



UNIVERSITI PUTRA MALAYSIA

***OPTIMIZATION OF LIQUID BIO-FERTILIZER AND COCO PEAT
APPLICATION ON NUTRIENT UPTAKE AND LOSSES OF PAK CHOI
(Brassica chinensis L.) VIA RSM APPROACH***

ANNA RENLY

FP 2021 47



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By

ANNA RENLY

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

June 2020

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Master of Science

OPTIMIZATION OF LIQUID BIO-FERTILIZER AND COCO PEAT APPLICATION ON NUTRIENT UPTAKE AND LOSSES OF PAK CHOI (*Brassica chinensis* L.) VIA RSM APPROACH

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ANNA RENLY

June 2020

Chair : Tee Tuan Poy, PhD
Faculty : Agriculture

Optimal liquid bio-fertilizer application rate and frequency with coco peat as growth media were estimated using Response Surface Methodology (RSM) to maximize Nitrogen Uptake Efficiency (NUE) and minimize Nitrogen loss (N loss) with favourable Pak Choi yield. A central composite design (CCD) was conducted to determine the experimental design of Pak Choi into 20 treatments based on 5 level codes ($-\alpha$, -1 , 0 , 1 , α) for three factors, as assigned coco peat (CP) (0%, 25%, 50%, 75%, 100%), bio-fertilizer rate (BFR) (0, 50, 100, 150, 200 kg N/ha) and application frequency (AF) (subsequent 1, 2, 3, 4, 5 day), respectively. The growth performance of plant height, girth, and fresh weight was measured at harvesting day and analysed the nutrients composition. These field data were used to determine the optimum value of plant yield, N loss and NUE by full quadratic polynomial model generated from RSM. The RSM analysis optimum output indicated combination of 25% CP with 150 kg N/ha of BFR at subsequent 4 days that produced the best growth performance on plant height, girth and yield of 11.88 cm, 9.31 cm, and 18.29 kg, respectively. No significant interaction of three factors found on the nutritive compositions (N, P, K, Ca and Mg) by RSM. Within RSM optimization process, an ANOVA output indicated significant terms that fitted two-factor interaction models on yield and NUE as well as a quadratic model on N loss which described their relationships found in this study. These two models enabled to predict response optimization range based on maximizing yield and NUE while minimizing N loss. The models result was predicted Pak Choi maximum yield (≥ 10378.43 kg/ha) were obtained at optimum range: 0 - 20% of CP with 155 - 200 kg N/ha of BFR and AF at subsequent day 4 to day 5. The maximum NUE (≥ 47.75 kg yield/ kg N) were obtained at the range of 0 - 20% CP, 175 - 200 kg N/ha BFR and subsequent 4 to 5 day of AF. While, 0 - 50% of CP, 175 - 200 kg N/ha of BFR and subsequent 3 - 5 day of AF were predicted to minimize N loss (≤ 13.86 kg N/ha). In line with these models optimum output ranges, the models could be used to predict the optimum value by setting the goal of yield, NUE and N loss align with adopting a numerical optimization function. Hence for example, this study was targeted to maximize the NUE (++++), minimize the N loss (++++), and maximize the yield

(+++ based on the degree of importance (+, the most “+”, the greater of desire). The output of predication shows the combination of 19% CP, 190 kg N/ha of BFR and AF at subsequent 4 day was given the best optimum value at maximum of NUE of 49.60 kg yield/kg N, with minimum N loss of 1.05 kg N/ha, ultimately achieved better Pak Choi yield of 10739.02 kg/ha. These results were used to conduct a validation experiment on field to validate the model prediction accuracy. Validation result proven a small deviation between the expected and observed value for yield (10725.47 kg/ha), NUE (47.07 kg yield/kg N), and N losses (1.09 kg N/ha) at 0.12%, 5.10%, 3.38%, respectively. This expresses that the models are valid and fit to be used for prediction on Pak Choi yield, NUE and N loss. In conclusion, this study proven that RSM model simulation result shows a promising Pak Choi yield, along with better NUE, while reduces in N losses at the value of CP < 20 %, together with BFR value < 200 kg N/ha at subsequent 4 days of AF. This also indicated that reduced in the cost of fertilizer and significant to environmental friendly (less pollution) by applied lower rate of fertilizer than common farming applied (200 kg N/ha). In addition, farmer able to achieve their targeted yield and reduce the fertilizer cost by using the model obtained from the RSM optimization. The research finding highlighted utmost effect of liquid bio-fertilizer and coco peat on Pak Choi yield, NUE and N loss.

