

PROBIOTIC CANDIDATES YEAST ISOLATED FROM *DANGKE*–INDONESIAN TRADITIONAL FERMENTED BUFFALO MILK

Gemilang Lara Utama¹, Siska Meliana¹,
Mohamad Djali¹, Tri Yuliana¹, Roostita L. Balia²

¹Faculty of Agro-Industrial Technology, Universitas Padjadjaran, Bandung, 45363, Indonesia

²Faculty of Animal Husbandry, Universitas Padjadjaran, Bandung, 45363, Indonesia

To link to this article: <https://doi.org/10.11118/actaun201967010179>

Received: 17. 8. 2018, Accepted: 22. 11. 2018

To cite this article: UTAMA GEMILANG LARA, MELIANA SISKI, DJALI MOHAMAD, YULIANA TRI, BALIA ROOSTITA L. 2019. Probiotic Candidates Yeast Isolated from *Dangke*–Indonesian Traditional Fermented Buffalo Milk. *Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis*, 67(1): 179–187.

Abstract

The aim of the study was to isolate and identify the yeast isolated from *Dangke* and its potential as probiotics. The purified isolates obtained were identified based on observations of macroscopic characteristics of colonies and microscopic cells. The ability as a probiotic yeast is obtained by testing the resistance towards acid conditions, bile salt resistance test and aggregation ability test against pathogenic bacteria using *Salmonella* sp.. The yeast isolates were identified using the RapID Yeast Plus System. The isolation result was obtained D.10.3.d isolate that identified as *Candida guilliermondii* which showed probiotic characteristic. The yeast colony is round, cream-colored, smooth surfaces, low convex elevations and entire edges, capable of growing on mediums with the pH of 4, containing 1 % and 5 % of bile salts and having the ability to aggregate *Salmonella* sp. at 15, 60, and 180 minutes.

Keywords: *Dangke*, yeast, identification, isolation, probiotics

INTRODUCTION

Dangke is a traditional fermented food that have similar tastes with cheese, but the appearance and texture are similar with white to yellowish tofu. As traditional fermented food, *Dangke* is an indigenous product for the Enrekang Regency that has been known by all the people of South Sulawesi and even Indonesia (Rahman, 2014).

Recently, the potential of *Dangke* has been developing into one of the functional food products. One of functional food product that is widely evolved

is probiotic products that control about 60–70 % of functional food markets (Holzapfel, 2005).

Various microorganisms such as bacteria or even yeasts can be used as probiotics (Khotimah *et al.*, 2015). Yeasts can be applied as a probiotic because it can survive in guts microflora and producing antibacterial metabolites (Vine *et al.*, 2006, Indah *et al.*, 2015).

The type of yeast used as a probiotic is very diverse. Several previous studies conducted in an in vitro and in vivo showed probiotic yeasts such *Saccharomyces boulardii* was able to prevent intestinal infections

caused by *Escherichia coli*, *Salmonella typhimurium*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Proteus vulgaris*, *Yersinia enterocolitica* and *Candida albicans* (Czerucka, *et al.*, 2007).

The nature of yeast as probiotics in dairy products has not been explored. Based on the above description, traditional fermented products such as *Dangke* can have potential as probiotic food obtained from yeast activities. So that the exploration of yeast probiotics from *Dangke* felt necessary to do in-depth research.

MATERIALS AND METHODS

The raw materials used in this study are *Dangke* obtained from Makassar, medium PDA/Potato Dextrose Agar (Oxoid Ltd.), NB/Nutrient Broth (Oxoid Ltd.), Brain Heart Infusion / BHI broth (Oxoid Ltd.), Vegemite (Kraft), Amoxicillin, 0.1 N HCl, synthetic bile salts (Ox Bite). The research method used is an experimental method with descriptive analysis. The yeast isolation was done by isolating the potentially probiotic yeast candidate from *Dangke* product. The parameters were tested based on yeast ability on resistance towards low pH, bile salts, and the aggregation ability.

