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Little Tuna (*Euthynnus affinis*) Waste-Based Medium for Fungal Growth and The Lipolytic Activity

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Abstract: Lipase demand keeps increasing as it has wide applications in many sectors of industry, hence producing lipase is economically prospective. Finding a cheap medium is important for lipase production for commercial purpose. Waste based medium KIT1, KIT2 and KIT3 were alternately assessed for the capability to support growth and lipolytic activity of the fungi. SDA either or PDA were used as the control Medium. Fungi isolated from Coconut or Avocado flesh were used. Observation was made on day 1 and day 2 after inoculation. The result suggests that KIT2 gave similar growth and lipolytic activity to SDA medium, and better growth and lipolytic activity on KIT3 medium as compare to SDA and PDA.

Keywords: Waste-based medium; lipase; lipolytic activity; Euthynnus affinis

Introduction

This research is conducted to assessed gradually the capability of cheap Little-Tuna Waste-based Medium in supporting fungal growth and the activity as preliminary study for lipase production in a low cost. Lipase is an enzyme that convert various substrates into various products leading to a wide range of its industrial application such as on food (Guerrand, 2017). drink, leather, and pulp industrial processes (Andualema and Gessesse, 2012; Hasan et al., 2006). It was suggested that lipase can work in harsh conditions including high pH and temperature, non-polar solvent and less water (Patel et al., 1996). The use of lipase as part of ingredient of detergent-based industry (Chauhan et al., 2013), particularly those that highly alkaline lipase (Cherif et al., 2011). Lipase also can be applied for bioremediation (Okino-Delgado et al., 2017). All these increase the market demand of lipase. Indonesia meets the demand of enzymes in general (including lipase) by importing them from abroad, hence, working on enzyme may help to meet the demand as well as economically prospective. Astuti (2014) suggests that producing lipase using agricultural waste medium in Indonesia is still beneficial.

Naturally fungi are growns on many substrates (Uthayasooriyan et al., 2016). Various carbohydrates were investigated to be used on bacterial medium, and it shown that canna edulis was best for Ecoli and Disocore for stapilococcus aureus (Anisah and Rahayu, 2015; Adesemoye & Adedire, 2005). Local grains and legumes protein source have reported to be used for alternative medium (Uthayasooriyan et al., 2016). Jannah et al. (2021) has formulated alternative medium for BAL using tofu waste. There are numerous alternative media have been researched that is not mentioned here. This is indicating that may not be difficult to grow fungi on any substrates.

Little-Tuna Fish is mostly available on traditional market in most of the year in Lombok Island particularly, and also common in Indonesia as an archipelago country, especially at the coastal area. In the market, the fish is available as raw fish or boiled that is called as 'pindang'. The waste of boiling fish is a huge. When it is not been handle properly, it potentially pollutes the environment (as due to the richness of organic molecules. As the broth is still rich in nutrition's

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therefore, some are used as row material for fish sauce (petis, Indonesian term), organic fertilizer, animal feed (Astuti, 2014). Hence the broth may be used as medium microbial growth.

For industrial purposes, lipase is generally produced from microbes, because of several reasons: 1) microbial substrates are cheap and abundant, 2) icrobial enzymes normally are more stable, 3) easy to produce on a large scale, in a short time and 4) mcrobial growth does not depend on the season. All of these things make lipase production cost is cheap (Guerrand, 2017; Pratush, et al., 2013).

From previous work, it was obtained fungi isolated from coconut and Avocado flesh showing a powerful lipolytic activity on SDA medium (Sedijani et al., 2021). In this report the assessment results tof he use of Little-Tuna broth as main ingredient to prepare medium for fungal growth and the lipolytic activity. The medium then is annotated as KIT medium, stand for Kuah Ikan Tongkol (Indonesian term for waste water after being used to boil Little-Tuna (Tongkol) Fish. The report discus if the Waste-based-medium is able to support fungal colony's growth and the lipolytic activity.