Abstrak teisis yang dikemukakan kepada Senat Universiti Putra Malaysia
Sebagai memenuhi keperluan untuk ijazah Master Sains

**MENGOPTIMUMKAN PENGGUNAAN BAJA BIO CECAIR DAN SABUT
KELAPA TERHADAP PENGAMBILAN DAN KEHILANGAN NUTRIEN
PADA PAK CHOI (*Brassica chinensis* L.) MENGGUNAKAN PENDEKATAN
RSM**

Oleh

ANNA RENLY

Jun 2020

Pengerusi : Tee Tuan Poy, PhD
Fakulti : Pertanian

Kadar dan frekuensi aplikasi baja bio cair yang optimum dengan gambut kelapa sebagai media pertumbuhan dianggarkan menggunakan Metodologi Permukaan Respons (RSM) untuk memaksimumkan kecekapan pengambilan Nitrogen dan meminimumkan kehilangan Nitrogen dengan hasil Pak Choi yang baik. Reka bentuk komposit pusat (CCD) dilakukan untuk menentukan reka bentuk eksperimen Pak Choi ke dalam 20 rawatan berdasarkan 5 tahap kod ($-\alpha$, -1 , 0 , 1 , α) untuk tiga faktor, seperti ditetapkan coco gambut (0%, 25%, 50%, 75%, 100%), kadar baja bio (0, 50, 100, 150, 200 kg N/ha) dan kekerapan aplikasi (setiap 1, 2, 3, 4, 5 hari). Prestasi pertumbuhan tinggi tanaman, lilitan, dan berat segar diukur pada hari penuaian dan menganalisis komposisi nutrien. Data lapangan ini digunakan untuk menentukan nilai optimum hasil tanaman, kehilangan N dan kecekapan pengambilan N oleh model polinomial kuadratik penuh yang dihasilkan dari RSM. Hasil analisis optimum RSM menunjukkan kombinasi 25% gambut kelapa dengan 150 kg N / ha kadar bio baja pada setiap 4 hari menghasilkan prestasi pertumbuhan terbaik pada 11.88 cm ketinggian, 9.31 cm lilitan dan hasil tanaman 18.29 kg. Tiada interaksi yang signifikan dari tiga faktor terhadap komposisi nutrien (N, P, K, Ca dan Mg) oleh RSM. Dalam proses pengoptimuman RSM, keluaran ANOVA menunjukkan istilah penting yang sesuai dengan model interaksi dua faktor pada hasil dan NUE serta model kuadratik kehilangan N yang menggambarkan hubungan mereka yang terdapat dalam kajian ini. Kedua-dua model digunakan untuk meramalkan jangkauan pengoptimuman tindak balas dengan memaksimumkan hasil dan kecekapan pengambilan N sambil meminimumkan kehilangan N. Model meramalkan hasil maksimum bagi Pak Choi (≥ 10378.43 kg/ha) diperoleh pada julat optimum: 0 - 20% gambut kelapa dengan 155 - 200 kg N/ha kadar bio baja dan kekerapan aplikasi setiap 4 ke 5 hari. Maksimum kecekapan pengambilan N (47.75 kg hasil /kg N) diperoleh pada julat 0 - 20% gambut kelapa, 175 - 200 kg N/ha kadar bio baja setiap 4 ke 5 hari berikutnya. Sementara, 0 - 50% gambut kelapa, 175 - 200 kg N/ha kadar bio baja dan kekerapan setiap 3 ke 5 hari berikutnya diramalkan akan mengurangkan kehilangan N (≤ 13.86 kg N/ha). Model ini digunakan

untuk menganggarkan nilai optimum dengan menetapkan sasaran hasil, kecekapan pengambilan N dan kehilangan N dengan mengadopsi fungsi pengoptimuman numerik. Sebagai contoh, kajian ini disasarkan untuk memaksimumkan kecekapan pengambilan N (++++), meminimumkan kehilangan N (++++) dan memaksimumkan hasil (+++) berdasarkan tahap kepentingan (+, yang paling banyak “+”, semakin besar keinginan). Hasil ramalan menunjukkan gabungan 19% gambut kelapa, 190 kg N/ha kadar bio baja dan kekerapan aplikasi setiap 4 hari berikutnya memberi nilai optimum terbaik dalam memaksimum kecekapan pengambilan N 49.60 kg hasil/kg N, dengan minimum kehilangan N 1.05 kg N/ha, turut menghasilkan hasil Pak Choi yang lebih baik iaitu 10739.02 kg/ha. Hasil ini digunakan untuk melakukan eksperimen validasi di lapangan untuk mengesahkan ketepatan ramalan model. Hasil pengesahan membuktikan penyimpangan kecil antara nilai yang diharapkan dan diperhatikan untuk hasil (10725.47 kg / ha), kecekapan pengambilan N (47.07 kg hasil/kg N), dan kehilangan N (1.09 kg N/ha) pada 0.12%, 5.10%, 3.38%, masing- masing. Ini menyatakan bahawa model-model ini sah dan sesuai untuk digunakan untuk membuat ramalan untuk hasil Pak Choi, kecekapan pengambilan N dan kehilangan N. Kesimpulannya, kajian ini membuktikan bahawa hasil simulasi model RSM menunjukkan hasil Pak Choi yang menjanjikan, bersama dengan kecekapan pengambilan N yang lebih baik, sementara penurunan kehilangan N pada nilai CP <20%, bersamaan dengan nilai kadar bio baja <200 kg N / ha pada kekerapan aplikasi setiap 4 hari. Ini juga menunjukkan berlaku pengurangan kos baja dan mesra alam (kurang pencemaran) dengan kadar baja yang lebih rendah daripada penggunaan pertanian biasa (200 kg N ha). Di samping itu, petani dapat mencapai hasil yang disasarkan dan mengurangkan kos baja dengan menggunakan model yang diperoleh dari pengoptimuman RSM. Penemuan kajian menunjukkan pengaruh besar baja bio cair dan gambut coco terhadap hasil Pak Choi, kehilangan kecekapan pengambilan N dan kehilangan N.