Isolation and identification of yeast

According to Roostita and Fleet (1999), yeast isolation can be obtained by taking 1 gram of *Dangke* and conducting a series of dilutions to be inoculated onto agar media. Media used for yeast growth is PDA which modified by adding 3 % vegemite as an extract of yeast and 10 ppm amoxicillin to prevent unwanted bacterial growth. Identification of yeast isolates done by using *RapID Yeast Plus System* and continued with the analysis using ERIC (Electronic Code Compendium) through www.remel.com/eric (Utama *et al.*, 2016).

Resistance Test towards Acid Conditions

The acid resistance test was performed using a modified PDA medium added with 0.1 N HCl to obtain pH 2, 3, 4 (corresponding to gastric pH). A total of 1 ml of each yeast isolate extracted from culture stock then inoculated on a modified-HCl PDA medium. Incubated for 48 hours at 37 °C. The yeast colony appears on PDA modified-HCl medium counted as acid resistance yeast probiotic candidates (Djide and Wahyuddin, 2008).


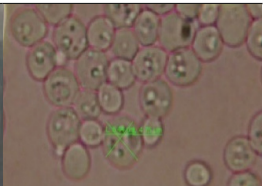
Resistance test towards bile salts

Modified PDA medium is added with synthetic bile salts, with concentrations of 1 % and 5 %. As many as 1 loops of yeast isolate taken from the culture stock then streaked on the bile salt-modified PDA medium. Incubation was done for 48 hours at 37 °C (Djide and Wahyuddin, 2008; Khotimah *et al.*, 2015). The results were obtained qualitatively from the comparison of the yeast colonies appears from control (not streaked PDA) with the streaked modified PDA (1 % and 5 % bile salts).

Salmonella sp. Aggregation Test

One ml of 10⁸ CFU/ml yeast isolates were grown in modified NB medium at 37 °C for 24 hours, with constant agitation (150 rpm). 0.5 ml of *Salmonella* sp. suspension contains 10⁹ CFU/ml was grown in BHI broth for 48 hours at 37 °C. Mix the two suspensions on six-well plates, the presence or absence of aggregation is observed microscopically after the 15th and 60th minutes, up to 180 minutes' maximum (Perez-Sotelo *et al.*, 2005; Khotimah *et al.*, 2015).

I: *Macroscopic and Microscopic Appearance of Yeast Isolated from Dangke*

Macroscopic Characteristic					Figure	
Form	Colour	Surface	Elevation	Edge	Macroscopic	Microscopic 1000× magnification
Round	Cream	Smooth	Low convex	Entire		

RESULTS AND DISCUSSION

Yeast Isolation

Results of microorganism isolation from *Dangke* found 4 bacteria isolates and 1 yeast-like isolate. The yeast-like isolates were identified macroscopically and microscopically, which the results can be seen in Tab. I.

Yeast-like isolate has an appearance with features of rounded colony shape, slightly creamy white color, prominent elevation, glossy surface, flat edges, oval cell shape, and has a size of 3.8 μm . According to Bhatia (2016) and Utama, *et al.* (2015), yeast has a round shape, cream-colored, smooth surface, soft texture and has a varied size, usually has a diameter of 3–4 μm .

Yeast Identification using RapID Yeast Plus System

The results of the biochemical test used for yeast identification is using RapID Yeast Plus System (Tab. II). Based on the results of the biochemical test obtained, yeast-like isolate showed color changes in the media of glucose, sucrose, and raffinose, the color changed from red to yellow. According to Bhowmik, (2011), the color changes due to yeast activity ferment glucose, sucrose, and raffinose,

so its produce acid and cause color changes in the media from red to yellow. If the carbohydrate particles are fermented, the final product produced is acidic, thus decreasing the pH so that the pH indicator will change (phenol red changes from red to yellow).

Positive test of ONPG will also produce a yellow color because according to Leboffe and Pierce (2011), β -galactosidase will break down o-Nitrophenyl- β D-galactopyranose (ONPG) to yellow β -galactose and o-Nitrophenol (ONP). Positive results on enzyme tests showed that these isolates were able to degrade complex compounds to be simpler. Urease test results showed negative results, this was indicated because there was no color change in the urea test media. Urease test is useful to identify organisms that are able to hydrolyze urea which can produce ammonia and carbon dioxide, especially to find out whether these microorganisms have urease enzymes or not. Urease is a constitutive enzyme that hydrolyzes urea to carbon dioxide and ammonia (NH_2) $2\text{CO} + \text{H}_2\text{O} \rightarrow \text{CO}_2 + 2\text{NH}_3$ (Brink, 2013).