Method

The work was conducted in 3 step of refinement medium. The control medium either-or SDA and PDA were used. Added material consist of 1% olive oil, 1% emulsifier and Rhodamin B, is notified as Substrate group. First KIT medium composed of 5% or 10% fish broth and 100 gram/L sliced potato. Fish broth was prepared as follow, separately fish or potato were put on a cooking pot containing 1 L of tap water, the level of water surface was marked. The pot was boiled for 1 h at low heat, hot water was added until the mark. The broth was filtered using cotton ball to reduce the turbidity. The broth was used to make medium by adding Substrate group within the broth, and pH8 was adjusted. A 20 gr/L agar was added to solidify the medium before autoclave. Second KIT2 medium was prepared as the First Medium but fish and sliced potato were boiled together within the same pot containing 1L of Tap water. While the third KIT3 medium was prepared as the KIT2 medium but using 200 grams/L instead of 100 grams/L sliced potato. Inoculation was done using tooth pick right in the middle of petri dish. The experiment was set in 3 replicates. The colony's growth and the clear zone were recorded on day 1 and day 2 after inoculation. The lipolytic activity index was calculated using formula (Kouker and Jeager, 1987) equation 1:

$$\text{Index Activity} = \frac{\text{Ø zona bening} - \text{Ø koloni} \times 100\%}{\text{Ø koloni}} \quad (1)$$

Result and Discussion

Growth and Lipolytic activity of Isolates on KIT1 Medium.

Appearance of clear zone on petri dish is presented on Figure 1.

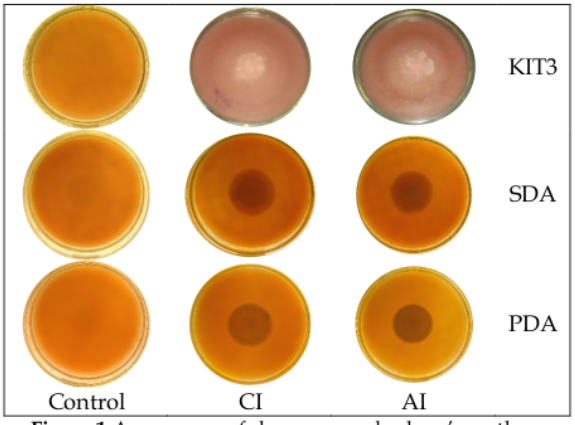
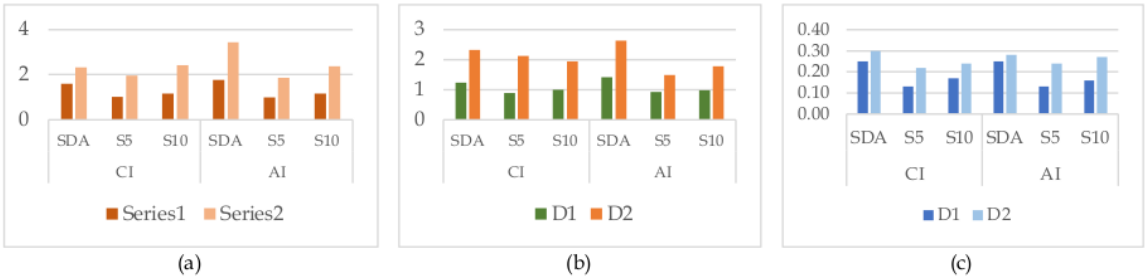


Figure 1. Appearance of clear zone and colony's groth on indicated edium. CI = Cococnut Isolate and AI = Avocado Isolate

The growth and the activity of both isolates on KIT1 and SDA medium is presented on Chart 1. The colony's growth and their activity on SDA medium were higher than those on KIT1 medium either on 5% nor at 10% of fish broth. Likewise with the clear zone and its lipolytic activity.

The result suggests that the KIT1 medium, fish broth in this case is little tuna up to 10% did not give enough support for growth and lipolytic activity as the control medium does.



(a) (b) (c)

Figure 2. Growth and lipolytic activity of colony on different KIT1 medium. S5 = 5% KIT1, S10 = 10% KIT1, SDA. Clear zone (Top), Colony's diameter (middle), Lipolytic activity (bottom).

It was suspected that 5 to 10% of fish broth may not give enough protein for the fungal growth. Hence an improvement of medium composition was done using the second medium in which fish broth without addition of water was used to prepared the medium. Colony's growth and lipolytic activity on the KIT2 medium is presented on Figure 3.