ACKNOWLEDGEMENT

First and foremost, I would like to acknowledge and express the deepest appreciation to my supervisor, Dr Tee Tuan Poy for her guidance, support and advice through the duration of my research. I appreciate the contribution made by each of my committee members, Prof Ir. Dr Chin Nyuk Ling, Prof Dr Dahlan Bin Ismail and Prof Madya Dr Siti Aishah Binti Hassan for all their invaluable suggestions, experience they provided me and generosity of using their resource and motivation that keep me positive during this study.

I also wish to express my sincere gratitude to staffs at Nutrition Lab and Livestock Waste Lab, Animal Science Department, Faculty of Agriculture, UPM Serdang, Selangor for their assistance during lab analysis.

Last but not least, I would like to thank my family for understanding and supporting me, to my beloved husband, Mr Stenly, for your love, support and confidence in me all these years. Thank you to my lovely parent, Mr Renly and Mdm Langai for their encouragement and endless love for raising me the way I am and putting me on the right path. To family and friends thanks for their unconditional loves, constant support and source of inspiration.

Most of all, thanks to God.

This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfillment of the requirement for the degree of Master of Science. The members of the Supervisory Committee were as follows:

Tee Tuan Poy, PhD
Senior Lecturer
Faculty of Agriculture
Universiti Putra Malaysia
(Chairman)

Chin Nyuk Ling, PhD
Professor Ir.
Faculty of Engineering
Universiti Putra Malaysia
(Member)

Dahlan Bin Ismail, PhD
Professor
Faculty of Agriculture
Universiti Putra Malaysia
(Member)

Siti Aishah Binti Hassan, PhD
Associate Professor
Faculty of Educational Studies
Universiti Putra Malaysia
(Member)

ZALILAH MOHD SHARIFF, PhD
Professor and Dean
School of Graduate Studies
Universiti Putra Malaysia

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Signature: _____

Name of Chairman of
Supervisory
Committee: Dr Tee Tuan Poy

Signature: _____

Name of Member of
Supervisory
Committee: Prof. Ir. Chin Nyuk Ling

Signature: _____

Name of Member of
Supervisory
Committee: Prof. Dr. Dahlan Bin Ismail

Signature: _____

Name of Member of
Supervisory
Committee: Prof. Madya Siti Aishah

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LIST OF ABBREVIATIONS

AF	Application Frequency
BFR	Bio-fertilizer Rate
C	Carbon
Ca	Calcium
C:N	Carbon to Nitrogen ratio
CO ₂	Carbon Dioxide
CP	Coco Peat
Cu	Copper
DM	Dry Matter
DOA	Department of Agriculture
EM	Effective Microorganism
FR	Fertilizer Rate
K	Potassium
Mg	Magnesium
N	Nitrogen
NL	Nitrogen Losses
NUE	Nitrogen Uptake Efficiency
P	Phosphorus
PY	Plant Yield
RSM	Response Surface Methodology
Zn	Zinc
AF	Application Frequency

CHAPTER 1

INTRODUCTION

In Malaysia, livestock practices in term of waste management is always recklessly concerned and resulted in various environmental pollution issues, mainly due to inappropriate and “unrestricted” discharge of livestock waste (i.e. animal manure, bedding, animal food waste) into environment. In fact, these wastes can be recycled, reused and composed as bio fertilizer through conventional windrow method or advance in-vessel systems. The latest technology particularly, in-vessel composting that requires less space, shorter period and provides better control condition which can produce both solid and liquid form of compost known as bio fertilizer. According to Pangnakorn *et al.* (2009), bio fertilizer defined as a product from bio fermentation or digestion of vegetables, fruits and animal wastes fermented with sugar and effective microorganisms. It contains living microorganisms that may enhance the physical and chemical condition of soil and potentially useful for the efficient growth of plants.

The bio-fertilizer in liquid or solid form can substitute the expensive chemical fertilizer, as well as conserves the soil fertility as it caused by depleting soil organic matter in the long-term application of chemical substrate (Sundaravarathan and Kannaiyan, 2002). In the previous studies by (Harris, 2003; Schloter *et al.*, 2003; Timo *et al.*, 2004; Zhou and Ding, 2007; Ma *et al.*, 2010) indicated that compost or bio fertilizer was a good solution to replenish the loss of soil organic matter via sequesterate the soil carbon in reduce the CO₂ emission, as well as a good preventing the environment pollution. In addition, the utilization of bio fertilizer promotes organic farming concept while may give extra profit to farmer in substituting the expenses of mineral fertilizer/insecticide/pesticide used, also, promoting healthy food consuming with better quality (safe and nutritious) as a bio product. This bio-products produce from organic farming reduces dependence on non-renewable resources, which recycles by product from household, agriculture and other human activities.