Based on the biochemical test and after analyzed by ERIC, the isolates were identified similar to *Candida guilliermondii* species. *Candida guilliermondii* is a commensal yeast that can live as a saprophyte in the respiratory and

II: Biochemical Test Results using RapID Yeast Plus System

Material	Results	%
GLU	+	91 %
MAL	–	00 %
SUC	+	92 %
TRE	–	04 %
RAF	+	38 %
LIP	–	03 %
NAGA	–	00 %
α GLU	+	63 %
β GLU	–	40 %
ONPG	–	00 %
α GAL	+	70 %
β FUC	–	01 %
PHS	+	91 %
PCHO	+	88 %
URE	–	07 %
PRO	+	99 %
HIST	+	70 %
LGY	+	29 %

gastrointestinal tract. Changes from the form of saprophytes to pathogens, will not occur as long as the body's immunity is still stable, and there are no predisposing factors. This factor can increase yeast growth and facilitate yeast invasion into the system so that local colonization arises (Yang *et al.*, 2010).

Candida's presence in food products is a common thing. *Candida* has a beneficial composition so that it has a high potential for use in various industrial sectors. According to Nehal (2013), *Candida* has been found in many fermentation products in Asian countries especially in India and Nepal. *Candida* parapsilosis also found in soybean fermentation, *C. famata*, *C. guilliermondii*, *C. kefir*, and *C. inconspicua* on milk fermentation from kefir (Simova *et al.*, 2002; Sohliya *et al.*, 2009; Pogacic *et al.*, 2013).

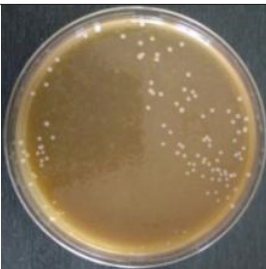
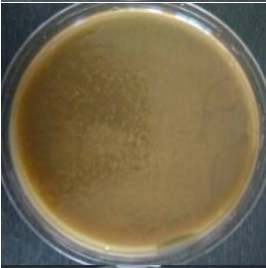
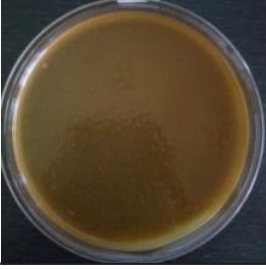
Probiotic yeast candidates resistance towards acids

According to Kanmani *et al.* (2010), resistance towards acid is one of the important conditions of an isolate to be categorized as probiotics. This is because if the isolate enters the digestive tract of the body, the isolate must be able to withstand the pH of the gastric acid which is around 3–5. The results of the resistance test of *C. guilliermondii* isolates for various pH values are presented in Tab. III.

Tabl. III shows that the *Candida guilliermondii* isolated from *Dangke* were able to grow at pH 4 with the number of colonies around 9.9×10^7 CFU/g while at pH 2–3 showed no growth. This is in line with the opinion of Shah, (2007) and Hardiningsing *et al.*, (2006), that the minimum number of probiotic strains present in probiotic foods is 10^6 CFU/g. The number of probiotic strains that must be consumed every day around 10^8 CFU/g and resistant to acidic conditions in the pH range 2–6, with the aim of compensating for the decreasing possibility to the number of probiotic microbes in the digestive tract.

Based on the testing results of low pH indicates that *Candida guilliermondii* isolates are able to survive acidic conditions at pH 4. Susanti *et al.* (2007) explained that very acidic conditions can cause membrane damage and release of intracellular components that can cause death. Acid resistant yeast has a greater resistance to membrane damage due to a decrease in extracellular pH. The high tolerance of yeasts to acids is usually due to the fact that the yeast is able to maintain a more alkaline pH of the cytoplasm than the extracellular pH. A more alkaline cytoplasmic pH defenses occur

III: The resistance of *Candida guilliermondii* towards acid conditions

pH	Number of Colony	Picture
4	9.9×10^7	
3	0	
2	0	

when cells have membranes which are barriers that limit the movement of compounds/protons. According to Usmiati, *et al.* (2011), microorganisms that are not resistant to acidic conditions are unable to maintain pH in their cells to become more alkalis than their extracellular pH, resulting in the cell membrane and the microbial intracellular component damage that causes death.

