



**UNIVERSITI PUTRA MALAYSIA**

***DETECTION OF SALMONELLA AND E.COLI IN EDIBLE BIRD'S NEST  
RANCHED IN HOUSING SYSTEM***

**NORFARIDAH BINTI MOHAMAD RAZAK**

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**DETECTION OF *SALMONELLA* AND *E.COLI* IN EDIBLE BIRD'S NEST  
RANCHED IN HOUSING SYSTEM**

**NORFARIDAH BINTI MOHAMAD RAZAK**

A project paper submitted to the  
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DEGREE OF DOCTOR OF VETERINARY MEDICINE

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## CERTIFICATION

It is hereby certified that we have read this project paper entitled “Detection of *Salmonella* and *E.coli* in Edible Bird’s Nest Ranched in Housing System”, by Norfaridah binti Mohamad Razak and in our opinion it is satisfactory in terms of scope, quality, and presentation as partial fulfillment of the requirement for the course VPD 4999-Project

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**PROF. DATIN PADUKA DR. AINI BT IDERIS**  
**DVM (UPM), MVSc (Liverpool), PhD (UPM)**  
Faculty of Veterinary Medicine  
Universiti Putra Malaysia  
(Supervisor)

---

**PROF. DR SALEHA BT ABDUL AZIZ**  
**DVM (UGM), Grad.Dip.Vet.Preventive Medicine (Guelph), PhD (UPM)**  
Faculty of Veterinary Medicine  
Universiti Putra Malaysia  
(Co-supervisor)

**DEDICATIONS**

This project paper is dedicated to the Almighty Allah, who had made all things possible,

*To my beloved family,*

*Mother & father*

*Brothers*

*Sisters-in-law*

*Nieces and nephews*

*My late nephew*

And to my friends and all my teachers and lecturers who have committed toward the noble cause of education.

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**LIST OF ABBREVIATION**

%	percent
BGA	Brilliant Green Agar
BPW	Buffer Peptone Water
CFU	Colony forming unit
EBN	Edible Bird's Nest
EMBA	Eosin Methylene Blue Agar
g	Gram
LIA	Lysine Iron Agar
ml	mililiter
°C	Degree celsius
RV	Rappaport-Vassiliadis Enrichment Broth
SIM	Sulphide Indole Motility
SIRIM	Standard and Industrial Research Institute of Malaysia
SPC	Standard Plate Count
TSI	Triple Sugar Iron agar
XLD	Xylose Lysine Deoxycholate

## **ABSTRACT**

An abstract of the project paper presented to the Faculty of Veterinary Medicine in partial fulfillment of the course VPD 4999 – Project.

### **DETECTION OF *SALMONELLA* AND *E.COLI* IN EDIBLE BIRD’S NEST RANCHED IN HOUSING SYSTEM**

by

**Norfaridah Mohamad Razak**

**2016**

**Supervisor : Prof. Datin Paduka Dr. Aini Ideris**

**Co-supervisor : Prof. Dr. Saleha Abdul Aziz**

The swiftlet industry in Malaysia is growing very fast due to the high demand for Edible Bird’s Nest (EBN) . The presence of bacteria may produce low quality of EBN that can lead to economic loss and may cause food-borne diseases. The aim of this study was to detect the presence of *Salmonella* and *E.coli* in EBN and guano of edible-nest swiftlets ranched in housing system and to enumerate the total number of bacteria and coliform in EBN. In this study, a total of 64 guano and nest swab samples were collected from three bird houses in Terengganu. The samples were pre-enriched and enriched before culturing on Brilliant Green Agar (BGA) and

Xylose Lysine Deoxycholate (XLD) for isolation of *Salmonella* and propagated in nutrient broth before culture on Eosin Methylene Blue Agar (EMBA) for isolation of *E.coli*. The isolation of the bacteria was carried out in Veterinary Public Health Laboratory, Faculty of Veterinary Medicine, UPM. Standard Plate Count (SPC) and Coliform count using 3M<sup>®</sup> Petrifilm were done. No *Salmonella* was isolated in EBN and the prevalence of *Salmonella* in guano was 12.5% as compared to *E. coli* in EBN at 3.13% and 68.75% in guano sample. The average SPC of EBNs was  $3.2 \times 10^5$  CFU per gram and CPC was  $\leq 100$  CFU per gram. *E.coli* in EBNs swab were lower than in guano samples. Hence, the absence of *Salmonella* and low number of *E.coli* in EBN is a good news for the growing swiftlet industry.

Keywords : *Edible-nest Swiftlet, Edible Bird's Nest, Housing system, Salmonella, E.coli*

**ABSTRAK**

Abstrak daripada kertas kerja projek yang dikemukakan kepada Fakulti Perubatan Veterinar untuk memenuhi sebahagian keperluan kursus VPD 4999 – Projek

