji, A., Ibrahim, E.S.K., Daud, N., Nadia, N., Akbar, H., Ghassem, M., Najafian, L. & Salma, M. 2018. Assessment on bioactive components of hydrolysed edible bird nest. *International Food Research Journal* 25(5): 1936-1941.
- Badruzaman, S.Z.S., Aminan, A.W., Ramli, A.N.M., Che Man, R. & Wan Azelee, N.I. 2021. Extraction and characterization of keratin from chicken and swiftlet feather. *Materials Science Forum* 1(1): 157-162.
- Barchi Jr., J.J. 2013. Mucin-type glycopeptide structure in solution: Past, present, and future. *Biopolymers* 99(10): 713-723.
- Bhari, R., Kaur, M. & Singh, R.S. 2021. Chicken feather waste hydrolysate as a superior biofertilizer in agroindustry. *Current Microbiology* 78(6): 1-19.
- Ben Hamad Bouhamed, S., Krichen, F. & Kechaou, N. 2020. Feather protein hydrolysates: A study of physicochemical, functional properties and antioxidant activity. *Waste and Biomass Valorization* 11(1): 51-62.
- Callegaro, K., Brandelli, A. & Daroit, D.J. 2019. Beyond plucking: Feathers bioprocessing into valuable protein hydrolysates. *Waste Management* 95: 399-415.
- Callegaro, K., Welter, N. & Daroit, D.J. 2018. Feathers as bioresource: Microbial conversion into bioactive protein hydrolysates. *Process Biochemistry* 75(1): 1-9.
- Careena, S., Sani, D., Tan, S.N., Lim, C.W., Hassan, S., Norhafizah, M., Kirby, B.P., Ideris, A., Stanslas, J., Hamidon, B.B. & Lim, C.T.S. 2018. Effect of edible bird's nest extract on lipopolysaccharide-induced impairment of learning and memory in Wistar rats. *Evidence-Based Complementary and Alternative Medicine* 2018(1): 9318789.
- Chan, G.K.L., Wong, Z.C.F., Lam, K.Y.C., Cheng, L.K.W., Zhang, L.M., Lin, H., Dong, T.T. & Tsim, K.W.K. 2015. Edible bird's nest, an Asian health food supplement, possesses skin lightening activities: Identification of n-acetylneuraminic acid as active ingredient. *Journal of Cosmetics, Dermatological Sciences and Applications* 5(4): 262-274.
- Chaturvedi, V., Agrawal, K. & Verma, P. 2021. Chicken feathers: A treasure cove of useful metabolites and value-added products. *Environmental Sustainability* 4(1): 231-243.
- Cheong, C.W., Lee, Y.S., Ahmad, S.A., Ooi, P.T. & Phang, L.Y. 2018. Chicken feather valorization by thermal alkaline pretreatment followed by enzymatic hydrolysis for protein-rich hydrolysate production. *Waste Management* 79: 658-666.
- Cheong, C.W., Ahmad, S.A., Ooi, P.T. & Phang, L.Y. 2017. Treatments of chicken feather waste. *Pertanika Journal of Scholarly Research Reviews* 3(1): 32-41.
- Chua, K.H., Lee, T.H., Nagandran, K., Md Yahaya, N.H., Lee, C.T., Tjih, E.T.T. & Abdul Aziz, R. 2013. Edible bird's nest extract as a chondro-protective agent for human chondrocytes isolated from osteoarthritic knee: *In vitro* study. *BMC Complementary and Alternative Medicine* 13: 19.
- Colombo, J., Garcia-Rodenas, C., Guesry, P.R. & Rey, J. 2003. Potential effects of supplementation with amino acids, choline or sialic acid on cognitive development in young infants. *Acta Paediatrica* 92(442): 42-46.
- Comb, D.G. & Roseman, S. 1960. The sialic acids I. The structure and enzymatic synthesis of N-acetylneuraminic acid. *Journal of Biological Chemistry* 235(9): 2529-2537.
- Comb, D.G. & Roseman, S. 1958. Composition and enzymatic synthesis of N-acetylneuraminic acid (sialic acid). *Journal of the American Chemical Society* 80(2): 497-499.