Growth and Lipolytic activity of Isolates on KIT2 Medium.

The Figure 3 shows that the KIT2 medium, colony diameter, clear zona and lipolytic activity showed similar to that on SDA medium upon both isolates CI and AI suggesting that the KIT2 medium was able to support the fungal colonies growth and their activity commensurate with the SDA medium. KIT2 medium therefore, can be used as an alternative for SDA medium. Fish powder can be used as a substitute for protein sources medium that is produced by factories (Soleymani et al., 2017). In KIT medium, however, the fish broth only that is used, therefore, the fish flesh still can be consumed reducing the medium cost. Other cheap raw material for medium preparation reported as waste of tofu production (Martínez-Moreno et al., 2021; Jannah et al., 2021; Prayekti and Lukiyono, 2022).

Further work then was done to obtained a medium that support growth and lipolytic activity better than SDA and PDA do. It is assumed that if the energy source increases, the growth and lipolytic activity might be increased, for this purpose the KIT3 medium was applied. This medium contained 200 gr/L instead of 100 gr/L sliced potato on KIT2 medium.

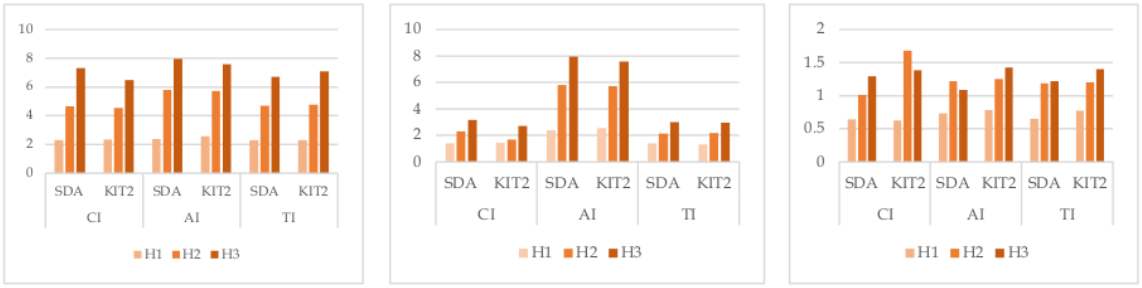
Growth and Lipolytic activity of Isolates on KIT3 Medium.

Colony's growth and lipolytic activity on the KIT3 medium is presented on Figure 1 and Figure 4. These suggest that colony's growth and lipolytic activity on KIT3 medium were higher than that on SDA and PDA media $p < 0.000$ based on Two Way ANOVA using SPSS application. The result also suggest that KIT3 medium might be an alternative for SDA or PDA that are expensive, with even better support for fungal growth and it activity.

Other protein source have also been reported, such as the use of local grains and legumes protein source (Uthayasooriyan et al., 2016). Jannah et al. (2021) has formulated alternative medium for BAL using tofu waste. This finding might also give information to school teacher where which the laboratory resource are limited. They can conduct practical class when they need medium for fungal growth. It might be test also to grow bacteria.

Weakness and strength o this experiment

The weakness on this experiment is shown by the inconsistence of the result between rounds of experiment. The lipolytic activity for instance, when isolate grown on the same medium but different lot of experiment showing different result for no reason or difficult to be explain.



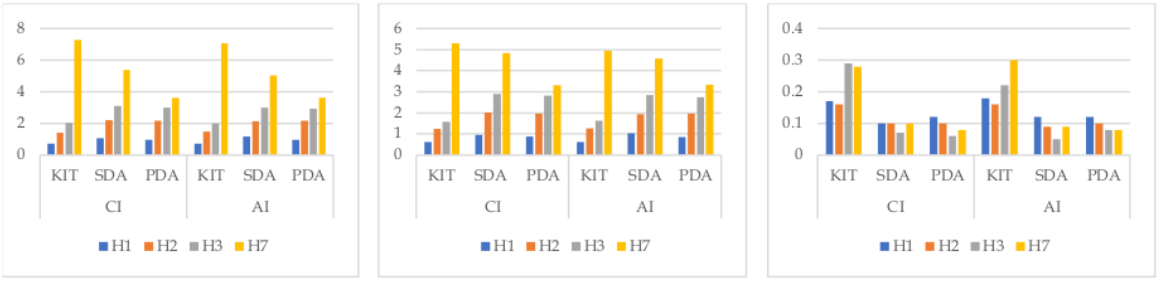
(a) (b) (c)

Figure 3. Growth and lipolytic activity of colony on KIT2 and SDA. Clear zone (Top), Colony's diameter (middle), Lipolytic activity (bottom).