Solid bio fertilizer as product of composting has been commonly used in study to determined bio fertilizer effect on planting compared to liquid form bio fertilizer. In this study, abundance of cattle manure with combination of food waste were composed inside in-vessel composter for 30 days to generate the liquid bio fertilizer. The liquid were collected and used to study its effect on Pak Choi (*Brassica chinensis*) growth and also effect on nutrients loss to the environment. In the study, Pak Choi used as a test crop and harvested at 42 to 75 days of planting. The Pak Choi were chosen due to it is a perennial plant grown commercially as an annual, native to China but is extensively grown throughout Asia and common prefer vegetable being consumed in Malaysia. Pak Choi also called Chinese white cabbage is high valued fresh cut vegetables. It has a light, sweet flavour, crispy texture, and high nutritional values, rich in minerals and vitamins (Able *et al.*, 2003; Lu, 2007).

In planting, growth media is known to be played an important role in germination rate and physiological parameters namely, plant height, number of leaves, yield and etc.

(Vendrame *et al.*, 2005). The media is important to supply roots with nutrients, air and water, allows for maximum root growth and provides physical support to the plant. Common plant media such as coco peat or coconut fiber is an alternative organic material that often being used as growing media due to it easily accommodates water in the pores. It can be a safety substitute for soil and healthy plants because characteristics of coco peat which saves a lot of water and air rich pores growth for seed germination (Awang *et al.*, 2009).

In line with abundance and availability of liquid bio fertilizer was produced but under-utilized that lead to waste of nutrients that available to be utilized by plant. The study aimed to investigate the optimum utilization of bio-fertilizer rate, application frequency and coco peat using Response Surface Methodology (RSM) approach. RSM, a statistical or mathematical technique for analysis of problems where an optimum response of interest influenced by several variables (Montgomery, 2005).

The RSM optimization analysis involves the processes of single and interactive effect of important factors (CP, BFR and AF) on Pak Choi yield, NUE and N loss value. The values of were investigated and optimized through response surface methodology (RSM), Model generate by RSM is able to display the optimum condition of maximum liquid bio fertilizer efficiency and allows prediction of nutrients loss and uptake in specific fertilizer application frequency and amount with growth media ratio according to the plant yield. Finally, best combination of liquid bio fertilizer application frequency and amount with growth media generated by RSM model then used to replanting for validation.

This study consists of three major steps (a) field experiment data collection, (b) RSM analysis and field validation. The general objective of study was investigated the response of the plant growth in terms of height, girth and fresh weight (yield) and plant nutritive compositions (N, P, K, Ca and Mg) that influent by the factors (independent variables) assigned as CP, BFR and AF. These response field data were collected to determine the Pak Choi yield, nitrogen uptake efficiency and nitrogen loss by RSM.

1.1 Specific Objectives

The specific objectives of this study were: -

- (i) To determine the effect of cattle liquid bio fertilizer application rate (concentration) and frequency on Pak Choi's growth traits and nutritive values at different coco peat ratio.
- (ii) To determine the effect of cattle liquid bio fertilizer application rate (concentration) and frequency on nutrients losses and nutrients uptake at different coco peat ratio.

- (iii) To determine the optimum fertilizer rate (amount), fertilizer frequency and coco peat that maximizes the plant nutrient uptake and minimizes the losses into the environment.

1.2 Significant Study

Desired production of Pak Choi and best utilization of liquid bio fertilizer can be achieved if the optimization model of Pak Choi has been developed. This study aims at improving the use of RSM to optimize nutrient uptake and losses of Pak Choi with favourable yield. This research will give new information about the relationship between coco peat, bio-fertilizer rate and application frequency, particularly the utilization of bio-fertilizer and its optimization on growth performance, nutritive value, nitrogen uptake efficiency and nitrogen losses.