- Dai, Y., Cao, J., Wang, Y., Chen, Y. & Jiang, L. 2021. A comprehensive review of edible bird's nest. *Food Research International* 140(1): 109875.
- Daud, N.A., Mohamad Yusop, S., Babji, A.S., Lim, S.J., Sarbini, S.R. & Hui Yan, T. 2021a. Edible bird's nest: Physicochemical properties, production, and application of bioactive extracts and glycopeptides. *Food Reviews International* 37(2): 177-196.
- Daud, N.A., Yusop, S.M., Lim, S.J. & Babji, A.S. 2021b. Evaluation of glycan compound from swiftlet's edible nest (*Aerodramus fuciphagus*) as potential prebiotic material. *Current Advances in Chemistry and Biochemistry* 4(1): 7-15.
- Daliri, E.B., Oh, D.H. & Lee, B.H. 2017. Bioactive peptides. *Foods (Basel, Switzerland)* 6(5): 32.
- Department of Standards Malaysia. 2010. MS 2333:2010 Good manufacturing practice (GMP) for processing raw-unclean and raw-clean edible-bird nest (EBN).

- Department of Veterinary Services Malaysia. 2021. *Lawatan Kerja YB Menteri Pertanian dan Industri Makanan ke Loji Pemprosesan Sarang Burung Walet: 2*. Kuala Lumpur, Malaysia.
- Etty Syarmila, I.K. 2019. *Sarang Burung Walet Suatu Dimensi Baru*. Bangi: Penerbit Universiti Kebangsaan Malaysia.
- Etty Syarmila, I.K., Ayub, M.K. & Babji, A.S. 2014. Effect of enzymatic hydrolysis of pancreatin and alcalase enzyme on some properties of edible bird's nest hydrolysate. *AIP Conference Proceedings* 1614(1): 427-432.
- Gan, J.Y., Chang, L.S., Mat Nasir, N.A., Babji, A.S. & Lim, S.J. 2020. Evaluation of physicochemical properties, amino acid profile and bioactivities of edible Bird's nest hydrolysate as affected by drying methods. *LWT Food Science & Technology* 131(1): 109777.
- Gausset, Q. 2002. A short history of birds' nests management in the Niah Caves (Sarawak). *Borneo Research Bulletin* 33(1): 127-140.
- Ghassem, M., Arihara, K., Mohammadi, S., Sani, N.A. & Babji, A.S. 2017. Identification of two novel antioxidant peptides from edible bird's nest (*Aerodramus fuciphagus*) protein hydrolysates. *Food and Function* 8(5): 2046-2052.
- Guo, C.T., Takahashi, T., Bukawa, W., Takahashi, N., Yagi, H., Kato, K., Kazuya, I.P., Miyamoto, D., Suzuki, T. & Suzuki, Y. 2006. Edible bird's nest extract inhibits influenza virus infection. *Antiviral Research* 70(3): 140-146.
- Guo, L., Wu, Y., Liu, M., Wang, B., Ge, Y. & Chen, Y. 2017. Determination of edible bird's nests by FTIR and SDS-PAGE coupled with multivariate analysis. *Food Control* 80(1): 259-266.
- Halimi, N.M., Kasim, Z.M. & Babji, A.S. 2014. Nutritional composition and solubility of edible bird nest (*Aerodramus fuchiphagus*). *AIP Conference Proceedings* 1614: 476-481.
- Hamzah, Z., Jeyaraman, S., Hashim, O. & Hussin, K. 2016. Application of Fourier transform infrared spectroscopy on edible bird nest authenticity. *Contemporary Issues and Development in the Global Halal Industry* 1(1): 557-566.
- Hobbs, J.J. 2004. Problems in the harvest of edible birds' nests in Sarawak and Sabah, Malaysian Borneo. *Biodiversity and Conservation* 13(12): 2209-2226.
- Hou, Z., Imam, M.U., Ismail, M., Azmi, N.H., Ismail, N., Ideris, A. & Mahmud, R. 2015. Lactoferrin and ovotransferrin contribute toward antioxidative effects of Edible Bird's Nest against hydrogen peroxide-induced oxidative stress in human SH-SY5Y cells. *Bioscience, Biotechnology, and Biochemistry* 79(10): 1570-1578.
