

## Anticandida Activity of Gut Symbiont Bacteria from Sea Urchins Against *Candida albicans*, the Causative Agent of Candidiasis

Dita Delia Rothman<sup>1</sup>, Risky Hadi Wibowo(\*)<sup>1,2</sup>, M. Eka Prastya<sup>4</sup>,  
Sipriyadi Sipriyadi<sup>1,2</sup>, Morina Adfa<sup>3</sup>, Uci Cahlia<sup>2</sup>

<sup>1</sup> Master Study Program of Biology, Department of Biology, Mathematics and Natural Sciences  
Faculty, Bengkulu University

<sup>2</sup> Department of Biology, Mathematics and Natural Sciences Faculty, Bengkulu University

<sup>3</sup> Department of Chemistry, Mathematics and Natural Sciences Faculty, Bengkulu University  
Jl. W. R. Supratman, Kandang Limun, Bengkulu, Indonesia Postcode 38122;

<sup>4</sup> Research Center for Drug Raw Materials and Traditional Medicine, National Research and  
Innovation Agency (BRIN)  
Chemistry Research Center Building, B.J. Habibie Science and Technology Area (Puspitek), Serpong,  
South Tangerang, Indonesia

\* Corresponding Author: [rhwibowo@unib.ac.id](mailto:rhwibowo@unib.ac.id)

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### Abstract

**Background:** *Candidiasis* is an opportunistic fungal infection caused by *Candida albicans*, particularly affecting immunocompromised individuals. Increasing antifungal resistance has encouraged the search for natural anticandidal agents, including sea urchin gut symbiotic bacteria. Information regarding the anticandidal activity of gut symbiotic from *Stomopneustes variolaris* and *Tripneustes ventricosus* remains limited. This study evaluated the anticandidal potential of gut symbiotic bacterial isolates from these sea urchins collected from Laguna Beach and Pengubaian Beach, Kaur Regency, Bengkulu. **Methodology:** Six bacterial isolates, TVL 6, TVL 11, TVL 13, SVP 1, SVP 6, and SVP 15 were subcultured on Zobell Marine Agar (ZMA) and tested against *C. albicans*. Anticandida activity was evaluated using the spot inoculation method for bacterial cultures and the disk diffusion method for bacterial pellets. Nystatin and sterile distilled water served as positive and negative controls, with all assays performed in triplicate. Inhibition zones were measured using a digital caliper. **Findings:** All isolates inhibited the growth of *C. albicans*. In the culture assay, isolate TVL 6 showed the highest inhibition zone diameter of 11.58 mm, while SVP 1 showed the lowest activity (8.08 mm). In the pellet assay, TVL 11 exhibited the highest activity (16.65 mm), whereas SVP 15 showed the lowest activity (5.50 mm). Overall, pellet preparations produced larger inhibition zones than culture preparations. **Contributions:** Gut symbiotic bacteria from sea urchins collected in Kaur Regency, Bengkulu, showed promising anticandidal activity against *C. albicans*, supporting their potential as marine-derived sources of alternative anticandidal against and providing a basis for future studies on isolate identification, bioactive metabolites, and mechanisms of action.

**Keywords:** Anticandida; Gut; Sea Urchin; Symbiont



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## INTRODUCTION

Fungal infections represent one of the major challenges in global health, particularly those caused by *Candida* species, including *Candida albicans*. *Candida albicans* is an opportunistic pathogen that naturally exists as part of the normal human microbiota, especially in oral cavity, gastrointestinal tract, and urogenital tract. However, under the certain conditions, such as immunosuppression, prolonged antibiotic use, or other chronic diseases, this fungus can overgrow and cause candidiasis (Talapko et al., 2021). Candidiasis can affect various parts of the body, ranging from superficial, such as oral and vaginal candidiasis, to life threatening systemic infections, particularly in immunocompromised patients. The increasing incidence of systemic candidiasis and the emergence of resistance to conventional antifungal agents have become serious concerns in the healthcare sector (Alves et al., 2022).

