

# **UNIVERSITI PUTRA MALAYSIA**

# IN VITRO STUDY OF ELEPHANT HINDGUT FERMENTATION USING FEED AND FAECAL SAMPLE

NABEELAH ANIYYAH BAHARUDIN

FP 2017 112

## IN VITRO STUDY OF ELEPHANT HINDGUT FERMENTATION USING FEED

## AND FAECAL SAMPLE



BY

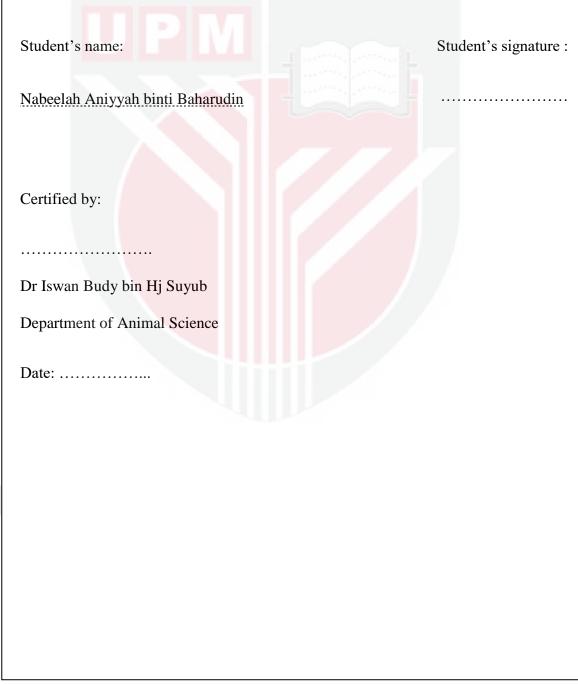
NABEELAH ANIYYAH BINTI BAHARUDIN

A project report submitted to Faculty of Agriculture, Universiti Putra Malaysia, in fulfilment of the requirement of SHW 4999 (Final Year Project) for the award of the degree of Bachelor of Agriculture (Animal Science)

> Faculty of Agriculture Universiti Putra Malaysia 2016/2017

## ENDORSEMENT

This project report entitled *In Vitro* Study of Elephant Hindgut Fermentation using Feed and Faecal Sample is prepared by Nabeelah Aniyyah binti Baharudin, Matric No. 173493 and submitted to the Faculty of Agriculture in fulfilment of the requirement of SHW 4999 (Final Year Project) for the award of the degree of Bachelor of Agriculture (Animal Science).



#### ACKNOWLEDGEMENT

This is a gratitude expression I would like to dedicate to every party involved in the success of completing this project. All praise to Allah s.w.t for allowing me this opportunity of undergoing and completing the course of final year project SHW 4999. First of all, I would like to thank plentifully to my supervisor Dr. Iswan Budy bin Hj Suyub on his support and guidance in my project's progression throughout this semester.

A very grateful I proposed to Zoo Negara, Kuala Lumpur for allowing me the chance to carry out this project and guided through the sampling procedure. Apart from that, I also would like to thank my academic assistance Prof Madya Dr Halimatun Yaakub for the endorsement and concern of my academic progression and on permitting me the opportunity to use the *In Vitro* Laboratory.

Also, thanks to both Syuhada and Haqique as my unofficial project members for all the great teamwork and encouragement given during our studies together. Last but not least, a profusely appreciation I dedicated to my families, lecturers, laboratory assistants, friends, classmates and all individuals who involved directly or indirectly in the project terms and completing this project for the assistance and full support.

To my parents whom never stop praying for me, this is for you.

## CONTENT

Chapter	Торіс	Page No.
1	INTRODUCTION	1
2	LITERATURE REVIEW	3
3	MATERIALS AND METHOD	6
4	RESULT	14
5	DISCUSSION	21
6	CONCLUSION	23
REFEREN	CES	24
APPENDIC	CES	26

C

## LIST OF TABLES

No.	Title Experimental Treatment Variables and Treatments	
Table 3.3		
Table 4.1.1	Mean nutrient composition percentage (%) among feed samples	14
<b>Table 4.1.2</b>	Mean nutrient composition percentage (%) among	16
Table 4.2 <mark>.1</mark>	treatments Cumulative In Vitro Gas Production for each treatments between both faecal and rumen inoculums	18
Table 4.3.1	Comparison of In Vitro Dry Matter Digestibility for faecal and rumen inoculums	20

## LIST OF FIGURES

No.	Title	Page No.
Figure 4.1.1	Mean nutrient composition percentage (%) among treatments	16
Figure 4.2.1	Cumulative <i>In Vitro</i> Gas Production for each treatments between both faecal and rumen inoculums	
Figure 4.3.1	Comparison of <i>In Vitro</i> Dry Matter Digestibility for faecal and rumen inoculums	20

G

Abbreviation	Full Term	
GP	Gas production	
F	Faecal	
R	Rumen	
EF	Elephant feed	
PN	Pakchong Napier	
М	Mixture of EF and PN	
IVDMD	In Vitro Dry Matter Digestibility	