REFERENCES

- Abdul Rahim, K. (2002). Biofertilizer in Malaysia agriculture : Perception, demand and promotion. *FNCA Joint Workshop on Mutation, Breeding and Bio-fertilizer*.
- Able, A.J., Wong, L.S., Prasad, A. and O'hare, T.J. (2003). The effect of *I-methylcyclopropane* on the shelf life of minimally processed leafy Asian vegetable. *Post-harvest Biology and Technology*, 27:157-161.
- Albiach, R. R. (2000). Microbial biomass content and enzymatic activities after application of organic amendments to horticultural soil. *Bioresource Technology*, 75:43-48.
- Amini, M. and Younesi H. (2009). Bioabsorption of Cd(II), Ni(II) and Pb(II) from aqueous solution by dried biomass of *Aspergillus niger*: Application of response surface methodology to the optimization of process parameters. *CLEAN-Soil, Air, Water*, 37(10): 776-786.
- Anna Witek-Krowiak, K. C. (2014). Application of response surface methodology and artificial neural network methods in modelling and optimization of biosorption process. *Bioresource Technology*, 160: 150-160.
- Anuar, N. (2014). *Effective microorganism effect on nutrients and economic value of broiler chicken manure*. (Unpublished bachelor's thesis). University Putra Malaysia, Malaysia.
- AOAC. (1990). *Official Method of Analysis. 15th Edition*. Washington, USA: Association of Official Analytic Chemists.
- Awang, Y., Shaharom, A., Mohamad, R, and Ahmad, S. (2009). Chemical and physical characteristics of cocopeat-based media mixture and their effect on the growth and development of celosia cristata. *America Journal of Agricultural and Biological Science*, 4 (1): 63-71.
- Barneix, A. (2007). Physiological and biochemistry of source-regulated protein accumulation in the wheat grain. *Journal of Plant Physiological*, 164: 581-590.
- Bernal, M.P., Albuquerque, J.A. and Moral, R. (2009). Composting of animal manure and chemical criteria for compost maturity assessment. *Bioresource Technology*, 100 (22): 5444-5453.
- Bhardwaj, D., Ansari, M.W. and Sahoo, R.K. (2014). Biofertilizers functions as key player insustainable agriculture by improving soil fertility, plant tolerance and crop productivity. *Microb Cell Fact*, 13: 66.

- Bolan, N. (2004). Distribution and bioavailability of trace elements in livestock and poultry manure by-products. *Environment Science Technology*, , 34:291-338.
- Box, G.E.P. and Wilson, K.B. (1951). On the experimental attainment of optimum conditions. *Statistic Social*, 13:1- 45.
- Braber, K. (1995). Anerobic digestion of munucipal solid waste: a morden waste disposal option on the verge of breakthrough. *Biomass Bioenergy*, 9(1-5): 365-376.
- Cameron, H. (2002). Nitrate leaching in temperate agroecosystems:sources, factors and mitigating strategies. *Nutrient Cycling in Agroecosystems*, 46 , 237-256.
- Cameron, K. (2013). NItrogen losses from the soil/plant system: a review. *Annals of Applied Biology*, 162 , 145-173.
- Cartea, M. E., Francisco, M., Soengas P. and Velasco, P. (2011). Phenolic compounds in *Brassica* vegetables. *Moleculas*, 16:251-280.
- Cassidy, E.S., West, P.C., Gabber, J.S. and Foley, J.A. (2013). Refining agriculture yields : from tonnes people nourish per hactare. *Environment Resource Letter*, 8, 1-8.
- Cassman, K.G., Dobermann, A. and Walters, D.T. (2001). Agrosystems, nitrogen use efficiency and nitrogen management. *AMBIO*, 31: 132-140.
- Castelli, F., Miceli F. and Piro F. (1995). Effect of harvesting and curing method on tobacco burley at different nitrogen fertilizer and plant population densities. *Agronomia*, 24: 308-316.
- Conder, C.C. (2014). *Modelling studies on nutrients (N,P,K) and economic value of goat's manure in farm.* (Unpublished bachelor's thesis). University Putra Malaysia, Malaysia.
- Cordell, M. and Verderame, F.D. (1954). The determination of nitrogen in copper-titanium alloys. *Analytica Chemica Acta*, 11: 40-47.
- Dadkhah, A. (2012). Effect of chemical and bio-fertilizerz on yield, growth parameters and essential oil contents of funnel (*Foeniculun vulgare Miller*). *Journal of Medicinal Plants and By-products*, 2 , 101-105.
- De Baere, L. (2000). Anaerobic degestion of solid waste: state of the art. *Water Science Technology*, 41(3): 283-290.
- Deniz, C., Demirci, A., Graves, R.H. and Davitt, N.H. (2005). Applicability of optimised in-vessel food waste composting for windrow system. *Biosystem Engineering*, 91(4) 479-486.

- Dominquez, J. (2011). Relationship between composting and vermicomposting . . *Vermiculture Technology Earthworms, Organic Wastes and Environmental Management*, 11-26.
- Dwivedi, B.S. and Meena, M.C. (2017). Efficient nitrogen management under predominant cropping system of india. *The Indian Nitrogen Assessment*, 95 - 115.
- Eckert, D. (2020). *Nitrogen*. Retrieved from The Mosaic Company: <https://www.cropnutrition.com/nutrient-management/nitrogen>
- Edelman, W. and Engeli, H. (1993). Combined digestion and composting of organic industrial and manucpal wastes in Switzeland. *Water Science Technology*, 27(2): 169-182.
- EEC. (1991). Protection of water against pollution caused by nitrates from agriculture.
- Fageria N. K. and Baligar, V.C. (2005). Enhancing nitrogen use efficiency in crop plants. *Advance Agronomic*, 88: 97-185.
- Fageria, N. (2014). *Nitrogen management in crop production*. New York: CRC Press.
- FAO. (2012). Current World Fertilizer Trends and Outlook to 2016. *Food and Agriculture Organization of the United Nations*. Rome.
- FAO. (2012). *FAO Statistical Year Book*. Retrieved from <http://www.fao.org/docep/015/i2490e00.htm>
- Gibbs, P. R. (2002). Environmental impact of cattle manure composting. *Microbiology of Composting*, 445-456.
- Goel, A. (1999). Use of bio fertilizer : Potential, constraints and future strategies review. *International Journal of Tropical Agriculture*, 17:1-18.
- Goulding, K. J. (2008). Optimizing nutrient management for farm systems. *Philosophical Transactions of the Royal Society of London. Series B, . Biological Sciences* , 363 , 667-680.
- Harris, J.A. (2003). Measurement of the soil microbial community for estimating the success of restoration. *European Journal of Soil Science*, 54: 801-808.
- He, M. W. (2009). Effect of composting process on phytotoxicity and speciation of copper, zin and lead in sewage sludge and swine manure. *Waste Management*, 29 : 590-597.