Globally, *Candida* bloodstream infections (candidemia) are estimated to affect approximately 1,5 million individuals annually and cause nearly 995,000 deaths. In Indonesia, candidemia is estimated to occur in approximately 26,700 individuals each year, with around 25% of cases reported among patients admitted to intensive care units (ICUs) (Mayasari & Siregar, 2026). The high incidence and mortality rates associated with *Candida* infections, together with the increasing prevalence of antifungal resistance, highlight the urgent need to discover new and effective anticandida agents.

One promising approach is the exploration of marine resources, including marine microorganisms known for their ability to produce diverse bioactive compounds. This capability results from adaptation to extreme marine environmental conditions, such as high salinity, temperature fluctuations, nutrient limitation, and competition with other organisms. These environmental pressures stimulate the production of various bioactive compounds with antimicrobial, antifungal, and anticandida activities (Setiasih et al., 2020). Sea urchins (*Echinoidea*) are marine invertebrates known to maintain symbiotic relationships with a wide range of microorganisms, particularly bacteria (Rompas et al., 2022). Symbiotic bacteria inhabiting the body and digestive tract of sea urchins play important roles in digestion, pathogen defense, and the production of metabolites that support host physiology (Chen et al., 2024).

The diversity of symbiotic bacteria associated with sea urchins has been widely reported. Rodríguez-Barrera et al. (2021) reported that *Echinometra lucunter* and *Diadema antillarum* harbor bacterial communities dominated by the phyla Planctomycetes, Proteobacteria, Fusobacteria, and Firmicutes. Similar bacterial diversity has also been observed in other sea urchin species, including *Lytechinus variegatus*, *Paracentrotus lividus*, and *Strongylocentrotus purpuratus*, which are associated with bacterial genera such as *Pleurocapsa*, *Rhodopirellula*, *Pelagibius*, *Arcobacter*, *Sulfurimonas*, *Psychromonas*, and *Propionigenium* (Faddetta et al., 2020). The high diversity of these bacterial communities indicates that sea urchins constitute a promising habitat for microorganisms capable of producing bioactive metabolites.

Numerous studies have demonstrated the potential of sea urchins as sources of bioactive compounds. Extracts obtained from sea urchin gonads, spines, and shells contain various compounds, including steroids, saponins, triterpenoids,

alkaloids, phenolics, and polyketides, which exhibit antimicrobial activities (Akaerina et al., 2015; Rompas et al., 2022). Abouelmaaty et al. (2020) reported that gonad and spine extracts of *T. gratilla* were able to inhibit the growth of *Enterococcus faecalis*. Likewise, gonad extracts of *Diadema setosum* containing alkaloids, phenolics, and saponins demonstrated inhibitory activity against *Escherichia coli* and *Staphylococcus aureus* (Rompas et al., 2022).

In addition, antimicrobial activity in marine organisms may also originate from their associated symbiotic bacteria. *Bacillus velezensis* isolated from marine environments exhibited antibacterial activity against *S. aureus*, producing inhibition zones of up to 35 mm (Chakraborty et al., 2021), whereas *Pseudoalteromonas flavipulchra* isolated from sea urchins was reported to inhibit the growth of *S. aureus* and *E. coli* (Fofied et al., 2018). These findings suggest that marine symbiotic bacteria constitute a valuable source of bioactive metabolites involved in host defense mechanisms against pathogenic microorganisms.

Previous studies have also shown that marine bacteria produce secondary metabolites, including lipopeptides, biosurfactants, and polyketides, which possess anticandida activity against various *Candida* species (Lei et al., 2019; Ridwan et al., 2024; Wu et al., 2019). Furthermore, several sea urchin species, including *T. ventricosus*, have been reported to exhibit anticandida activity against *Candida albicans* (Moreno-García et al., 2022). These findings support the hypothesis that symbiotic bacteria associated with marine organisms are capable of producing secondary metabolites with potential anticandida properties.