However, lot1 (experiment on KIT1) and Lot3 (experiment on KIT3) show similar result, only lot2 (experiment on KIT2) showed higher result on the parameters used than that on KIT1 and KIT3. This could possibly doe to the nutrition variability of the fish being boiled although it has been carefully selected, however, the season and location they grown were uncontrollable. The broth nutrition analysis is vary depended on the area as well as the company in which the broth is collected from (Astuti, 2014). Although in this experiment, the process of producing the broth was strictly controlled except the season and their place/origin of the fish. Beside mention, KIT media, appears turbidity that reduce visibility to see the clear zone. These could be the constrains of using fish broth for research purposes. For rpoduction medium, however, it might not be problem.

The strength of this research, all those differences are proportional on every medium within the lot. Each lot of experiment showed consistent result according to the medium. For instance, lipolytic activity on KIT 2 Experiment showed higher than that of KIT1 and KIT 3.

This phenomenon was also shown on SDA and PDA media, both upon CI and AI. Likewise, clear zone, and colony's growth.



(a) (b) (c)

Figure 4. Growth and lipolytic activity of colony on KIT3, SDA and PDA. Clear zone(Top), Colony's diameter (middle), Lipolytic activity (bottom).

These suggest that the medium determined the growth and activity of the isolates, and therefore this weakness is actually become the strength of the experiment, high light the medium really does determined the result. KIT3, therefore gives the better growth and better lipolytic activity for the fungi than SDA and PDA do. Considering that the fish broth is no limit in amount, and availability, it is very prospective to use the broth for lipase production.

Further experiment is still on going, such as adding different concentration of sugar that indicates to increase lipase production (personal data, not shown); inducer and glucose (Ardani et al., 2021). Trials to make clear medium by filtering vigorously as well as replacing potato by sugar will be done, at the same time try to increase lipase yield.

Conclusion

From above discussion, it can be concluded that waste based KIT2 medium might be used as an alternative medium to grow fungi (CI and AI) and their lipolytic activity similar to SDA and PDA and KIT3 showed a better performance.

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References

Adesemoye, A. O., & Adedire, C. O. (2005). Use of cereals as basal medium for the formulation of alternative culture media for fungi. *World journal of microbiology and biotechnology*, *21*(3), 329-336. https://doi.org/10.1007/s11274-004-3907-4

Andualema, B., & Gessesse, A. (2012). Microbial Lipases and Their Industrial Applications: Review. *Biotechnology*, *11*(3), 100-118. https://doi.org/10.3923/biotech.2012.100.118

Anisah, A., & Rahayu, T. (2015). Media Alternatif untuk Pertumbuhan Bakteri Menggunakan Sumber Karbohidrat yang Berbeda. *Seminar Nasional XII Pendidikan Biologi FKIP UNS: Biologi, Sains, Lingkungan, dan Pembelajarannya*, 855-860. Retrieved from https://www.google.com/url?sa=t&rct=j&q=&esrc=s&source=..AOvVaw0DamdsZups1VSKfoCtYFKY

Ardani, A., Rismayanti, S., & Abdillah, H. (2021). Effect of surfactant type modifications, glucose inducer concentrations, and mineral sources towards lipase enzyme activity of Aspergillus niger ITBCC L74 on rice bran substrate. *Basic and Applied Science Conference (BASC) 2021. NST Proceedings*. pages 1-7. https://doi.org/10.11594/nstp.2021.1101

Astuti, A. D. (2014). Pemanfaatan Limbah Cair Pemindangan Ikan. *Jurnal Litbang*, *10*(2), 114-122. https://doi.org/10.33658/jl.v10i2.83