- Hill, T. (1990). The effect of nitrogenous fertilizer and plant spacing on the yield of three chinese vegetables-Kai Lan, Tsoi Sum and Pak Choi. *Scientia Horticulture*, 45 (1-2): 11-20.
- IFOAM (2005). (04 11, 2013). *Wikipedia*. Retrieved from Principles of organic agriculture:
https://en.m.wikipedia.org/wiki/Principles_of_Organic_Agriculture#:~:text=1%20principles,them%20and%20help%20sustain%20them.
- Intensive animal farming*. (11, 2017). Retrieved from Wikipedia:
https://en.wikipedia.org/w/index.php?title=intensive_animal_farming&oldid=966349263
- Iversen, G. A. (1987). *Analysis of variance(Quantitative application in social science)*. Thousand Oaks, CA: SAGE Publication Inc.
- James C. As cough II, E. M. (2013). Using response surface methodology for economic and environmental trade-offs at the farm level. *Air, Soil and Water Research* , 6:73-89.
- Jarvis, S., Hutchings, N., Brentrup, F., Olesen, J.E., Van Der Hock, K.W. (2011). Nitrogen flows in farming system across Europe. In M. H. Sutton, *The European Nitrogen Assesment* (pp. 211-228). New York: Cambridge University Press.
- Joung, D.K., Joon, S.E., Yung, H.I., Dae, K.K and Wan, N. (2008). Evaluation of pilot-scale in vessel composting for food waste treatment. *Journal of Hazardous Materials*, 272-277.
- Kala, D. (2011). Commercial organic fertilizers and their labelling in Malaysia. *Management Journal of Soil Science* , 15:147-157.
- Keerthi, P. R. (2017). Effect of sowing time and nitrogen on growth, yield and nutrient uptake. *Chemical Science Review and Letters*, 6(24), 2526-2532.
- Lakesh, K.S and Sukhninder, K.B. (2018). A review of methods to improve nitrogen uptake efficiency in agriculture. *Sustainability*, 10: 51.
- Larney, F. A. (2007). A review of composting as a management alternative for beef cattle feedlot manure in Southern Alberta, Canada. *Bioresource Technology* , 98:3221-3227.
- Lazcano, C. and Dominguez, J. (2011). *The use of vermicompost in sustainable agriculture: Impact on plant growth and soil fertility*. Nova Science Publishers.

- Lazcona, C. M. (2008). Comparison of the effectiveness of composting and vermicomposting for the biological stabilization of cattle manure. *Chemosphere*, 7: 1013-1019.
- Lim, W.J. (2018) *Modelling of Pilot -Scale Anaerobic Waste Composting Process with Dry Leaves or Cow Manure*. (Published bachelor's thesis). University Putra Malaysia, Malaysia.
- Li-zhilin, S. R. (1997). Physiological effect of nitrogen application on aromatic rice. *Journal of South China Agricultural University*, 18(3): 13-17.
- Lopez Alonso, M., Benedito, J.L., Miranda, M., Castillo, C., Hernandez, J. and Shore, R.F. (2000). The effect of pig farming on copper and zinc accumulation in cattle in Galicia (North-Western Spain). *Veterinary Journal*, 160: 259-266.
- Lu, S. (2007). Effect of packaging on shelf life of minimally processed Bok choy (*Brassica chinensis L.*). *Food Science and Technology*, 40:460-464.
- Ma, N.N., Li, T.L. Wu, C.C. and Zhang, E.P. (2010). Effects of long term fertilization on soil enzyme activities and soil physiochemical properties of facility vegetable field. *Chinese Journal of Applied Ecology*, 21, 1766-1771.
- Malaysia, D. O. (21 2, 2019). *Department of Veterinary Services, Minisrty of Agriculture & Agro Based Industry*. Retrieved from <https://www.dvs.gov.my>
- Majuntin, J. (2010). *Estimation on manure nutrients value in malaysia pig farm*. (Unpublished bachelor's thesis). University Putra Malaysia, Malaysia.
- Mansouri, H. M. (2014). Management of nitrogen fertilizer, irrigation and plant density in onion production using response surface methodology as optimization approach. *African Journal pf Agricultural Research*, 9(7): 676-687.
- Marscher, H. (1995). *Mineral nutrition of higher plants*. London: Academic Press.
- Martinez, J., Pabert, P., Barrington, S., & Burton, C. (2009). Livestock waste treatment system for environmental quality, food safety and sustainability. *Bioresource Technology*, 100: 5527-5536.
- Mata-Alvarez, J., Mace, S. and Liabres, P. (2000). Anaerobic digestion of organic solid wastes. A overview of research achievements and perspectives. *Bioresource Technology*, 74 : 3-16.
- McLellan, E.L., Cassman, K.G., Eagle, A.J., Woodbury, P.B., Sela, S., Tonitto, C. and Marjerison, H.M. (2018). The nitorgen balancing act: Tracking thr environemntal performance of food production. *Bioscience*, 68(3): 194-203.