Kaur Regency, Bengkulu Province, possesses relatively pristine coastal ecosystems that serve as natural habitats for various sea urchin species, including *S. variolaris* and *T. ventricosus* (Hafiza et al., 2025). The stable environmental conditions in this region facilitate complex interactions between sea urchins and their associated symbiotic microorganisms. Such interactions may promote the production of diverse bioactive metabolites that remain largely unexplored, including compounds with anticandida activity.

Although numerous studies have reported the biological activities of sea urchin extracts and the diversity of their associated bacterial communities, investigations on the anticandida potential of gut symbiotic bacteria from sea urchins remain very limited. Most previous studies have focused on bioactive compounds derived from sea urchin tissues, whereas the utilization of gut symbiotic bacteria as a source of anticandida metabolites has received little attention. Moreover, to the best of our knowledge, no studies have reported the anticandida activity of gut symbiotic bacteria associated with *S. variolaris* and *T. ventricosus* collected from the coastal region of Kaur Regency, Bengkulu.

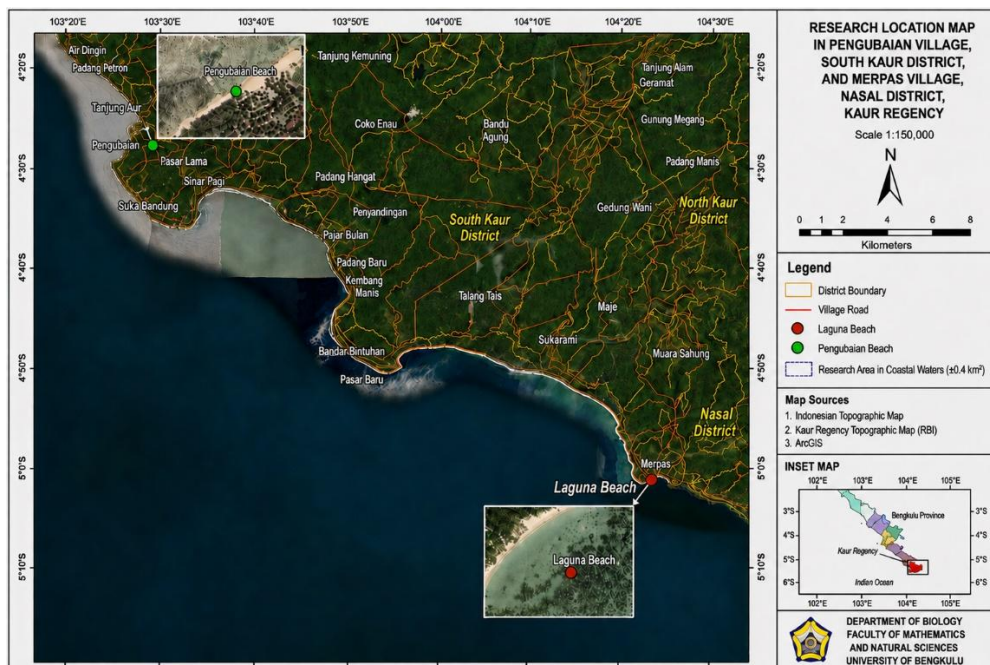
Therefore, this study was conducted to investigate the anticandida potential of gut symbiotic bacterial isolates obtained from *S. variolaris* and *T. ventricosus* collected from Pengubai Beach and Laguna Beach, Kaur Regency, Bengkulu Province. The novelty of this study lies in the exploration of gut symbiotic bacteria from two sea urchin species as potential producers of anticandida metabolites and the comparison of their inhibitory activities using both bacterial culture and bacterial pellet assays against *Candida albicans*. The findings of this study are expected to provide preliminary

information regarding the anticandida potential of sea urchin gut symbiotic bacteria and serve as a foundation for future research on the development of novel anticandida agents derived from marine microorganisms.