## LIST OF ABBREVIATIONS

No.	Title	Page No
Figure 8.1	Feed samples being prepared for proximate analysis	26
Figure 8.2	Elephant faecal sampling	26
Figure 8.3	Filtered faecal sample	26
Figure 8.4	Incubated syringes of <i>in vitro</i> gas production	
8.5	Composition of the solutions that make up the buffer medium for in vitro gas production;	27
Table 8.6	ANOVA table of feed proximate analysis	28
Table 8.7	ANOVA table of <i>in vitro</i> dry matter digestibility	28
Table 8.8	ANOVA table of <i>in vitro</i> gas production using faecal inoculums	29
Table 8.9	ANOVA table of <i>in vitro</i> gas production using rumen inoculums	30

LIST OF APPENDICES

#### ABSTRACT

Both elephant and cattle are herbivorous animal that differs by site of fermentation but similar in inability of producing fibre-degrading enzyme which instead produced by the microbes in the gut. But it is appears to be that elephant have much lower daily dry matter intake than ruminant's maintenance requirement. Hence, an in vitro gas production (GP) technique is used to study the fermentation ability and compare the rate between elephant and cattle by using faecal and rumen samples as inoculums respectively. The fresh faecal (F) and rumen (R) were collected, filtered and mixed with buffer solution before incubated in 39°C distilled water to digest 3 types of substrates which were the treatments; elephant feed (EF), Pakchong Napier grass (PN) and 50:50 mixture of both feed (M) for 72 hours. Produced gas volume (mL) were recorded every 4<sup>th</sup> hour. In Vitro Dry Matter Digestibility (IVDMD) were determined post-incubation. F versus R inoculums, higher GP recorded for FEF and FPN (0 to 20 and 24<sup>th</sup> hour respectively) but become lower than REF and RPN till incubation periods ends. FPN recorded constantly lower GP than RPN. For IVDMD, comparing with R, F inoculums obtained higher percentage for FEF and FM but lower for FPN. In conclusion, though hypothesis was rejected but this study found that elephants' F inoculums achieved faster asymptotic gas production.

#### ABSTRAK

Gajah dan lembu keduanya merupakan haiwan herbivor yang berbeza lokasi utama fermentasi tetapi sama dari segi ketidakupayaan dalam menghasilkan enzim mendegradasi serat yang sebaliknya dihasilkan oleh mikrob di dalam perut. Akan tetapi, gajah mempunyai pengambilan bahan kering harian yang lebih rendah daripada keperluan penyelenggaraan pengambilan bahan kering haiwan ruminan. Justeru itu, satu teknik penghasilan gas (GP) in vitro digunakan untuk mengkaji keupayaan dan kadar fermentasi antara gajah dan lembu dengan menggunakan sampel najis dan sampel rumen sebagai inokulum. Sampel segar najis (F) dan rumen (R) telah dikumpul, ditapis dan dicampur bersama larutan penampan sebelum diinkubasi di dalam 39°C air suling untuk mencerna 3 jenis substrat yang juga merupakan rawatan kajian; makanan gajah (EF), rumput Napier Pakchong (PN) campuran kedua-dua jenis makanan dengan nisbah 50:50 (M) selama 72 jam. Jumlah penghasilan gas (mL) telah direkod setiap jam ke-4. Prosedur pencernaan bahan kering *in vitro* (IVDMD) telah dilakukan pasca-inkubasi. Inokulum F berbanding R, GP lebih tinggi dicatatkan bagi substrat FEF dan FPN (jam 0 - 20 dan 24 masingmasing) tetapi menjadi semakin rendah daripada REF dan RPN sehingga berakhir tempoh inkubasi. FPN dicatat mempunyai GP yang sentiasa lebih rendah daripada RPN. Bagi IVDMD pula, berbeza dengan R, inokulum F memperoleh peratus yang lebih tinggi bagi FEF dan FM tetapi lebih rendah bagi FPN. Kesimpulannya, walaupun hipotesis ditolak tetapi kajian ini mendapati bahawa inokulum F gajah mencapai penghasilan asimptot lebih cepat. gas yang

viii

#### **CHAPTER 1**

### **INTRODUCTION**

#### 1.1. Background of Study

Elephants are an example of largest surviving herbivores with hind gut fermenter. Similar to other herbivores, the fibre-degrading enzyme in their digestion system is being produced by the gut microflora. The microorganism that populates the gastrointestinal tract is responsible to digest plant fibre in the form of cellulose and hemicelullose. There are several evident differences of the fermentative physiology between hind gut and fore gut fermenters. Fore gut fermenters usually refers to the ruminant animals that ferment the digesta before reaching the abomasums, which is the true stomach. In hind gut fermenter, the fermentation process is similar to the fore gut fermenters (Godoy-Vitorino *et al*, 2012) except apart from having rumen as the site of bacterial fermentation, hind gut fermenters have enlarged caecum located after the stomach and small intestine as the first microbial fermentation site (Fowler, 2006).

