# การเพิ่มคุณค่าทางโภชนะของกากถั่วเหลืองโดยใช้ยีสต์หมักร่วมกับ แลคติกแอซิดแบคทีเรียในน้ำนมดิบ

# The improving of soybean meal nutritive value by using yeast and lactic acid bacteria in raw milk

สินีนาฏ พลโยราช<sup>1\*</sup>, อรอนงค์ พวงชมภู<sup>1</sup>, เมธา วรรณพัฒน์<sup>2</sup>, อัญชณา สุภาชาติ<sup>1</sup>, อรอุมา ไสย์ฮาตวงษ์<sup>1</sup> และ จีราวุฒ ไชยยายงค์<sup>1</sup>

Sineenart Polyorach<sup>1\*</sup>, Onanong Poungchompu<sup>1</sup>, Metha Wanapat<sup>2</sup>, Unchana Supachat<sup>1\*</sup>, Onuma Saihatwong<sup>1</sup> and Geerawut chaiyayong<sup>1</sup>

บทคัดย่อ: งานทดลองครั้งนี้ เพื่อศึกษาถึงการเพิ่มคุณค่าทางโภชนะของกากถั่วเหลืองโดยใช้น้ำนมหมัก งานทดลอง ครั้งนี้ศึกษาโดยใช้แผนการทดลองแบบ t-test เพื่อศึกษาเปรียบเทียบองค์ประกอบทางเคมีระหว่าง กากถั่วเหลืองไม่หมัก นมและกากถั่วเหลืองหมักนม โดยนำผลิตภัณฑ์อาหารที่ได้ในงานทดลองครั้งนี้มาวิเคราะห์หาค่า DM, OM, CP, EE, CF, NDF และ ADF จากผลการทดลองพบว่าในกลุ่มที่หมักนมสามารถเพิ่ม - CP และ EE โดยเฉพาะอย่างยิ่งค่าของ CP เพิ่ม ขึ้นจาก 48.6 เป็น 70.6 %DM ในขณะที่ ADF ลดลงอย่างมีนัยสำคัญทางสถิติ (P<0.05) มากไปกว่านั้นทั้ง CF และ NDF ก็มีค่าลดลงเช่นกันเมื่อเปรียบเทียบกับกลุ่มที่ไม่หมัก จากผลการทดลองครั้งนี้สามารถสรุปได้ว่า กากถั่วเหลืองหมักนม สามารถเพิ่มคุณค่าทางโภชนะของกากถั่วเหลืองได้โดยการเพิ่ม CP และ EE และการลดลงของ ADF มากไปกว่านั้น งาน วิจัยในอนาคตควรมีการศึกษาเพิ่มเติมถึงองค์ประกอบของ ในโตรเจนที่ไม่ใช่โปรตีน โปรตีนแท้ กรดอะมิโนและกรดไขมัน ในกากถั่วเหลืองหมักนม

**คำสำคัญ**: กากถั่วเหลือง, น้ำหมักนม, ยีสต์, แลคติกแอซิดแบคทีวีย, แหล่งโปรตีน

**ABSTRACT:** This experiment was conducted the to improve soybean meal nutritive value by using yeast and lactic acid bacteria in raw milk. Experiments were used t-test to compare chemical composition between unfermented milk soybean meal and fermented milk soybean meal. Experimental products were analyzed for DM, OM, CP, EE, CF, NDF and ADF. The result was shown that in fermented milk group significantly (P<0.05) increased CP and EE, especially, CP was increased from 46.8 up to 70.6 % of DM while ADF was significantly (P<0.05) decreased. In addition, both, CF and NDF were numerically decreased when compared with unfermented group. In conclusion, fermented milk could improve nutritive value of soybean meal by increasing crude protein and ether extract and decreasing acid detergent fiber. Moreover, the further research should be investigate more composition in term of non protein nitrogen, true protein, amino acid and fatty acid profile in soybean meal fermented milk. **Keywords:** Soybean meal, fermented milk, yeast, lactic acid bacteria, protein source