## METHOD

### Sample Collections

The collection of sea urchins was conducted in April 2024. Samples were collected from Pengubaian Beach and Laguna Beach, Kaur Regency, Bengkulu (Figure 1). A total of four sea urchin specimens were collected, consisting of two individuals of *S. variolaris* and two *T. ventricosus*. The recorded body weights of *S. variolaris* were 66,08 g and 71.59 g, while those of *T. ventricosus* were 235,14 g and 250,11 g. Sample preparation and analyses were carried out in the Microbiology Laboratory and Biotechnology Laboratory, Basic Science Building, Biology Study Program, Faculty of Mathematics and Natural Sciences, University of Bengkulu.



**Figure 1.** Sampling Locations of Sea Urchins at Pengubaian Beach and Laguna Beach, Kaur Regency, Bengkulu

The sea urchins *S. variolaris* and *T. ventricosus* were collected using a purposive sampling method, targeting individuals inhabiting sandy flats and coral reef ecosystems. The specimens were carefully retrieved using tongs and placed into sterile ziplock plastic bags. The samples were then documented, and abiotic parameters were recorded. Water temperature was measured using a Thermo Scientific thermometer (accuracy  $\pm 0.5$  °C), pH using a Thermo Scientific pH meter (accuracy  $\pm 0.01$ ), and salinity using an Atago refractometer (accuracy  $\pm 0.006$ ).

### **Preparation of Sea Urchins Digestive Tract Sample**

The collected sea urchin specimens were transported to the laboratory and rinsed with sterile distilled water to remove debris and surface contaminants. All procedures were performed under aseptic conditions. Each specimen was dissected by carefully cutting the test into two halves using a sterile scalpel. The digestive tract was aseptically removed using sterile forceps and transferred into a sterile mortar. Subsequently, the digestive tract tissue was homogenized using a sterile pestle until a uniform homogenate was obtained (Padang et al., 2019).

### **Isolation and Morphological Characteristics**

Sea urchin symbiotic bacteria were isolated using a homogenization method. 1 gram of sample was placed in a screw-cap tube, homogenized with sterile distilled water, and serially diluted to  $10^{-1}$ ,  $10^{-3}$ , and  $10^{-5}$ . Subsequently, 0,1 ml of each dilution was spread onto the culture medium using a sterile spreader and incubated at 30 °C for 48 hours. the resulting bacterial colonies were purified on sterile *Zobell Marine Agar* (ZMA) using the quadrant streak method and incubated again at 30 °C for 48 hours. the purified isolates were then characterized based on colony morphology through macroscopic observation, including colony appearance, surface form, color, margin, and elevation (Wibowo et al., 2020).

### **Subculturing of Gut Symbiont Bacteria from Sea Urchins**

The bacterial isolates used in this study (SVP 1, SVP 6, SVP 15, TVL 6, TVL 11, and TVL 13) were obtained from a previous study on the diversity of bacteria associated with the digestive tract of sea urchins. The isolates were subsequently subcultured and purified on *Zobell Marine Agar* (ZMA), followed by incubation at 30 °C for 48 hours. The purified isolates were then stored in at 4 °C for further analysis.

### **Anticandida Activity Test**

Anticandida activity was tested using both culture and pellet of bacterial isolates. *Candida albicans* was cultured in 25 ml of *Sabouraud Dextrose Broth* (SDB) and incubated on a shaker for 24 hours. 1 ml of the culture was added to 100 ml of *Sabouraud Dextrose Agar* (SDA) that had been cooled to approximately 40 °C, homogenized, and poured into Petri dishes at a volume of approximately 15 ml per dish. The symbiont bacterial isolates were subsequently inoculated onto the SDA medium containing *C. albicans* using the spot inoculation method and incubated at 37 °C for 24 hours (Collins & Lyne, 2004).