- Hou, Z.P., Tang, S.Y., Ji, H.R., He, P.Y., Li, Y.H., Dong, X.L., Du, M.N., Maznah, I. & He, W.J. 2021. Edible bird's nest attenuates menopause-related bone degeneration in rats via increasing bone estrogen-receptor expression. *Chinese Journal of Integrative Medicine* 27(4): 280-285.
- Houdret, N., Lhermitte, M., Degand, P. & Roussel, P. 1975. Purification and chemical study of a *Collocalia* glycoprotein. *Biochimie* 57(5): 603-608.
- Howe, C., Lee, L.T. & Rose, H.M. 1961. *Collocalia* mucoid: A substrate for myxovirus neuraminidase. *Archives of Biochemistry and Biophysics* 95(3): 512-520.
- Howe, C., Lee, L.T. & Rose, H.M. 1960. Influenza virus sialidase. *Nature* 188(4746): 251-252.
- Huang, X., Li, Z., Zou, X., Shi, J., Elrasheid Tahir, H., Xu, Y., Zhai, X. & Hu, X. 2019. A low-cost smart system to analyze different types of edible bird's nest adulteration based on colorimetric sensor array. *Journal of Food and Drug Analysis* 27(4): 876-886.
- Huda, M.Z., Abu Bakar, M.Z., Goh, Y., Shuhaimi, H., Awang Junaidi, A.H. & Zairi, M.S. 2008. Proximate, elemental and free fatty acids of pre-processed edible bird's nest (*Aerodramus fuciphagus*): A comparison between regions and type of nest. *Journal of Food Technology* 6(1): 39-44.
- Hun, L.T., Wani, W.A., Tjih, E.T.T., Adnan, N.A., Le Ling, Y. & Aziz, R.A. 2015. Investigations into the physicochemical, biochemical and antibacterial properties of edible bird's nest. *Journal of Chemical and Pharmaceutical Research* 7(7): 228-247.
- Hwang, E., Park, S.W. & Yang, J.E. 2020. Anti-aging, anti-inflammatory, and wound-healing activities of edible bird's nest in human skin keratinocytes and fibroblasts. *Pharmacognosy Magazine* 16(69): 336-342.
- Ismail, M.Y. 1999. Social control and bird's nest harvesting among the Idahan: A preliminary observation. *Japanese Journal of Southeast Asian Studies* 37(1): 3-17.
- Jang, A. & Lee, M. 2005. Purification and identification of angiotensin converting enzyme inhibitory peptides from beef hydrolysates. *Meat Science* 69(4): 653-661.
- Jin, J., Okagu, O.D., Yagoub, A.E.A. & Udenigwe, C.C. 2021. Effects of sonication on the *in vitro* digestibility and structural properties of buckwheat protein isolates. *Ultrasonics Sonochemistry* 70(1): 105348.
- Kathan, R.H. & Weeks, D.I. 1969. Structure studies of *Collocalia* mucoid: I. Carbohydrate and amino acid composition. *Archives of Biochemistry and Biophysics* 134(2): 572-576.
- Khalid, S.K.A., Abd Rashed, A., Aziz, S.A. & Ahmad, H. 2019. Effects of sialic acid from edible bird nest on cell viability associated with brain cognitive performance in mice. *World Journal of Traditional Chinese Medicine* 5(4): 214-232.
- Kong, H., Wong, K.H. & Lo, S.C. 2016. Identification of peptides released from hot water insoluble fraction of edible bird's nest under simulated gastro-intestinal conditions. *Food Research International* 85(1): 19-25.
- Lee, S.H., Qian, Z.J. & Kim, S.K. 2010. A novel angiotensin I converting enzyme inhibitory peptide from tuna frame protein hydrolysate and its antihypertensive effect in spontaneously hypertensive rats. *Food Chemistry* 118(1): 96-102.

- Lee, T.H., Wani, W.A., Lee, C.H., Cheng, K.K., Shreaz, S., Wong, S., Hamdan, N. & Azmi, N.A. 2021. Edible bird's nest: The functional values of the prized animal-based bioproduct from Southeast Asia - A review. *Frontiers in Pharmacology* 12(1): 871-887.