Composite components such as fatty acids and proteins among yeast species also affect the ability to resist acidic conditions (Halim and Zubaidah, 2013). The yeast cell wall is an elastic structure that provides physical protection and osmotic support and determines cell shape. This cell wall is a complex and dynamic structure containing glucans, chitin, and mannoproteins (Orlean, 2012).

The components of the *Candida* cell wall consist of β -glucan, chitin, mannoprotein, and glucomannan (Drabikova *et al.*, 2009). β -glucan is the main component which covers about 50–60 % of the inner layer of the cell wall, while chitin covers 1–10 %. The outer layer of the cell wall consists of mannoproteins which represent 30–40 % of the total polysaccharide of the cell

wall. Mannoproteins are involved in cell-to-cell recognition, determine the nature of cell surfaces and play an important role in interactions with the host (Vickova *et al.*, 2004). The components of the yeast cell wall such as mannoprotein affect the ability of *Candida* to resist acidic conditions. Another component of the cell wall is glucomannan, the antioxidant properties of glucomannan can protect the system against harmful activities associated with free radicals, iron ion chelation, and intracellular modulation of signaling pathways (Miadokova *et al.*, 2006).

Probiotic candidates yeast candidates resistance to bile salts


Salminen *et al.* (2004) explained that microorganism can be categorized as probiotic if it is able to survive and grow in the digestive tract, especially when it enters the upper intestinal tract, where bile salts are secreted in the intestine. The period of observing *Dangke* indigenous yeast resistance against bile salts was carried out for 48 hours. According to Surono (2004), the length of microorganisms living in the intestine is about 4–6 hours. However, yeast that has passed bile salts must be able to colonize the lower intestinal tract so that can be defined as probiotic yeast, and so the observation time carried out in this study was extended to 48 hours. The ability of *Candida guilliermondii* to survive in bile salts for 48 hours with a concentration of 1 % and 5 % can be seen in Tab. IV.

The results showed that the isolates succeeded in living on a medium which added 1 % and 5 % bile salts, based on these results, *Candida guilliermondii* was resistant against bile salts. 1 % concentration of bile salts is the lowest concentration and 5 % is a critical concentration, a value that is high enough to select isolates resistant to bile salts (Pacheco *et al.*, 2010).

Bile salts are a mixture of bile acids, cholesterol, fatty acids, phospholipids, bile pigments, and a number of detoxified xenobiotics. The combination is bactericidal for commensal microorganisms in humans so that a number of pathogenic microbes do not hold bile salts, except for some genera of intestinal inhabitants or out of it that have resistance to bile salts (Salen and Batta, 2004; Bijl *et al.*, 2009).

Bile salt compounds will be toxic to living cells, so microorganisms in the digestive tract must have a defense mechanism to protect themselves from that toxic activity (Puspawati and Arihantana, 2016). A number of yeasts can

IV: Results of *Candida guilliermondii* Resistance Observation of Bile Salts

Treatment	Result
Control	
Bile Salt 1 %	
Bile Salt 5 %	

survive against bile salts due to the presence of fatty acids which can reduce cell leakage caused by bile salts. The presence of fatty acids is to improve the stability of membrane lipids. Lipids are the main compounds found in gram-positive microbial cell membranes and play an important role in maintaining the membrane structure (Kimoto *et al.*, 2002). The presence of other cell walls components such as polysaccharides can tolerate the presence of bile salts to prevent cell leakage (Puspawati and Arihantana, 2016). In addition, the ability to deconjugate bile salts is also thought to be a factor that allows an isolate to have resistance against bile salts. The deconjugation process can reduce the level of toxicity from conjugated bile salts to another microbe (De Smet *et al.*, 1995).

Probiotic yeast candidates aggregation towards *Salmonella* sp.

One of many features of yeast that must be possessed as a candidate for probiotics is to have the ability to interact and stick among species, so as to prevent pathogenic bacteria from attaching to

the intestine. These conditions can improve health by improving the balance of intestinal micro-flora. Aggregation ability is an easy and reliable way to select various probiotic yeasts. Adhesion of pathogenic bacteria panel to the surface of various yeasts is determined as aggregation (Khotimah *et al.*, 2015). The results of *Candida guilliermondii* aggregation on *Salmonella sp.* can be seen in Tab. 5.