- Mishra, D.J., S. Rajvir, Mishra U.K. and S.S. Kumar. (2012). Role of bio-fertilizer in organic agriculture: A Review. *Research Journal of Recent Science*, 2:39-41.
- Mohd Hanafiah, N.H., Samsuri, S., Yusup, S. and Amran, N.A. (2019). Effect of nutrients on the growth of pak choi (*Brassica chinensis L.*) seedlings in a hydroponic system. *PLATFORM- A Journal of Science and Technology*.
- Moll, R. E. (1982). Analysis and interpreting of factors which contribute to efficiency of nitrogen utilization. *Agronomic Journal*, 74:562-564.
- Montgomery, D. (2005). *Design and Analysis of Experiments (6th ed.)*. New York: John Wiley & Sons.
- Motsara, M.R. and Roy R.N. (2008). *Guide to laboratory establishment for plant nutrient analysis*. Rome: Food and Agriculture Organization of the United Nation.
- Myers, R. A. (2002). *Response Surface Methodology*. Wiley, New York, 10.
- Nitrogen*. (2020). Retrieved from The Mosaic Company.
- Norton, R.D., and Roberts, T. (2015). Nitrogen use efficiency and nutrients performance indicators. *Global Partnership on Nutrient Management Task Team Workshop*, (pp. 1-14). Washington D.C.
- Nuran, B. (2007). The response surface methodology. *Graduate Faculty, Indiana University South Bend*.
- Agriculture and Food Organization (2008). *Guide to laboratory establishment for plant nutrient analysis*. Rome.
- Pain, B. (1999). Gaseous pollutants from organic waste use in agriculture. *International Conference of the FAO network on recycling agricultural, municipal and industry residues in agriculture (Ramiran 98)* (pp. 233-246). Rennes: Cemegref FAO Editions.
- Pandey, N. (2018). Role of plant nutrients in plant growth and physiology. *Plant Nutrients and Abiotic Stress Tolerance*, 51-93.
- Pangnakorn, U. S. (2009). Application of wood vinegar to fermented liquid bio fertilizer for organic agriculture on soybean. *Asia J. Food & Agro Ind.*, 189-196.
- Piya, S., I. Shrestha, D.P. Gauchan and J. Lamichhane. (2018). Vermicomposting in organic Agriculture: Influence on the soil nutrients and plant growth. *International Journal of Research*, 5(20): 1055-1063.

- Pushpa, T., Sekaran, V., Basha, S.J. and Jegan, J. (2016). Investigation on preparation, characterization and application of effective microorganisms (EM) based composts-an ecofriendly solution. *Nature Environment and Pollution Technology*, 15:153-158.
- Rathke, G.W., Behrens, T. and Diepenbrock, W. (2006). Integrated nitrogen management strategies to improve seed yield, oil content and nitrogen efficiency of winter oilseed rape (*Brassica napis L.*): A review. *Agriculture Ecosystem and Environment*, 117 (2-3): 80-108.
- Raun, W.R. and Johnson, G.V. (1999). Improving nitrgen use efficiency for cereal production. *Agronomic Journal*, 91: 357-363.
- Renly, A. (2009). *Modeling and evaluation of nutrients flow for feedlot cattle manure in malaysia.* (Unpublished bachelor's thesis). University Putra Malaysia, Malaysia
- Ruiz, J.M. and Ramero, L. (1999). Cucumber yield and nitrogen metabolism in response to nitrogen supply. *Science Horticulture*, 82: 309-316.
- Sabier, K.S., Ahmed, S.A., Hassan, I.A. and Ahmed , P.H. (2015). Effect of bio-fertilizer and chemical fertilizer on growth and yield in cucumber (cucumis sativus) in green house condition. *Journal of Biological Science*, 18 : 129-134.
- Sakawi, Z. and Ismail, L. (2015). Managing odour pollution from livestock sources in Malaysia: Issue and challenges. *Malaysia Journal of Society and Space*, 13: 86-103.
- Sakeri, Z and Ismail, L. (2015). Managing odour pollution from livestock sources in malaysia: Issues and challenges. *Malaysia Journal of Society and Space*, 11 (13): 96-103.
- Sampaio, F.C., D.DeFaveri, H.C. Montovani, F.M.L Passos, P. Perego and A. Converti. (2006). Use of reeponse surface methodology for optimization of xylitol production y the new yeast strain Debaryomyces hansenii UFV-170. *Journal of Food Engineering*, 76: 376-386.
- Schloter, M., Dilly, O. and Munch. J.C. (2003). Indicators for evaluating soil quality. *Agriculture, Ecosystems and Environment*, 98: 255-262.
- Sekeran, V. C. (2005). Evaluation of effective microorganism in solid waste management. . *Alagappa Chettiar College of Engineering and Technology* .
- Shristi Piya, I. S. (2018). Vermicomposting in organic Agriculture:Influnce on the soil nutrients and plant growth. *International Journal of Research*, 5(20): 1055-1063.