The anticandida activity of bacterial pellet initiated by culturing each gut symbiont bacterial in *Zobell Marine Broth* (ZMB) and incubating the cultures in a shaker incubator at 120 rpm for 24 hours. Subsequently, 1,5 ml of the culture was centrifuged at 10,000 rpm for 5 minutes. The resulting pellet was separated from the supernatant and resuspended in 150 µl of sterile distilled water, followed by vortexing. A volume of 10 µl of the resuspended pellet was then applied onto a sterile paper disc (6 mm diameter) and air-dried. The anticandida activity was subsequently evaluated using the disk diffusion method by placing the discs onto *Sabouraud Dextrose Agar*

(SDA) plates previously inoculated with *C. albicans*, followed by incubation at 37 °C for 24 hours. The procedure was performed in triplicate. As a positive control, paper discs containing nystatin were used, while discs loaded with sterile distilled water served as the negative control. Anticandida activity was indicated by the formation of a clear inhibition zone surrounding the disc, which was measured using a digital caliper (Wibowo et al., 2023).

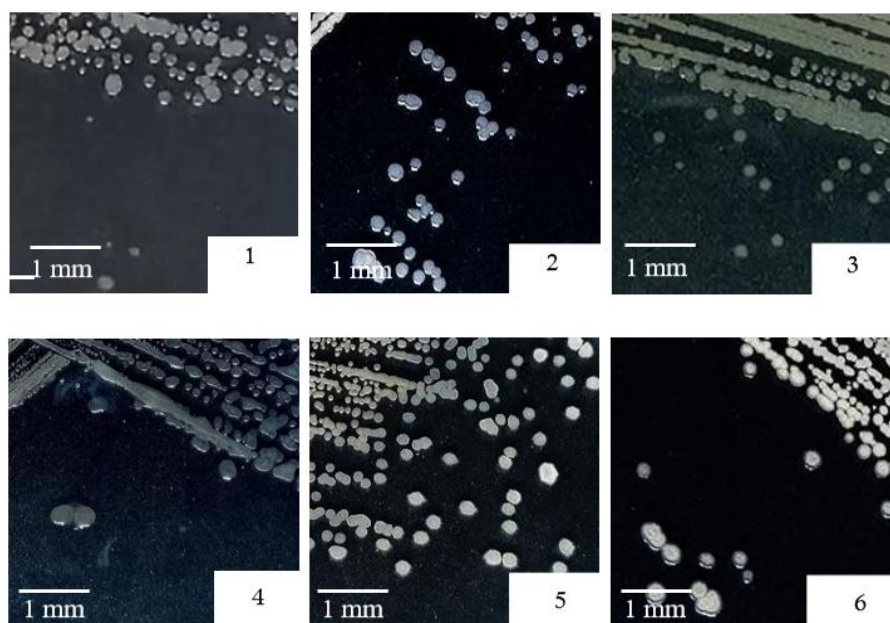
### Data Analysis

Data were analyzed using qualitative and quantitative descriptive methods. Antimicrobial inhibition was determined based on the inhibition index of Ouchari et al., (2019) by comparing inhibition zone diameters, measured with a digital caliper after substrating the disk diameter.

## RESULT AND DISCUSSION

### Subculturing of Gut Symbiont Bacteria from Sea Urchins

The gut symbiont bacterial isolates from the sea urchins *S. variolaris* and *T. ventricosus* were obtained from the Microbiology Laboratory and Biotechnology Laboratory collections, University of Bengkulu, encoded with SVP 1, SVP 6, SVP 15 and TVL 6, TVL 11, TVL 13. The isolates were subcultured on ZMA using the quadrant streak plate method and incubating them for 48 hours at 30 °C. The quadrant streak technique provides the necessary nutrient stimulation and spatial distribution required for bacterial cells to resume growth from a dormant phase. The subculturing process aims to reactivate dormant cells so that the bacteria regain metabolic activity and return to optimal growth (Carlina et al. 2020; Kantari & Ariyanti, 2024). The results of purified bacterial isolates are presented in Figure 2.