- Lee, Y.S., Phang, L.Y., Ahmad, S.A. & Ooi, P.T. 2016. Microwave-alkali treatment of chicken feathers for protein hydrolysate production. *Waste and Biomass Valorization* 7(5): 1147-1157.
- Lim, C.K. & Cranbrook, E.O. 2002. *Swiftlets of Borneo: Builders of Edible Nests*. Natural History Publications (Borneo) Sdn. Bhd.
- Lim, G.W. & Rahman, M.A. 2005. Patterns of genetic variation among white-nest swiftlets (*Aerodramus fuciphagus*) in Malaysia based on partial cytochrome b region. In *Wallace in Sarawak - 150 Years Later*, edited by Tuen, A.A. & Das, I. Kota Samarahan: Institute of Biodiversity and Environmental Conservation, Universiti Malaysia Sarawak.
- Lim, S.J., Chang, L.S., Fazry, S., Mustapha, W.A.W. & Babji, A.S. 2021. Functional food & ingredients from seaweed, edible bird's nest and tropical fruits: A translational research. *LWT-Food Science and Technology* 151: 112164.
- Ling, J.W.A., Chang, L.S., Babji, A.S. & Lim, S.J. 2020. Recovery of value-added glycopeptides from edible bird's nest (EBN) co-products: Enzymatic hydrolysis, physicochemical characteristics and bioactivity. *Journal of the Science of Food and Agriculture* 100(13): 4714-4722.
- Maciel, J.L., Werlang, P.O., Daroit, D.J. & Brandelli, A. 2017. Characterization of protein-rich hydrolysates produced through microbial conversion of waste feathers. *Waste and Biomass Valorization* 8(4): 1177-1186.
- Manikkam, V., Vasiljevic, T., Donkor, O.N. & Mathai, M.L. 2016. A review of potential marine-derived hypotensive and anti-obesity peptides. *Critical Reviews in Food Science and Nutrition* 56(1): 92-112.
- Mansor, M. & Rahman, M.K.A. 2013. *Merekayasa Kearifan Tempatan: Alam dan Manusia*. Penang: Penerbit Universiti Sains Malaysia.
- Marcone, M.F. 2005. Characterization of the edible bird's nest the "Caviar of the East." *Food Research International* 38(10): 1125-1134.
- Marni, S., Marzura, M.R., Norzela, A.M., Khairunnisak, M., Bing, C.H. & Eddy, A.A. 2014. Preliminary study on free sialic acid content of edible bird nest from Johor and Kelantan. *Malaysian Journal of Veterinary Research* 5(1): 9-14.
- Matsukawa, N., Matsumoto, M., Bukawa, W., Chiji, H., Nakayama, K., Hara, H. & Tsukahara, T. 2011. Improvement of bone strength and dermal thickness due to dietary edible bird's nest extract in ovariectomized rats. *Bioscience, Biotechnology, and Biochemistry* 75(3): 590-592.
- Ministry of Agriculture and Food Industries Malaysia. 2021. *National Agrofood Policy 2.0 2021-2030*. Malaysia: Putrajaya.
- Ministry of Agriculture and Food Industries Malaysia. 2011. *National Agrofood Policy 2011-2020*. Malaysia: Putrajaya.
- Mintah, B.K., He, R., Dabbour, M., Golly, M.K., Agyekum, A.A. & Ma, H. 2019. Effect of sonication pretreatment parameters and their optimization on the antioxidant activity of *Hermitia illucens* larvae meal protein hydrolysates. *Journal of Food Processing and Preservation* 43(9): e14093.
- Mohd. Rashid, R. 2016. *Kajian Penandaarasan Teknologi Pertanian Terpilih: Industri Pemprosesan Sarang Burung Walit*. Laporan Kajian Sosioekonomi 2015. MARDI. Pusat Penyelidikan Ekonomi dan Sains Sosial. pp. 123-129.
- Mohd. Rashid, R., Ahmad Zairy, Z.A. & Mohd Syauqi, N. 2013. *Prospek dan cabaran industri sarang burung walit negara*. Buletin Teknologi Pusat Penyelidikan Ekonomi dan Pengurusan Teknologi. Institut Penyelidikan dan Kemajuan Pertanian Malaysia (MARDI). pp. 57-69.