Based on the appearance results in the microscope, it can be seen that *Candida guilliermondii* has an aggregation ability towards *Salmonella sp.*. The aggregation ability of *Candida guilliermondii* was shown at intervals of 15, 60 and 180 minutes. The attachment of *Salmonella sp.* aggregation is influenced by the presence of structural virulence factors possessed by bacteria such as capsule, surface protein and hemagglutinin (Wahyuni *et al.*, 2006). Bacteria that have hemagglutinin can be more easily attached to the mucosal surface. Hemagglutinin will initiate colonization by mediating bacteria to receptors binding (usually oligosaccharides) in human cells because this bacterium requires hemin for growth,

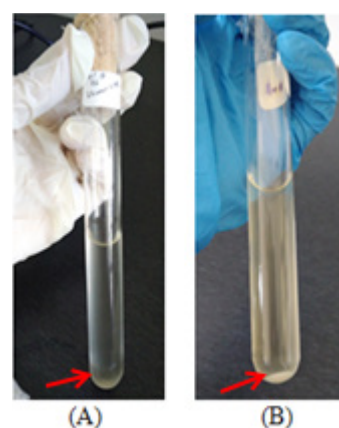
so its bond with the cell body's erythrocytes also provides nutritional functions (Irianto, 2008).

The results show *Candida guilliermondii* is potential to be used as a probiotic. According to Annuk *et al.*, (2001) and Leboffe and Pierce, (2011), some microorganism can maintain their ability and autoagglutination properties because they have an S layer that binds tightly to the cell surface. Some researchers have suggested the role of carbohydrates such as teichoic acids, lipoteichoic acids and polysaccharides to the aggressiveness of *Salmonella* from yeast (Annuk *et al.*, 2001; Corcoran *et al.*, 2005). Some researchers also state that adherence in the mucosal epithelium is mediated by lectins (Jin *et al.*, 2001; Rickard *et al.*, 2000). Lectin is a non-immunologic protein or glycoprotein that can be involved in adhesion phenomena with specific molecules (carbohydrates, glycoproteins or glycolipids). The multimeric lectin structure causes the lectin to have the ability for agglutinate cells or form precipitates with glycoconjugates in similar ways as antigen-antibody reactions. The aggregation ability of isolates in a single form (aggregation) is influenced by the production of EPS (Extra Cellular Polysaccharides) in the outer membrane of the cell, in addition to gravity and motility or bacteria movement when EPS production is low (Surono, 2004).

Beside microscopically, aggregation ability of *Candida guilliermondii* towards *Salmonella sp.* can qualitatively be supported by observing the presence of sediments in the broth which are incubated for 48 hours. The aggregation ability is asserted positively when the incubated culture

V: Aggregation ability towards *Salmonella sp.*

Time (minutes)	The appearance of aggregation ability on a microscope
15	
60	
180	



1: Results of a qualitative aggregation test of *C. guilliermondii* and *Salmonella sp.*

Description: A = Suspension of *C. guilliermondii* and *Salmonella sp.* before Incubation

B = Suspension of *C. guilliermondii* and *Salmonella sp.* After Incubation

looks clear with the aggregate of cells precipitate at the base of the tube. Aggregation is considered negative (non-aggregating) if the incubated culture exhibits significant turbidity. The result of sedimentation at *Candida guilliermondii* during the incubation period can be seen in Fig. 1.

Based on the precipitate, the results showed that the aggregate formed at the base of the liquid

culture was an indication that the isolate had aggregation ability as one of the preconditions of a yeast isolate before being determined as probiotics. The aggregation ability itself will cause the probiotic to be able to colonize the mucosal epithelium by forming bacterial films that play a role in removing pathogenic bacteria from the intestinal mucosa (Khotimah *et al.*, 2015).