### 1.2. Research Problem

Both elephant and ruminant's livestock are herbivore animals which unable to produce fibre-digesting enzyme by itself but being produce by microorganism in the gut. However, study estimated that captive or wild adult Asian elephants daily dry matter intake to be 1.5 - 1.9% of body weight (Hatt, 2006) compared to the dry matter intake for maintenance requirement of ruminant is 3% of

body weight. This questioned whether that elephant have better feed conversion ratio by the gut microflora compared to ruminants.

### **1.3. Research Objectives**

The general objective was to study the elephant hindgut fermentation ability and the specific objective were to compare *in* vitro digestibility between elephant feed and Pakchong Napier grass by faecal inoculums and to compare the fermentation rate between elephant and cattle.

### 1.4. Research Hypothesis

Since elephant and cattle are both herbivores with heavily reliance on microbial fermentation for fibre-degrading enzyme, based on the dry matter intake differences this study hypothesised that the rate of fermentation in elephant hind gut may be better than the rumen fermentation rate.

#### REFERENCES

- Aganga, A. A., et al. (2005), "Chemical composition of napier grass (Pennisetum purpureum) at different stages of growth and napier grass silages with additives." *Journal of Biological Sciences* 5.4 493-496.
- Barrios, Edmundo. (2004). "The *in vitro* dry matter digestibility (IVDMD) method." Modelling Nutrient Management in Tropical Cropping Systems 114: 62.
- Blümmel, M., Makkar, H. P. S., & Becker, K. (1997). In vitro gas production: a technique revisited. Journal of Animal Physiology and Animal Nutrition,77(1-5), 24-34.
- Clubb, R, Georgia M. (2002). A review of the welfare of zoo elephants in Europe. Horsham, UK: RSPCA,
- Demeyer, D.I., De Graeve, E.G., (1991). Differences in stoichiometry between rumen and hindgut fermentation. Adv. Anim. Physiol. Anim. Nutr. 22, 50±61.
- Dumonceaux, G. A.(2006). Digestive System. In *Biology, Medicine, and Surgery* of *Elephants*, ed.
- Fernando, Prithiviraj, Peter L. (2011) "Asian elephants and dry forests." The Ecology and Conservation of Seasonally Dry Forests in Asia: 151-163.
- Fowler, M. E. and Mikota, S. K. (eds) (2006) Front matter, in *Biology, Medicine,* and Surgery of Elephants, Blackwell Publishing Ltd, Oxford, UK. doi: 10.1002/9780470344484.fmatter
- Franz, R., *et al.* (2011). "Intake, selection, digesta retention, digestion and gut fill of two coprophageous species, rabbits (Oryctolagus cuniculus) and guinea pigs (Cavia porcellus), on a hay-only diet." Journal of animal physiology and animal nutrition 95.5: 564-570.
- Getachew, G., *et al.* (2005). "Methane production from commercial dairy rations estimated using an in vitro gas technique." Animal feed science and technology 123: 391-402.
- Godoy-Vitorino *et al.* (2012). "Comparative analyses of foregut and hindgut bacterial communities in hoatzins and cows." The ISME journal 6.3: 531-541.
- Hatt, J-M., and M. Clauss. (2006). "Feeding Asian and African elephants Elephas maximus and Loxodonta africana in captivity." International Zoo Yearbook 40.1: 88-95.
- Makkar, H. P. S. (2002). "Applications of the *in vitro* gas method in the evaluation of feed resources, and enhancement of nutritional value of tannin-rich tree/browse leaves and agro-industrial by-products." Development and Field Evaluation of Animal Feed Supplementation Packages, IAEA-TECDOC-1294, IAEA, Vienna: 23-39.

- Mauricio, Rogerio M., *et al.* (2001), "Comparison of bovine rumen liquor and bovine faeces as inoculum for an *in vitro* gas production technique for evaluating forages." *Animal Feed Science and Technology* 89.1: 33-48.
- Pandian, C. S., *et al.* (2016). "Faecal matter as inoculum for *in vitro* gas production technique." *Animal Nutrition and Feed Technology* 16.2:271-281.
- Pellikaan, W. F., *et al.* (2011). "A novel method to determine simultaneously methane production during in vitro gas production using fully automated equipment." Animal feed science and technology 168.3: 196-205.
- Ramin, M., *et al.* (2015). "Comparison of rumen fluid inoculum vs. faecal inoculum on predicted methane production using a fully automated in vitro gas production system." Livestock Science 181:65-71.
- Rymer, C., *et al.* (2005). "In vitro cumulative gas production techniques: History, methodological considerations and challenges." Animal Feed Science and Technology 123: 9-30.
- Sakaguchi, Ei. (2003). "Digestive strategies of small hindgut fermenters." Animal Science Journal 74.5: 327-337.
- Tagliapietra, F., *et al.*, (2010). In vitro rumen fermentation: effect of head space pressure on the gas production kinetics of corn meal and meadowhay. Anim. FeedSci. Technol. 158, 197–201.
- Ullrey, Duane E., Susan D. Crissey, and Harold F. Hintz. (1997). Elephants: Nutrition and dietary husbandry. Nutrition Advisory Group,.