- Mohd Syauqi, N., Ahmad Zairy, Z.A., Mohd. Rashid, R., Nor Amna A'liah, N.M. & Che Nurul Akmal, C.M. 2020. *Daya saing dan amalan teknologi industri sarang burung walit di Malaysia dan Indonesia: Satu perbandingan umum*. Buletin Teknologi Pusat Penyelidikan Ekonomi dan Pengurusan Teknologi. Institut Penyelidikan dan Kemajuan Pertanian Malaysia (MARDI). pp. 169-176.
- Möller, N.P., Scholz-Ahrens, K.E., Roos, N. & Schrezenmeir, J. 2008. Bioactive peptides and proteins from foods: Indication for health effects. *European Journal of Nutrition* 47(4): 171-182.
- Muhammad, N.N., Babji, A.S. & Ayub, M.K. 2015. Antioxidative activities of hydrolysates from edible birds nest using enzymatic hydrolysis. *AIP Conference Proceedings* 1678(1): 50038.
- Ng, S.R., Noor, H.S.M., Ramachandran, R., Tan, H.Y., Ch, S.E., Chang, L.S., Babji, A.S. & Lim, S.J. 2020. Recovery of glycopeptides by enzymatic hydrolysis of edible bird's nest: The physicochemical characteristics and protein profile. *Journal of Food Measurement and Characterization* 14(5): 2635-2645.
- Noor, H.S.M., Babji, A.S. & Lim, S.J. 2018. Nutritional composition of different grades of edible bird's nest and its enzymatic hydrolysis. *AIP Conference Proceedings* 1940(1): 20088.
- Norhayati, M., Azman, O. & Wan Mohamud, W.N. 2010. Preliminary study of the nutritional content of Malaysian edible bird's nest. *Malaysian Journal of Nutrition* 16(3): 389-396.
- Nurfatin, M.H., Ety Syarmila, I.K., Nur'Aliah, D., Zalifah, M.K., Babji, A.S. & Ayob, M.K. 2016. Effect of enzymatic hydrolysis on angiotensin converting enzyme (ACE) inhibitory activity in swiftlet saliva. *International Food Research Journal* 23(1): 141-146.
- Nurul Nadia, M., Babji, A.S., Ayub, M.K. & Nur'Aliah, D. 2017. Effect of enzymatic hydrolysis on antioxidant capacity of cave edible bird's nests hydrolysate. *International Journal of Chemical Technology Research* 10(2): 1100-1107.

- Nurul Nadiah, M.N., Mohamad Ibrahim, R., Abu Bakar, M.Z., Mahmud, R. & Ab Razak, N.A. 2021. Characterization and extraction influence protein profiling of edible bird's nest. *Foods* 10(10): 2248-2267.
- Paul, T., Das, A., Mandal, A., Halder, S.K., DasMohapatra, P.K., Pati, B.R. & Mondal, K.C. 2014. Valorization of chicken feather waste for concomitant production of keratinase, oligopeptides and essential amino acids under submerged fermentation by *Paenibacillus woosongensis* TKB2. *Waste and Biomass Valorization* 5(4): 575-584.
- Pozsgay, V., Jennings, H. & Kasper, D.L. 1987. 4, 8-Anhydro-N-acetylneuraminic acid: Isolation from edible bird's nest and structure determination. *European Journal of Biochemistry* 162(2): 445-450.
- Quek, M.C., Chin, N.L., Yusof, Y.A., Law, C.L. & Tan, S.W. 2018. Pattern recognition analysis on nutritional profile and chemical composition of edible bird's nest for its origin and authentication. *International Journal of Food Properties* 21(1): 1680-1696.
- Ramachandran, R., Babji, A.S. & Sani, N.A. 2018. Antihypertensive potential of bioactive hydrolysate from edible bird's nest. *AIP Conference Proceedings* 1940(1): 20099-1-20099-7.
- Saengkrajang, W., Matan, N. & Matan, N. 2013. Nutritional composition of the farmed edible bird's nest (*Collocalia fuciphaga*) in Thailand. *Journal of Food Composition and Analysis* 31(1): 41-45.
- Sánchez, A. & Vázquez, A. 2017. Bioactive peptides: A review. *Food Quality and Safety* 1(1): 29-46.