CONCLUSIONS

Yeast isolated type from Dangke is *Candida guilliermondii* species with the morphology of colonies showed characteristics have a round shape, cream color, smooth surface, elevation of low convex and entire edge. Based on the results of acidic, bile salts and aggregation ability against *Salmonella sp.*, *Candida guilliermondii* were able to resist pH 4, against bile salts with concentrations of 1 % and 5 % and had the ability to aggregate *Salmonella sp.* which increased until minutes 15, 60 and 180.

Acknowledgement

Author would like to thank Rector of Universitas Padjadjaran for the Academic Leadership Grant of Hibah Internal Unpad. Also thanked the Student Research Group, Vivi Fadila Sari, Isfari Dinika and Syarah Virgina who helped in the laboratory. This research / article's publication is supported by the United States Agency for International Development (USAID) through the Sustainable Higher Education Research Alliance (SHERA) Program for Universitas Indonesia's Scientific Modeling, Application, Research and Training for City-centered Innovation and Technology (SMART CITY) Project, Grant #AID-497-A1600004, Sub Grant #IIE-00000078-UI-1.

REFERENCES

- ANNUK, H., HYNES, S.O., HIRMO, S., MIKELSAAR, M. and WADSTROM, T. 2001. Characterization and differentiation of lactobacilli by lectin typing. *J. Med. Microbiol.*, 50(12): 1069–1074.
- BHATIA, S.C. 2016. *Food Biotechnology*. New Delhi: Woodhead Publishing India Ltd.
- BHOWMIK, G. 2011. *Analytical Techniques in Biotechnology*. New Delhi: Tata McGraw Hill Education Private Ltd.
- BIJL, N., A. VELDE, V. D. and GROEN, A. K. 2009. Bile Acids and Their Role in Cholesterol Homeostasis. In: EHNHOLM C. (Eds.). *Cellular Lipid Metabolism*. Berlin, Heidelberg: Springer.
- BRINK, B. 2013. *Urease test protocol*. [Online]. American society for microbiology. Available at: [http://www.microbelibrary.org/library/laboratory test/2871-urease-test protocol](http://www.microbelibrary.org/library/laboratory%20test/2871-urease-test%20protocol) [Accessed: 2018, August 21].
- CORCORAN, B. M., STANTON, C., FITZGERALD, G. F. and ROSS, R. P. 2005. Survival of probiotic in acidic environments is enhanced in the presence of metabolizable sugars. *Appl. Environ. Microbiol.*, 71(6): 3060–3067.
- CZERUCKA, D., PICHE, T. and RAMPAL, P. 2007. Review article: yeast as probiotics *Saccharomyces boulardii*. *Aliment. Pharmacol. Therap.*, 26(6): 767–778.
- DE SMET, I., HOORDE, L. V., WOESTYNE, M. V., CHRISTIAENS, H. and VERSTRAETE, W. 1995. The significance of bile salt hydrolytic activities of *lactobacilli*. *J. App. Bacteriol.*, 79(3): 292–301.
- DJIDE, M. N. and WAHYUDDIN, E. 2008. Isolasi Bakteri Asam Laktat dan Air Susu Ibu dan Potensinya dalam Penurunan Kadar Kolesterol secara In Vitro. *Majalah Farmasi dan Farmakologi*, 12(3): 73–78.
- DRABIKOVA, K., PERECKO, T., NOSAL, R., BAUEROVA, K., PONIST, S., MIHALOVA, D., KOGAN, G. and JANCINOVA, V. 2009. Glucmannan reduces neutrophil free radical production in vitro and in rats with adjuvant arthritis. *Pharmacol. Res.*, 59(6): 399–403.
- HALIM, C. N. and ZUBAIDAH, E. 2013. Probiotic Assay of Lactic Acid Bacteria as High Exopolysaccharides Producer from Sour Pickled Mustard (*Brassica juncea*). *Jurnal Pangan dan Agroindustri*, 1(1): 129–137.
- HARDININGSIH, R., NAPITUPULU, R. N. R. and YULINERY, T. 2006. Isolation and resistance test of several isolates of *Lactobacillus* in low pH. *Biodiversitas*, 7(1): 15–17.