**PENGESANAN *SALMONELLA* DAN *E.COLI* PADA SARANG BURUNG  
YANG DIBIAKKAN DI DALAM SISTEM RUMAH BURUNG**

oleh

**Norfaridah Mohamad Razak**

**2016**

**Penyelia : Prof. Datin Paduka Dr. Aini Ideris**

**Penyelia bersama : Prof. Dr. Saleha Abdul Aziz**

Industri sarang burung walit di Malaysia semakin pesat berkembang seiringan dengan permintaan sarang burung yang tinggi. Kewujudan bakteria mengurangkan kualiti sarang burung yang menyebabkan kerugian dan menimbulkan penyakit berpunca pemakanan. Tujuan utama penyelidikan ini adalah untuk mengenalpasti kehadiran *Salmonella* dan *E.coli* di dalam sarang burung walit dan najis burung walit yang dibiak dalam sistem rumah dan untuk mengira jumlah bakteria dan koliform di dalam sarang burung. Dalam kajian ini, sejumlah 64 sampel najis dan

swab sarang burung telah diambil daripada tiga rumah burung di Terengganu, Malaysia. Sampel dibiakkan di dalam cecair nutrisi sebelum dikultur di atas Brilliant Green Agar (BGA) and Xylose Lysine Deoxycholate (XLD) untuk isolasi *Salmonella* dan Eosin Methylene Blue Agar untuk isolasi *E.coli*. Prosedur isolasi telah dijalankan di Makmal Veterinar Kesihatan Awam, Fakulti Perubatan Veterinar, UPM. Kaedah Standard Plate Count (SPC) dan bilangan koliform dikira menggunakan 3M<sup>®</sup> Petrifilm. Tiada *Salmonella* dalam sarang burung dan 12.5 % *Salmonella* positif dalam sampel najis berbanding hanya 3.13% untuk sarang burung positif *E.coli* dan 68.75% dari sampel najis. Purata SPC dari sarang burung walit adalah  $3.2 \times 10^5$  CFU/g manakala purata CPC sebanyak 100 CFU/g. Isolasi *E.coli* dari swab sarang burung lebih rendah berbanding sampel najis. Ketiadaan *Salmonella* dan jumlah *E.coli* yang sedikit di dalam sarang burung merupakan satu berita yang baik untuk perkembangan industri sarang burung walit.

Kata kunci : *burung walit, sarang burung, sistem rumah, Salmonella, E.coli*

## 1.0 INTRODUCTION

Edible Bird's Nest (EBN) is a natural saliva nest (Saengkrajang and Matan, 2011), produced by white-nest swiftlet (*Aerodramus fuciphagus*) and black-nest swiftlet (*Aerodramus maximus*), which are highly traded worldwide (Babji *et al.*, 2015). In Malaysia, the swiftlet industry is growing very fast due to the high demand and high value of EBN at the international market. The major demand is from the Chinese communities around the world, mainly China, Taiwan, Singapore, North America, there are new emerging markets such as Middle East, Japan and Korea (Babji *et al.*, 2015). EBN has been consumed as an expensive delicacy for various health benefits. Eventhough many studies has been done to evaluate the nutritional properties in EBN. So far, the benefits, nutritional and non-nutritional contents of EBNS remain undetermine clearly (Kew *et al.*, 2014). However, the demand for EBN remain high because it is not just a pleasant food to be consumed, but also it has been traditionally used to provide health benefits, such as aiding digestion, raising libido, improving the voice, alleviating asthma and improving concentration (Babji *et al.*,2015).

In 2011, China has listed EBNS from Malaysia as banned products due to the high level of nitrites (Kew *et al.*, 2014). To prevent such incidence from reoccur, food safety aspects is very important to improve the sustainability of the swiftlet industry in Malaysia. However, as in other food, contaminants that are present in the EBN as well as the growth of microbes such as bacteria,virus,yeast and fungi (Oktorina *et al.*, 2005) may produced low quality EBN that lead to economic loss,

but also may cause food-borne diseases. There have been reports that EBN may cause side effects such as allergic symptoms and food-induced anaphylaxis among children (Goh *et al.*, 1999 ; Kemp *et al.*,2010; Kew *et al.*,2014) and adults (Thong *et al.*, 2005,2007 ; Kew *et al.*,2014).

As the price of the EBN is determined by the quality of EBN, a standard guideline has been set by the Standard and Industrial Research Institute of Malaysia (SIRIM). It is mainly to ensure the acceptability of EBNs at the international market. It is stated in the guideline that EBN must not contain *Salmonella* spp. and the microbial content by Total Plate Count should be  $\leq 2.5 \times 10^6$  CFU/g, whereas the Coliforms Count should be  $\leq 100$  CFU/g (Kew *et al.*, 2014) (Refer to Table 1).

Table 1. Microbiological requirements of raw-unclean EBN after pre-cleaning

Category	Parameters	Tolerance level	Method of tests
Microbiological analysis	Total Plate Count	$\leq 2.5 \times 10^5$ cfu/g	Bacteriological
	Coliform Count	$\leq 1.0 \times 10^2$ cfu/g	Analytical manual (BAM) Method or equivalent method
	<i>Salmonella enteritidis</i>	nil	
	<i>Salmonella typhimurium</i>		
	<i>Salmonella pullorum</i>		
	<i>Salmonella gallinarum</i>		

Following this standard, the level of food safety of EBN can be continuously monitored by the authorities.



Eventhough the bird nests are made up of the nest cement produced by the salivary gland of the birds (Babji *et al.*,2015) solely, EBN also contains a lot of impurities such as feathers, eggs fraction, bird droppings, dirt fleas, and sands which may contaminate the nest (Utomo *et al.*, 2014). Hence, as the EBN is intended for human consumption, it is crucial to evaluate the food from any food borne pathogens which are of major public health concern all over the world.

Thus, the main aim to study were :

1. to determine the presence of *Salmonella* and *E.coli* bacteria in EBNs and guano
- 2.to enumerate the total number of bacteria and coliform in EBNs.

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