- Sarmadi, B.H. & Ismail, A. 2010. Antioxidative peptides from food proteins: A review. *Peptides* 31(10): 1949-1956.
- Shi, J., Hu, X., Zou, X., Zhao, J., Zhang, W., Holmes, M., Huang, X., Zhu, Y., Li, Z., Shen, T. & Zhang, X. 2017. A rapid and nondestructive method to determine the distribution map of protein, carbohydrate and sialic acid on Edible bird's nest by hyper-spectral imaging and chemometrics. *Food Chemistry* 229(1): 235-241.
- Sitanggang, A.B., Putri, J.E., Palupi, N.S., Hatzakis, E., Syamsir, E. & Budijanto, S. 2021. Enzymatic preparation of bioactive peptides exhibiting ace inhibitory activity from soybean and velvet bean: A systematic review. *Molecules* 26(13): 3822-3836.
- Spiro, R.G. 2002. Protein glycosylation: Nature, distribution, enzymatic formation, and disease implications of glycopeptide bonds. *Glycobiology* 12(4): 43R-56R.
- Tagliacozzo, E. & Chang, W.C. 2011. *Chinese Circulations: Capital, Commodities, and Networks in Southeast Asia*. Duke University Press.
- Tan, S., Sani, D., Lim, C., Ideris, A., Stanslas, J. & Thiam Seong Lim, C. 2020. Proximate analysis and safety profile of farmed edible bird's nest in Malaysia and its effect on cancer cells. *Evidence-Based Complementary and Alternative Medicine* 2020: ID8068797.
- Thorburn, C. 2014. The edible birds' nest boom in Indonesia and South-east Asia: A nested political ecology. *Food, Culture & Society* 17(4): 535-553.
- Tung, C.H., Pan, J.Q., Chang, H.M. & Chou, S.S. 2008. Authentic determination of bird's nests by saccharides profile. *Journal of Food Drug Analysis* 16(4): 86-91.
- Utomo, B., Rosyidi, D., Radiati, L.E., Puspaningsih, N.N.T. & Proborini, W.D. 2014. Protein characterization of extracted water from three kinds of edible bird nest using SDS-PAGE CBB staining and SDS-PAGE glycoprotein staining and LC-MS/MS analyses. *IOSR Journal of Agriculture and Veterinary Science* 7(9): 33-38.
- Varki, A. 2008. Sialic acids in human health and disease. *Trends in Molecular Medicine* 14(8): 351-360.
- Wang, B. & Brand-Miller, J. 2003. The role and potential of sialic acid in human nutrition. *European Journal of Clinical Nutrition* 57(11): 1351-1369.
- Wang, C.Y., Cheng, L.J., Shen, B., Yuan, Z.L., Feng, Y.Q. & Lu, S. 2019. Antihypertensive and antioxidant properties of sialic acid, the major component of edible bird's nests. *Current Topics in Nutraceutical Research* 17(4): 376-380.
- Wang, C.C. 1921a. The isolation and the nature of the amino sugar of Chinese edible birds' nests. *Journal of Biological Chemistry* 49(2): 441-452.
- Wang, C.C. 1921b. The composition of Chinese edible birds' nests and the nature of their proteins. *Journal of Biological Chemistry* 49(2): 429-439.
- Wang, H., Zhang, S., Sun, Y. & Dai, Y. 2013. ACE-inhibitory peptide isolated from fermented soybean meal as functional food. *International Journal of Food Engineering* 9(1): 1-8.
- Wang, L.L., Chen, N., Zhang, W.W., Wu, G.H. & Lai, X.P. 2013. Study on a collagenase protocol to extract DNA from remnant feathers in edible bird's nest. *Journal of Chinese Medicinal Materials* 36(8): 1224-1226.
- Wieruszkeski, J.M., Michalski, J.C., Montreuil, J., Strecker, G., Peter-Katalinic, J., Egge, H., Van Halbeek, H., Mutsaers, J.H. & Vliegthart, J.F. 1987. Structure of the monosialyl oligosaccharides derived from salivary gland mucin glycoproteins of the Chinese swiftlet (genus *Collocalia*). Characterization of novel types of extended core structure, Gal beta (1---3)[GlcNAc beta (1---6)] GalNAc alpha (1---3) GalNAc(-ol), and of chain termination, [Gal alpha(1---4)]0-1[Gal beta(1---4)]2GlcNAc beta(1---). *Journal of Biological Chemistry* 262(14): 6650-6657.