**Figure 2.** Results of subculturing of gut-symbiont bacterial isolates from sea urchins on ZMA, incubated at 30°C for 48 hours; 1= SVP 1; 2= SVP 6; 3= SVP 15; 4= TVL 6; 5= TVL 11; 6= TVL 13

Based on the observation of colony morphology, the bacterial isolates obtained from the digestive tracts of *S. variolaris* and *T. ventricosus* exhibited considerable morphological diversity. Variations were observed in colony appearance, surface, margin, and color, although most isolates displayed a flat elevation and were predominantly white in color. These differences indicate the presence of diverse bacterial populations associated with both sea urchin species, as summarized in Table 1.

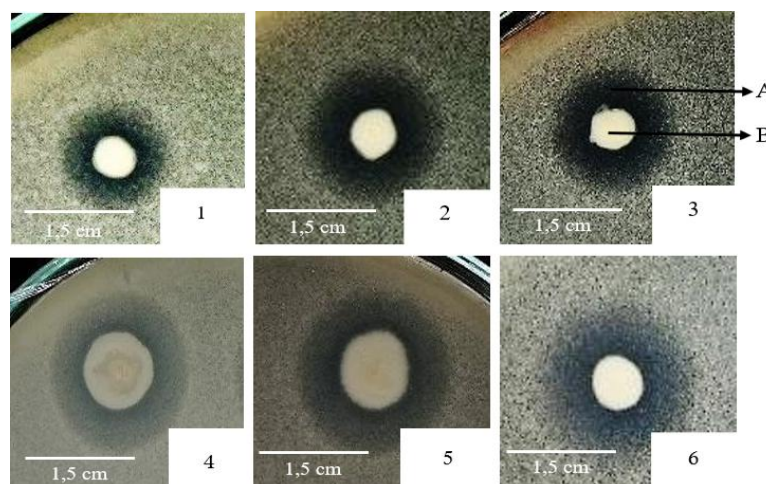
**Table 1.** Observation of morphological colony of symbiotic bacteria isolated from the sea urchins *S. variolaris* and *T. Ventricosus*

Isolate Code	Appearance	Surface	Elevation	Margin	Color
SVP 1	Circular	Smooth	Flat	Entire	White
SVP 6	Circular	Smooth	Flat	Entire	White
SVP 15	Circular	Wrinkled	Flat	Entire	Yellow
TVL 6	Irregular	Smooth	Flat	Entire	White
TVL 11	Irregular	Contoured	Flat	Undulate	White
TVL 13	Circular	Radiated	Flat	Entire	Cream

Description SVP: *Stomopneustes variolaris* Pengubaian; TVL: *Tripneustes ventricosus* Laguna

### Anticandida Activity Test

The anticandida activity test of symbiotic bacterial isolates from the sea urchins digestive tract showed that several isolates were capable in producing inhibition zones against *C. albicans*. The bacterial isolates that produced inhibition zones against *C. albicans* were SVP 1, SVP 6, SVP 15, and TVL 6, TVL 11, and TVL 13. The strongest anticandida activity against *C. albicans* was exhibited by TVL 6 isolate with an inhibition zone of 11.58 mm, whereas the weakest anticandida activity showed by SVP 1 isolate, with an inhibition zone of 8.08 mm. According to [Faturrahman et al., \(2022\)](#), the wider the inhibition zone produced by a bacterial isolate, the greater its potency to inhibit the growth of pathogenic fungi. The results of the anticandida activity test are presented in Figure 3.