- HOLZAPFEL, W. H. 2005. Introduction to Prebiotics and Probiotics. In: GOKTEPE, I., JUNEJA, V.K., and AHMEDNA, M. (Eds.). *Probiotics in Food Safety and Human Health*. CRC Taylor & Francis Group.
- INDAH, H., PUTRI, F. and UTAMA, G. L. 2015. Preliminary studies of halophilic yeasts antimicrobial activities isolated from cocoa bean pulp towards *E. coli* and *Salmonella spp.* *Int. J. on Adv. Sci. Eng. and Information Technology*, 5(2): 107–109.
- IRIANTO, A. 2008. *Efek Antibakteri Aloe Vera Terhadap Porphyromonas gingivalis In Vitro (Perbandingan Metode Ekstraksi Maserasi dan Infundasi)*. Jakarta: Fakultas Kedokteran Gigi Universitas Indonesia.
- JIN, Y. L., RITCEY, L. L. and SPEERS, R. A. 2001. Effect of cell surface hydrophobicity, charge and zymolectin density on the flocculation of *Saccharomyces cerevisiae*. *J. Am. Soc. Brew. Chem.*, 59(1): 1–9.
- KANMANI, P., KUMAR, R. S., YUVARAJ, N., PAARI, K. A., PATTUKUMAR, V. and ARUL, V. 2010. Comparison of antimicrobial activity of probiotic bacterium *Streptococcus phocae* P180, *Enterococcus faecium* MC13 and *Carnobacterium* divergence against the fish pathogen. *World Journal of Dairy and Food Sciences*, 5(2): 145–151.
- KHOTIMAH, K., BALIA, R., CHAERUNNISA, H., SAFITRI, R., UTAMA, G. L., SAPUTRA, M. I. and SYARA, A. 2015. Isolation and Identification Probiotic Candidates Bacteria from Crossbreed Fries Holland (CFH) Colostrum that Grown on Nutrient Agar with Olive Oil and Casein Enrichment. *Adv. J. of Food Sci. and Technol.*, 8(1): 22–26.
- KIMOTO, H., OHMOMO, S. and OKAMOTO, T. 2002. Enhancement of Bile Tolerance in *Lactococci* by Tween 80. *J. Of Applied Microbiology*, 92(1): 41–46.
- LEBOFFE, M. J. and PIERCE, B. E. 2011. *A Photographic Atlas for The Microbiology Laboratory*. 4th Edition. Morton Publishing Company.
- MIADOKOVA, E., SVIDOVA, S., VLCKOVA, V., DUHOVA, V., NADOVA, S. RAUKO, P. and KOGAN, G. 2006. Diverse biomodulatory effects of glucomannan from *Candida utilis*. *Toxicol. In Vitro*, 20(5): 649–657.
- NEHAL, N., 2013. Knowledge of traditional fermented food products harbored by the tribal folks of Indian Himalayan belt. *Int. J. Agric. Food Sci. Technol.*, 4(5): 401–414.
- ORLEAN, P. 2012. Architecture and biosynthesis of the *Saccharomyces cerevisiae* cell wall. *Genetics*, 192(3): 775–818.
- PACHECO, K. C., DEL TORO, G. V., MARTINEZ, F. R. and DURAN-PARAMO, E. 2010. Viability of *Lactobacillus delbrueckii* under human gastrointestinal conditions simulated in vitro. *Am. J. Agric. Biol. Sci.*, 5(1): 37–42.
- PEREZ-SOTELO, L. S., TALAVERA-ROJAS, M., MONROY-SALAZAR, H. G., LAGUNAS-BERNABE, S., CUARON-IBARGUENGOYTIA, J. A., JIMENEZ, R. M. and VAZQUEZ-CHAGOYAN, J. C. 2005. In vitro evaluation of the binding capacity of *Saccharomyces cerevisiae* Sc47 to adhere to the wall of *Salmonella spp.* *Rev. Latinoam Microbiol.*, 47(3-4): 70–75.
- POGACIC, T., SINKO, S., ZAMBERLIN, S. and SAMARZIJA, D., 2013. Micro-biota of kefir grains. *Mljekarstvo*, 63(1): 3–14.
- PUSPAWATI, N. N. and ARIHANTANA, N. M. I. H. 2016. Viability of Lactic Acid Bacteria Isolated from Kombucha Tea Against Low pH and Bile Salt. *Media Ilmiah Teknologi Pangan*, 3(1): 18–25.
- RAHMAN, S. 2014. Studi Pengembangan Dangke sebagai Pangan Lokal Unggulan dari Susu di Kabupaten Enrekang. *J. Apl. Tek. Pangan.*, 3(2): 41–45
- RICKARD, A. K., LEACH, S. A., BUSWELL, C. M., HIGH, N. J. and HANDLEY, P.S. 2000. Coaggregation between aquatic bacteria is mediated by specific growth phase-dependent lectinsaccharide interactions. *Appl. Environ. Microbiol.*, 66(1): 431–434
- ROOSTITA, R. and FLEET, G. H., 1999. Growth of Yeasts Isolated from Cheeses on Organic Acids in the Presence of Sodium Chloride. *Food Technol. and Biotechnol.*, 37: 73–80.
- SALEN, G. and BATTA, A. K. 2004. Bile Formation. In: *Encyclopedia of Gastroenterology*. Elsevier Inc, USA.
- SALMINEN, S., GORBACH, S., KUN LEE, Y. and BENNO, Y. 2004. Human studies on probiotics: What is scientifically proven today? In: SALMINEN, S., VON WRIGHT, A. and OUWEHAND, A. (Eds.). *Lactic Acid Bacteria: Microbiological and Functional Aspect Third Edition*. New York: Marcel Dekker Inc.
- SIMOVA, E., BESHKOVA, D., ANGELOV, E., HRISTOZOVA, T., FRENGOVA, G. and SPASOV, Z., 2002. Lactic acid bacteria and yeasts in kefir grains and kefir made from them. *J. Ind. Microbiol. Biotechnol.*, 28(1): 1–6.
- SHAH, N. P. 2007. Functional cultures and health benefits. *Int. Dairy J.*, 17(11): 1262–1277.
- SOHLIYA, I., JOSHI, S. R., BHAGOBATY, R. K. and KUMAR, R., 2009. Tungrymbai - a traditional fermented soybean food of the ethnic tribes of Meghalaya. *Indian J. Tradit. Knowl.*, 8(4): 559–561.