ชาขาวิชาสัตวศาสตร์ คณะทรัพยากรธรรมชาติ มหาวิทยาลัยราชมงคลอีสาน วิทยาเขตสกลนคร Department of Animal Science, Faculty of Natural Resources, Rajamangala University of Technology-Isan, Sakon Nakhon Campus, Phang Khon, Sakon Nakhon, 47160, Thailand

<sup>&</sup>lt;sup>2</sup> ศูนย์วิจัยและพัฒนาทรัพยากรอาหารสัตว์เขตร้อน ภาควิชาสัตวศาสตร์ คณะเกษตรศาสตร์ มหาวิทยาลัยขอนแก่น Tropical Feed Resources Research and Development Center, Department of Animal Science, Faculty of Agriculture, Khon Kaen University, 40002. Thailand

#### Introduction

The significant increase in demand livestock product in recent years in developing countries has required increasing and improving the quality of animal feed supply. Many researchers try to find strategies to improve nutritive value of byproduct and local feed such as cassava chip, rice straw, rice bran, soybean meal, etc. Incorporation of microbial additives such as a culture of *Saccharomyces cerevisiae* and lactic acid bacteria to the diet has become a common practice in ruminant nutrition.

Fermented milk products are regarded as predominantly lactic fermentations, the frequent occurrence of yeasts and lactic acid bacteria has led to the suggestion that interactions may occur that can influence product characteristics and quality. Certainly, the presence of yeasts is crucial for the desirable properties of carbon dioxide and ethanol production in east European and Asian products such as kefir, koumiss and airag. However, the mechanisms of possible interaction between yeasts and the lactic flora have not been widely studied. Such interaction may be stimulation or inhibition of growth of one, or both, of the co-cultured strains (Narvhus and Gadaga, 2003).

Soybean meal is a protein source in animal feed worldwide. Its universal acceptability in animal feed has been due to favourable attributes such as relatively high protein content and suitable amino acid profile except methionine, minimal variation in nutrient content, ready availability year-round, and relative freedom from intractable anti-nutritive factors if properly processed. Also, attention has been focused on soy-

bean meal utilization as an alternate protein source in animal diets due to the changing availability or allowed uses of animal proteins. However, the improving quality of feedstuff is very interesting by nutritionist and also the data of method to improve soybean meal quality by using microorganism fermented milk still limited. Therefore, the objective in this experiment is to study the improving of soybean meal nutritive value by using fermented milk.

#### Materials and methods

#### Fermented milk process

Activated yeast: weight Baker's yeast 20g into a flask mixed with sugar 20 g and distill water 100 ml. then incubated at room temperature for 1 h (A). Activated Lactic acid bacteria: mixed commercial yoghurt 50 g, molasses 25 g and distilled water 25 ml,- incubated at room temperature for 2 h (B). Mix (A) and (B) with 500 g of raw milk (pH 5-7) then flushed with air for 4 days at room temperature by using air pump (600 W).

#### Soybean meal fermented milk production

After fermented milk for 4 days, mixed fermenting milk with soybean meal in tank at proportion 1.5: 1 kg, fermented for 3 days. The pellet were did using soybean meal fermented milk 2 kg with cassava chip mashed 1 kg and then sundry for 2 days. Products were used as a feed-source for animal. Soybean product: prepared by mixed soybean meal with water in tank at proportion 1.5: 1 kg and then mixed with cassava chip mashed at proportion 2:1, after that sundry for 2 days.

## Chemical analysis

Soybean meal and soybean meal fermented milk product were analyzed for Dry matter (DM), organic matter (OM), ether extract (EE), crude fiber (CF) and crude protein (CP) determined according to AOAC (1990). The sample was also analyzed for neutral detergent fiber (NDF) and acid-detergent fiber (ADF) according to Van Soest et al. (1991).