- Wong, C.F., Chan, G.K.L., Zhang, M.L., Yao, P., Lin, H.Q., Dong, T.T.X., Li, G., Lai, X.P. & Tsim, K.W.K. 2017. Characterization of edible bird's nest by peptide fingerprinting with principal component analysis. *Food Quality and Safety* 1(1): 83-92.
- Wong, Z., Chan, G., Wu, Q., Poon, K., Chen, Y. & Dong, T. 2018. Completed digestion of edible bird's nest releases free N-acetyl- neuraminic acid and small peptide: An efficiency method to improve functional properties. *Food and Function* 9(10): 5139-5149.
- Xu, H., Zheng, L., Xie, Y., Zeng, H., Fan, Q., Zheng, B. & Zhang, Y. 2019. Identification and determination of glycoprotein of edible bird's nest by nanocomposites based lateral flow immunoassay. *Food Control* 102(1): 214-220.

- Yan, Y.H., Lim, S.J., Babji, A.S., Rawi, M.H. & Sarbini, S.R. 2021. Enzymatic hydrolysis: Sialylated mucin (SiaMuc) glycoprotein of edible swiftlet's nest (ESN) and its molecular weight distribution as bioactive ESN SiaMuc-glycopeptide hydrolysate. *International Journal of Biological Macromolecules* 175(1): 422-431.
- Yan, T.H., Babji, A.S., Lim, S.J. & Sarbini, S.R. 2021. A systematic review of edible Swiftlet's nest (ESN): Nutritional bioactive compounds, health benefits as functional food, and recent development as bioactive ESN glycopeptide hydrolysate. *Trends in Food Science and Technology* 115(1): 117-132.
- Yao, Y., Yuan, X., Wang, M., Han, L. & Liu, X. 2021. Efficient pretreatment of waste protein recovery from bovine bones and its underlying mechanisms. *Waste and Biomass Valorization* 12(1): 5413-5423.
- Yeo, B.H., Tang, T.K., Wong, S.F., Tan, C.P., Wang, Y., Cheong, L.Z. & Lai, O.M. 2021. Potential residual contaminants in edible bird's nest. *Frontiers in Pharmacology* 12(1): 312.
- Yew, M.Y., Koh, R.Y., Chye, S.M., Zainal Abidin, S.A., Othman, I. & Ng, K.Y. 2019. Neurotrophic properties and the *de novo* peptide sequencing of edible bird's nest extracts. *Food Bioscience* 32: 100466.
- You, Y., Cao, Y., Guo, S., Xu, J., Li, Z., Wang, J. & Xue, C. 2015. Purification and identification of α 2–3 linked sialoglycoprotein and α 2–6 linked sialoglycoprotein in edible bird's nest. *European Food Research and Technology* 240(2): 389-397.
- Zainab, H., Jeyaraman, S., Othman, H. & Kamarudin, H. 2015. Waste to wealth for the edible bird nest industry. *Applied Mechanics and Materials* 754-755: 990-997.
- Zhao, R., Li, G., Kong, X.J., Huang, X.Y., Li, W., Zeng, Y.Y. & Lai, X.P. 2016. The improvement effects of edible bird's nest on proliferation and activation of B lymphocyte and its antagonistic effects on immunosuppression induced by cyclophosphamide. *Drug Design, Development and Therapy* 10: 371-381.
- Zou, T.B., He, T.P., Li, H.B., Tang, H.W. & Xia, E.Q. 2016. The structure-activity relationship of the antioxidant peptides from natural proteins. *Molecules* 21(1): 72-85.
- Zulkifli, A.S., Babji, A.S., Lim, S.J., Teh, A.H., Daud, N.M. & Rahman, H.A. 2019. Effect of different hydrolysis time and enzymes on chemical properties, antioxidant and antihyperglycemic activities of edible bird nest hydrolysate. *Malaysian Applied Biology* 48(2): 149-156.

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