- SURONO, I. S. 2004. *Probiotic Susu Fermentasi dan Kesehatan*. Yayasan Pengusaha Makanan dan Minuman Seluruh Indonesia, Jakarta.
- SUSANTI, I., KUSUMANINGTYAS, R. W. and ILLANINGTYAS, F. 2007. Probiotic Characteristics of Lactic Acid Bacteria as Candidate for Functional Food. *J. Teknol. Indust. Pangan*, 18(2): 89–95.
- USMIATI, S., BROTO, W. and SETIYANTO, H. 2011. Characteristic of cow milk dadih using starter of probiotic of lactic acid bacteria. *JITV*, 16(2): 140–152.
- UTAMA, G. L., KURNANI, T. B. A., SUNARDI and BALIA, R. L. 2016. The isolation and identification of stress tolerance ethanolfermenting yeasts from mozzarella cheese whey. *Int. J. on Adv. Sci. Eng. and Information Technology*, 6(2): 252–257.
- VICKOVA, V., DUHOVA, V., SVIDOVA, S., FARKASSOVA, A., KAMASOVA, S., VLCEK, D., KOGAN, G., RAUKO, P. and MIADOKOVA, E. 2004. Antigenotoxic potential of glucomannan on four model test systems. *Cell Biol. Toxicol.*, 20(6): 325–332.
- VINE, N. G., DUKES, W. D. and KAISER, H., 2006. Probiotics in marine larviculture. *FEMS Microbiol. Rev.*, 30(3): 404–27
- WAHYUNI, A. 2006. Genetic analysis of *S. agalactiae* caused subclinical mastitis in dairy cattle by Pulsed Field Gel Electrophoresis (PFGE). *Journal of Sain Veteriner*, 26(1): 110–118.
- YANG, Y. L., HUNG, C. C., WANG, A. H., TSENG, F. C., LEAW, S. N., TSENG, Y. T., SU, C. L., CHEN, H. T., LAUDERDALE, T. L. and LO, H. J. 2010. Oropharyngeal colonization of HIV-infected outpatients in Taiwan by yeast pathogens. *J. Clin. Microbiol.*, 48(7): 2609–12.

Contact information

Gemilang Lara Utama: g.l.utama@unpad.ac.id