



UNIVERSITI PUTRA MALAYSIA

**CHARACTERIZATION AND PRIMARY RECOVERY OF FUNCTIONAL
BIOACTIVE COMPOUNDS FROM EDIBLE BIRD'S NEST**

LIM HONG KWONG

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By

LIM HONG KWONG

**Thesis Submitted to the School of Graduate Studies, Universiti Putra
Malaysia, in Fulfilment of the Requirements for the Degree of Doctor of
Philosophy**

June 2014

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Abstract of the thesis presented to the Senate of Universiti Putra Malaysia in
fulfilment of the requirement for the degree of Doctor of Philosophy

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June 2014

Chairman : Professor Tan Chin Ping, PhD.

Faculty : Food Science and Technology

The edible bird's nest (EBN) is a saliva-cemented nest built by swiftlets (*Collocalia spp.* /*Aerodramus spp.*) during the breeding season, and it is found predominantly in Southeast Asia. However, little is known or is published about EBN and no scientific and technological findings on recovering bioactive compounds from EBN. Therefore, this study was conducted to characterize the physico-chemical, rheological behavior properties and recover the functional bio-compounds derived from EBN. The application of an aqueous two-phase system (ATPS) strategy to the potential recovery of bio-compounds (sialoglycoproteins) from the EBN was evaluated. This study revealed that the amino acid composition of EBN-extract was significantly higher ($P < 0.05$) than that of intact EBN, with the exception of the methionine content. Overall, the most abundant amino acids in the chemical structure of EBN-extract were aspartic acid (9.80 %), serine (9.50 %), and proline (8.81 %). The hydrophilic glycoprotein in EBN-extract was a heavily glycosylated protein, and this result was further supported by the presence of two glycoprotein bands at 110 and 130 kDa, as observed by SDS-PAGE analysis. The non-Newtonian flow behavior of *Collocalia* mucoid (EBN-extract) dispersions indicated that all concentrations (5 – 100 mg/mL) of the mucoid dispersions exhibited shear-thinning behaviors, which could best be described using the Herschel-Bulkley model. As the concentration of *Collocalia* mucoid dispersions increased, the appearance of a plateau shear modulus indicated the dispersion was exhibiting more gel-like behaviors. The *Collocalia* mucoid dispersions were thought to involve the physical entanglement of network

formations, which were able to form weak gels at high concentrations. The effects of hydrophilic solvent in the ATPS and the system parameters on partition behavior were evaluated. The partitioning of the EBN sialoglycoprotein was pH-dependant. Slightly more than 97 % of the total EBN sialoglycoproteins were recovered in the top phase of ATPS under selected conditions. According to this study, the ATPS technique provides a simple, efficient, and economic recovery process for sialoglycoproteins derived from EBN and EBN by-products for the food, nutraceutical, pharmaceutical, cosmetic, and other related industries. This ATPS separation technique has great potential to be carried over to an industrial scale.



Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk Ijazah Doktor Falsafah

**PENCIRIAN DAN PEMULIHAN AWAL SEBATIAN BIO-AKTIF
FUNGSIAN DARIPADA SARANG BURUNG WALIT**

Oleh

LIM HONG KWONG

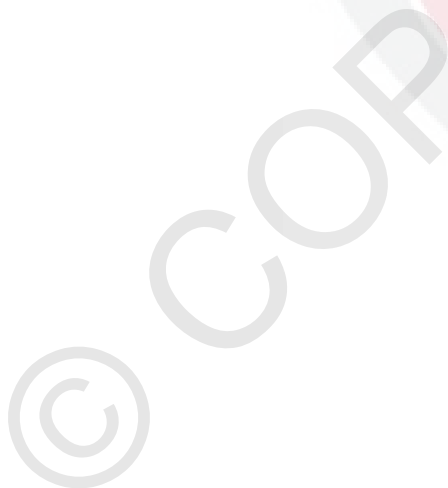
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Pengerusi : Profesor Tan Chin Ping, PhD.

Fakulti : Sains dan Teknologi Makanan

Sarang burung walit (EBN) merupakan sarang burung yang dihasilkan daripada air liur burung walit (*Collocalia spp.* /*Aerodramus spp.*) semasa musim bertelur dan biasanya ditemui di Asia Tenggara. Walau bagaimanapun, hanya sedikit maklumat mengenai EBN telah diketahui atau diterbitkan dan tiada penemuan saintifik dan teknologi mengenai pemulihan sebatian bio-aktif daripada EBN. Justeru itu, kajian ini telah dijalankan untuk mencirikan sifat fiziko-kimia, kelakuan reologi dan pemulihan bio-sebatian fungsian yang diperolehi daripada EBN. Potensi penggunaan sistem akueus dua fasa (ATPS) strategi untuk pemulihan bio-sebatian (sialoglikoprotein) daripada EBN telah dinilai. Keputusan kajian ini menunjukkan bahawa komposisi asid amino daripada ekstrak EBN adalah lebih tinggi ($P < 0.05$) berbanding dengan EBN mentah, kecuali kandungan asid amino metionina. Secara keseluruhannya, kandungan asid amino dalam struktur kimia ekstrak EBN yang utama terdiri daripada asid aspartik (9.80 %), serin (9.50 %), dan prolin (8.81 %). Glikoprotein hidrofilik yang terdapat dalam ekstrak EBN adalah protein terglykosilat, dan keputusan ini turut disokong dengan kehadiran dua jalur glikoprotein pada 110 dan 130 kDa, seperti yang diperhatikan dalam analisis SDS-PAGE. Larutan lendir *Collocalia* (ekstrak EBN) yang berkelakuan aliran bukan “Newtonian” dan menunjukkan bahawa semua kepekatan (5 – 100 mg/mL) daripada larutan lendir mempamerkan tingkah laku yang ricih penipisan yang dipadankan dengan model “Herschel-Bulkley”. Dengan peningkatan kepekatan larutan lendir *Collocalia*, kemunculan dataran modulus ricih telah menunjukkan larutan tersebut lebih bersifat seperti gel. Keputusan ini

mencadangkan bahawa lendir *Collocalia* yang terlibat dalam pembentukan rangkaian secara fizikal yang akan menyebabkan larutannya bersifat gel yang lemah pada kepekatan yang tinggi. Kesan-kesan pelarut hidrofilik dalam sistem akueus dua fasa (ATP) dan beberapa parameter sistem terhadap tingkah laku pemisahan telah dinilai. Pemisahan sialoglikoprotein EBN adalah bergantung kepada pH. Lebih kurang 97 % daripada jumlah sialoglikoprotein EBN telah didapati dalam fasa atas ATPS di bawah keadaan yang terpilih. Kajian ini menunjukkan bahawa teknik ATPS merupakan satu proses pemulihan sialoglikoprotein daripada EBN dan hasil sampingan EBN yang mudah, berkesan, dan ekonomik untuk penggunaan dalam industri makanan, nutraseutikal, farmaseutikal, kosmetik, dan industri lain yang berkenaan. Teknik pemisahan ATPS ini amat berpotensi untuk dibawa ke skala industri.



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I certify that a Thesis Examination Committee has met on 18 Jun 2014 to conduct the final examination of Lim Hong Kwong on his thesis entitled “Characterization and Primary Recovery of Functional Bioactive Compounds from Edible Bird’s Nest” in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The Committee recommends that the student be awarded the Doctor of Philosophy.

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DECLARATION

Declaration by graduate student

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