#### Analysis of data

Data were analyzed by using the t-test (SAS, 1998) to compare the chemical composition of the two treatments (soybean meal and soy bean meal fermented milk). Each treatment was containing 5 replication. Treatment means were statistically compared using Duncan's New Multiple Range Test (Steel and Torrie, 1980). Differences were assumed to be significant at P<0.05.

# Results and discussion

Table 1, showed effect of soybean meal fermented milk on chemical composition of experimental feed. It was found that when fermented soybean meal with yeast and lactic acid bacterial fermented milk could significantly (P<0.05) increased crude protein and ether extract, especially, crude protein the value increased from 46.8 up to 70.6 % of dry matter while acid detergent fiber was significantly (P<0.05) decreased. However, both, crude fiber and neutral detergent fiber were numerically decreased as well.

The increasing of crude protein ether extract that might be due to effect of soybean meal, cassava chip mashed, raw milk, micro-organism

(yeast and lactic acid bacteria) in soybean meal fermented milk could utilized milk and soybean meal to product them self and also products from fermented milk. Nasseri et al. (2011) reported that yeast contain protein, fat, ash and nucleic acid around 45-55, 2-6, 5-10 and 6-12 % dry weight, respectively. Moreover, Yamada and Sgarbieri, (2005) study compare the composition and functional and nutritional properties of whole yeast cells (WY) an ethanol distillery with those of a phosphorylated protein concentrate (PPC) prepared from the same cells. It was found that yeast (S. cerevisiae) is rich source of protein, soluble fiber, some minerals, and saturated fatty acids predominated over monounsaturated and polyunsaturated in both WY and PPC. Moreover, fermented soybean meal also could be used as an energy source for animals because it contain cassava chip mashed at 33.3% in fermented soybean meal. Many recherchers interested in using microorganism to improved nutritive value of feed. Thongkratok et al. (2010) reported that microbial fermentation played a significant role in nutritional enrichment of erstwhile worthless and often discarded agro-industrial by-products generated through the harvesting and processing of cassava roots. Recently, Wanapat et al. (2011) and Polyorach et al. (2012, 2013, 2014) reported that yeast could prepare to increased crude protein content of cassava chip up to 30-47% of dry matter.

Most of the important microorganisms applied in the production of fermented foods have been studied for decades, yielding a wealth of information on their physiology and genetics in relation to product functionalities, such as the development of flavor, taste, texture and chemical

KHON KAEN AGR. J. 42 SUPPL. 1: (2014).

composition of feedstuff. The quantity as well as quality of the food proteins as expressed by biological value, and often the content of water soluble vitamins is generally increased, while the antinutritional factors show a decline during fermentation (Paredes-López and Harry, 1988). Fermentation results in a lower proportion of dry matter in the food and the concentrations of vitamins, minerals and protein appear to increase when measured on a dry weight basis (Adams, 1990). Ws-

zolec et al. (2001) reported that fermented milk product which utilizes a wide variety of microorganisms to produce a wide variety of products in addition to lactic acid including ethanol, free fatty acids, and acetaldehyde. Inaddition, Mixed culture fermentation of pearl millet flour with *S. diastaticus, S. cerevisiae, Lactobacillus brevis* and *L.fermentum* was found to improve its biological utilisation in rats (Khetarpaul and Chauhan, 1991).

Table 1. Effect of fermented milk on chemical composition of experimental feed

Item	Unfermented soybean meal	Fermented soybean meal	SEM	P-value
Dry matter	97.4	96.0	2.29	0.3599
Organic matter	93.3	95.2	1.52	0.0839
Crude protein	46.8 <sup>a</sup>	70.6 <sup>b</sup>	1.68	<.0001
Ether extract	3.2°	5.6 <sup>b</sup>	0.60	0.0002
Crude fiber	6.6	5.8	0.66	0.0801
Neutral detergent fiber	12.8	10.6	1.89	0.1026
Acid detergent fiber	10.2 <sup>a</sup>	6.8 <sup>b</sup>	1.61	0.0102

<sup>&</sup>lt;sup>a,b</sup> Value on the same row with different superscripts differ (P<0.05), SEM= standard error of the means

#### Conclusions

In conclusion, - fermented milk could improve nutritive value of soybean meal by increasing crude protein and ether extract and decreasing acid detergent fiber. Moreover, the further research should be investigate more composition in term of non protein nitrogen, true protein, amino acid and fatty acid profile in soybean meal fermented milk.