Chauhan, M., Chauhan, R. S., Garlapati, V. K. (2013). Evaluation of a new lipase from Staphylococcus sp. for detergent additive capability. *Biomed Research International*, 1-6. https://doi.org/10.1155/2013/374967

Cherif, S., Mnif, S., Hadrich, F., Abdelkafi, S., & Sayadi, S. (2011). A newly high alkaline lipase: an ideal choice for application in detergent formulations. *Lipids in Health and Disease*, *10*(221). https://doi.org/10.1186/1476-511X-10-221

Guerrand, D. (2017). Lipase Industrial Application: Focus on Food and Agro industri. *Oilseeds & Fats Crops and Lipids*. *24*(4), D403.

https://doi.org/10.1051/ocl/2017031

Hasan, F., Shah, A.A., & Hameed, A. (2006). Industrial Applications of Microbial Lipases. *Enzyme and Microbial Technology*, *39*(2), 235–251. https://doi.org/10.1016/j.enzmictec.2005.10.016

Jannah, S. N., Pujiyanto, S., Rosiana, E., and Purwantisari, S. (2021). Formulation and optimization of alternative culture media for probiotic bacteria growth using tofu liquid waste. *Journal of Physics: Conference Series 1943*(012070). https://doi.org/10.1088/1742-6596/1943/1/012070

Kouker, G,. and Jeager, K-R. (1987). Specific and Sensitive Plate Assay for Bacterial Lipases. *Applied Science and Environmental Microbiology*, *53*(1), 211-213. https://doi.org/10.1128/aem.53.1.211-213.1987

Okino-Delgado., Prado, D. Z., Facanali, R., Marques, M. M. O., Nascimento, A. S., Fernandes, C. J. D., Zambuzzi, W. F., and Fleun, L. F. (2017) Bioremediation of cooking oil waste using lipases from wastes. *PLoS One*, *12*(10), e0186246. https://doi.org/10.1371/journal.pone.0186246

Patel, M.T., Kilara, A., & Nagarajan, R. (1996). Lipase-catalyzed biochemical reactions in novel media: A review. *Chemical Engineering Communications*, *152-153*(1), 365-404. https://doi.org/10.1080/00986449608936574

Pratush, A., Gupta, A., Vyas, G., & Sharma, P. (2013). *Bacterial Lipases: Production Strategies and Industrial Applications* In Book: Microbiology Application (64-83). India: Bhalla publishers, Dehradun.

Prayekti, E., & Lukiyono. (2022). Penggunaan Tepung Ampas Tahu Untuk Media Pertumbuhan Candida albicans dan Candida sp. *Journal of Indonesian Medical Laboratory and Science*, *3*(2). https://doi.org/10.53699/joimedlabs.v3i2.122

Martínez-Moreno, F., Jofre y Garfias, A., Hernandez-Orihuela, A., & Martínez-Antonio, A. (2021). Avocado seed hydrolysate as an alternative growth medium for fungi. *Revista Mexicana De Ingeniería Química*, *20*(2), 569-580. https://doi.org/10.24275/rmiq/Bio1951

Sedijani, P., Rasmi, D. A. C. ., Kusmiyati, K., & Anggriani, R. A. (2021). Powerful Lipolytic Activity of Fungi Isolated from Coconut and Avocado Flesh on Different pH and Temperature. *Jurnal Penelitian Pendidikan IPA*, *7*(Special Issue), 365–369. https://doi.org/10.29303/jppipa.v7iSpecialIssue.1261

Soleymani, S., Alizadeh, H., Mohammadian, H., Rabbani, E., Moazen, F., MirMohammad, Sadeghi, H., Shariat, ZS., Etemadifar, Z., & Rabbani, M. (2017). Efficient Media for High Lipase Production: One Variable at a Time Approach. *Avicenna J Med Biotechnol*, *9*(2), 82-86. Retrieved from https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5410133/pdf/AJMB-9-82.pdf

Uthayasooriyan, M., Pathmanathan, S., Ravimannan, N., and Sathyaruban, S. (2016). Formulation of alternative culture media for bacterial and fungal growth. *Scholars Research Library Der Pharmacia Lettre*, *8*(1), 431-436. Retrieved from http://scholarsresearchlibrary.com/archive.html

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