**Figure 3.** Inhibition zones produced by symbiotic bacterial isolates from the digestive tract of *S. variolaris* (Pengubaian) and *T. ventricosus* (Laguna) against *C. Albicans*. 1 = SVP 1; 2 = SVP 6; 3 = SVP 15; 4 = TVL 6; 5 = TVL 11; 6 = TVL 13, all of isolates were able to inhibit the growth of *C. albicans*; A = Inhibition zone; B = isolates culture

The anticandida activity of a material is influenced by the bioactive compounds it contains. The bioactive components that can inhibit the fungal growth are generally originate from the groups of saponins, triterpenoids, and steroids. This is consistent with the findings of [Akaerina et al. \(2015\)](#), who reported that the bioactive compounds present in sea urchin gonad extracts are flavonoids, saponins, steroids, and triterpenoids. Flavonoids are widely recognized as anticandida agents. These compounds possess hydroxyl groups that interact with the phospholipids in fungal cell membranes, forming complexes that disrupt membrane structure. As a result, fungal growth is inhibited, membrane permeability increases, and the cells undergo denaturation ([Ratnah et al., 2023](#)).

Saponins are found in several marine organisms, including sea cucumbers and sea urchins ([Yulia et al., 2023](#)). Saponins act as anticandida agents by lowering surface tension and increasing membrane permeability, which leads to leakage of cellular contents. Their polar surfactant properties can disrupt lipids in fungal cell membranes, interfere with the diffusion of essential substances, and ultimately cause the fungal cells to rupture or swell ([Anjelin et al., 2023](#)). This is further supported by [Prastya et al. \(2022\)](#), who stated that secondary metabolites such as flavonoids and saponins possess anticandida activity, particularly through mechanisms involving membrane disruption and interference with cellular metabolism. Symbiotic bacterial isolates that exhibited anticandida activity were evaluated by measuring the diameter of the inhibition zones using a digital caliper. The results of the anticandida activity measurements are presented in Table 2.

**Table 2.** Results of Anticandida Activity Test using Bacterial Culture

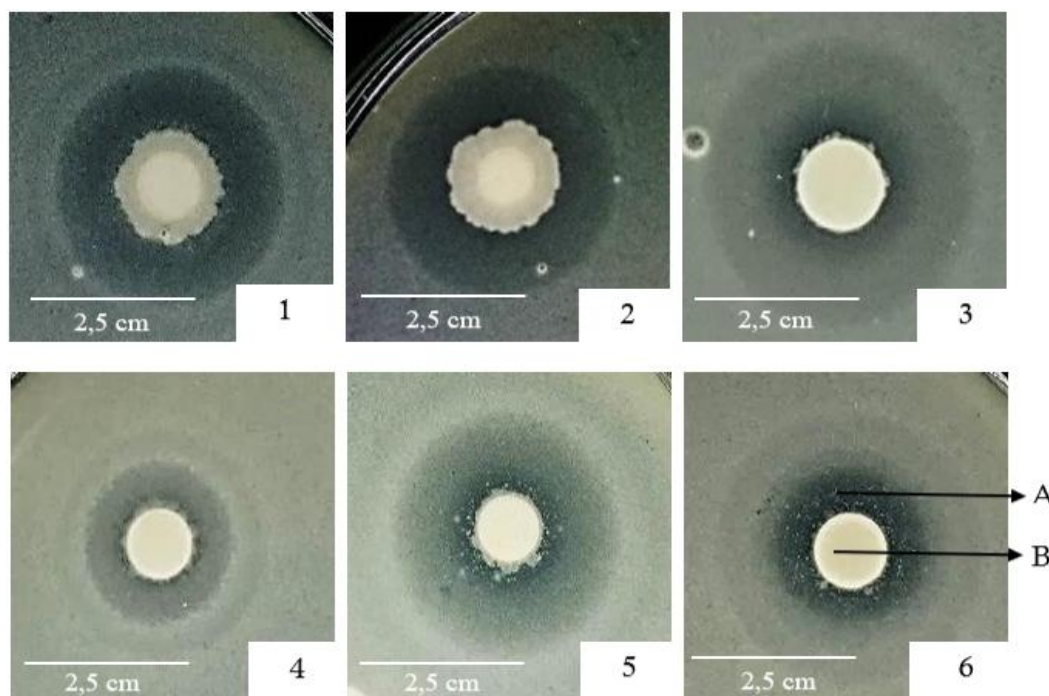
No	Isolate Code	Inhibition Zone Against <i>C. albicans</i> (mm)
1	SVP 1	8,08±0,45
2	SVP 6	8,53±0,10
3	SVP 15	8,58±0,23
4	TVL 6	11,58±0,10
5	TVL 11	10,98±0,48
6	TVL 13	8,40±0,10