### Acknowledgements

The authors would like to express their most sincere thanks to all who have assisted and supported the research in this study, particularly Department of Animal Science, Faculty of Natural Resources, Rajamangala University of Technology-Isan, Sakon Nakhon Campus, Phang Khon, Sakon Nakhon, Thailand and Tropical Feed Resources Research and Development Center, Department of Animal Science, Faculty of Agriculture, Khon Kaen University, 40002, Thailand for providing financial support of research and the use of research facilities.

#### References

Adams, M. J. 1990. Beginning to Read: Thinking and Learning about Print. Cambridge, MA: Bolt, Beranek, and Newman, Inc. ED 317 950.

- AOAC. 1990. Official methods of analyses, 15th edn. Assoc. Offic. Anal. Chem, Arlington, VA.
- Khetarpaul, N., and B.M. Chauhan. 1991. Biological utilisation of pearl millet flour fermented with yeasts and lactobacilli. Plant Foods for Human Nutrition. 41: 309-319.
- Narvhus, J. A., and T. H. Gadaga. 2003. The role of interaction between yeasts and lactic acid bacteria in African fermented milks: a review. Int. J. Food Microbiol. 86: 51–60.
- Nasseri, A.T., S. Rasoul-Amini, M.H. Morowvat, and Y. Ghasemi. 2011. Single cell protein: Production and process. Am. J. Food Technol. 6: 103-116.
- Paredes-López, O., and G. I. Harry. 1988. Food biotechnology review: traditional solid-state fermentations of plant raw materials - application, nutritional significance and future prospects. CRC Critical Reviews in Food Science and Nutrition. 27: 159-187.
- Polyorach, P., M. Wanapat, and S. Wanapat. 2012. Increasing protein content of cassava (Manihot esculenta, Crantz) using yeast in fermentation. KHON KAEN AGR. J. 40 (supplement 2): 178-182.
- Polyorach, S., M. Wanapat, and S. Wanapat. 2013. Enrichment of protein content in cassava (*Manihot esculenta* Crantz) by supplementing with yeast for use as animal feed. Emir. J. Food Agric. 25 (2): 142-149.

- Polyorach, S., M. Wanapat, and A. Cherdthong. 2014. Influence of Yeast Fermented Cassava Chip Protein (YEFECAP) and Roughage to Concentrate Ratio on Ruminal Fermentation and Microorganisms Using *In vitro* Gas Production Technique. Asian Australas. J. Anim. Sci. 27(1): 36-45.
- SAS. 1998. User's Guide: Statistic, Version 6.12<sup>th</sup> Edition. SAS Inst. Inc., Cary, NC.
- Steel, R. G. D., and J. H. Torrie. 1980. Principles and Procedures of Statistics. McGraw Hill Book Co, New York.
- Thongkratok, R., S. Khempaka, and W. Molee. 2010. Protein enrichment of cassava pulp using microorganisms fermentation techniques for use as an alternative animal feedstuff. J. Anim. Vet. Adv. 9:2859-2862.
- Van Soest, P. J., J. B. Robertson, and B. A. Lewis. 1991. Methods for dietary fiber neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. J. Dairy Sci. 74: 3583–3597.
- Wanapat, M., S. Polyorach, V. Chanthakhoun, and N. Sornsongnern. 2011. Yeast-fermented cassava chip protein (YEFECAP) concentrate for lactating dairy cows fed on urea-lime treated rice straw. Livest. Sci. 139: 258-263.
- Yamada, E. A., and V. C. Sgarbieri. 2005. Yeast (Saccharomyces cerevisiae) protein concentrate: Preparation, chemical composition, and nutritional and functional properties. J. Agric. Food Chem. 53:3931-3936.