- Srivastava, P., Gupta, M., Upadhyaya, R., Sharma, S., Shikha, Singh, N., Tiwari, S. and Singh, B. (2012). Effect of combined application of vermicompost and mineral fertilizer on the growth of *Allium cepa* L. and soil fertility. *Journal Of PLant Nutrition Soil Science*, 175:101-107.
- Subedi, K.D. and Ma, B.L. (2005). Nitrogen uptake and partitioning in stay-green and leafy maize hybrids. *Crop Science*, 45:740-747.
- Sundaravarathan, S and Kannaiyan, S. (2002). Influence of *Azolla* and *Sesbania rostrata* application changes in microbial population and enzymes in rice soils. *Biotechnology of Biofertilizers*, 225-251.
- Sung, Y. and Chen, S.W. (2018). Response of nitrogen metabolism in pak choi plants treated with different sodium selenate (Na_2SeO_4) concentrations. *The Japanese Society for Horticultural Science*.
- Tajeda, M. G. (2008). Agricultural use of leachates obtained from two different vermicomposting process. *Bioresources Technology*, 99 : 6228-6232.
- Timo, K., Stephen, W. and Frank, E. (2004). Microbial activity in a sandy arable soil is governed by the fertilization regime. *European Journal of Soil Biology*, 40, 87-94.
- Tripathi, K.M., Dhakal, D.D., Sah, S.C., Baral, D.R. and Sharma, M.D. (2015). Evaluation of vermicompost and chemical fertilizer on performance of pak choi (*Brassica rapa* CV. *Hong Tae*) and soil biological process. *Journal of Institute Agriculture Animal Science*, 33-34: 243 - 250.
- Umesha, S., Srikantiah, M., Prasanna, K.S., Sreeramulu, K.R., Divya, M. and R.N. Lakshmi pathi. (2014). Comparative effect of oand biofertilizer in growth and yield of maize (*Zea mays*.L). *Current Agriculture Research*, 56-62.
- Ummar, G. and Mohee, R. (2008). Assessing the effect of biodegradable and degradable plastics on the composting of green wastes and compost quality. *Bioresource Technology*, 99 (15): 6738-6744.
- Vendreme, A.W., Maguire, K.K. and Moore K.K. (2005). Growth of selected bedding plants are effected by different compost percentages. *Florida State Horticulture Science*, 18:368-371.
- Wang, J.P., Chen, Y.Z., Ge, X.W. and Yu H.Q. (2007). Optimization of coagulation-flocculation process for a paper recycling wastewater treatment using response surface methodology. *Colloid Surfaces A: Physicochemical and Engineering Aspects*, 302: 204-210.
- Wu, Y., S.W. Cui, J. Tang and X. Gu. (2007). Optimization of extraction process of crude polysaccharides from boat-fruited sterculia seeds by responsesurface methodology. *Food Chemistry*, 105: 1599-1605.

- Yosefi, K., Galavi, M., Ramrodi, M. and Mousavi S.R. (2011). Effect of bio-phosphate and chemical phosphorus fertilizer accompanied with micronutrient foliar application on growth, yield and yield components of maize (Single cross 704). *Australian Journal of Crop Science*, 5 (2): 175-180.
- Yousaf, M.M., M. Zeshan, M. Hussain, M.M. Raza, M.J. Shah, B. Ahmed and S.H. Shah. (2018). Effect of source and placement timing of nitrogen fertilizers on growth and yield of raya (*Brassica juncea L.*). *Pakistan Journal of Agriculture Research*, 31(3): 285-290.
- Zhang, X., Davidson, E.A., Mauzerall, D.L., Searchinger, T.D., Dumas, P. and Shen Y. (2015). Managing nitrogen for sustainable development. *Nature*, 528 (7580): 51-59.
- Zhou, L.X., and Ding, M.M. (2007). Soil microbial characteristics as bio-indicators of soil health. *Biodiversity Science*, 15, 162-171.

PUBLICATION

Submitted

Renly, A., Abdul Aziz, M. S., Chin, N. L., and Tee, T. P. (2018). RSM Optimization of Cattle Liquid Bio Fertilizer Application and Growth Media on Pak Choi (*Brassica chinensis*) Growth Performance.