SVP: *Stomopneustes variolaris* Pengubaiian; TVL: *Triploneustes ventricosus* Laguna

Based on the results obtained, the symbiotic bacterial isolates exhibited varying levels of potency in inhibiting *C. albicans*. The measurements of the inhibition zones were then compared with their Standard Deviation (SD) values. According to [Febriani \(2022\)](#), the higher the standard deviation, the greater the deviation of the data from the mean that indicating high data variability, and vice versa. When the inhibition zone produced by a bacterial isolate is large, it indicates that the bacteria are able to secrete the secondary metabolites in greater and more optimal amounts ([Alouw et al., 2022](#)). These compounds are produced when bacteria are in unfavorable conditions, prompting the cells to release bioactive anticandida substances to survive.

The formation of inhibition zones is influenced by several factors, including the production of antibiotics, hydrogen peroxide, lysozymes, siderophores, and protease enzymes. Differences in inhibition zones among isolates against *C. albicans*

indicate the diversity of secondary metabolites produced by each isolate. The diameter of the inhibition zone reflects the antagonistic properties of the gut symbiont bacteria, demonstrating their ability to produce bioactive compounds that suppress the growth of *C. albicans*. This occurs because the bioactive compounds can inhibit the production and secretion of extracellular enzymes by the test microorganism (Purniasih et al., 2022).



**Figure 4.** Inhibition zones produced by pellet of symbiotic bacterial isolates from *S. variolaris* and *T. Ventricosus*; 1= SVP 1; 2= SVP 6; 3= SVP 15; 4= TVL 6; 5= TVL 11; 6= TVL 13, the pellet were able to inhibit *C. albicans*; A= Inhibition zone, B= Paper disc.

The anticandida activity test using pellets of the symbiotic bacterial isolates from sea urchins showed that several isolates were able to inhibit the growth of the test fungus *C. albicans*. The strong anticandida activity showed by TVL 11 isolate, with an inhibition zone of 16.65 mm, while the medium anticandida activity was observed in SVP 15 isolate, with an inhibition zone of 5.50 mm. The inhibition exhibited by the symbiotic bacterial isolates against *C. albicans* represents anticandida activity, which can be grouped into three mechanistic classes based on their site of action: inhibition of ergosterol synthesis (the primary fungal sterol), physicochemical interaction with fungal membrane sterols, and inhibition of macromolecule synthesis. Secondary metabolites produced by marine bacteria exhibit antimicrobial properties, including anticandida activity, through mechanisms that inhibit growth and damage the cellular structure of pathogenic fungi, including *Candida albicans* (Wibowo et al., 2021). Various resistance mechanisms may develop, including alterations in drug targets, modifications in sterol biosynthesis, reduction of intracellular enzyme concentrations, and overexpression of anticandida drug target sites (Hossain et al., 2022).

Differences in growth among bacterial isolates arise from variations in their reproductive capabilities, which depend on growth media and nutritional availability.

Bacterial growth is also influenced by factors such as pH and temperature. Moreover, each bacterium's ability to adapt to its environment, divide, and survive contributes to differences in growth rates (Pamungkas et al., 2023). Growth rates can vary due to differences in enzymatic content that affect metabolic processes and the production of secondary metabolites. The production of bioactive compounds by bacteria is closely associated with their growth phase. During the logarithmic phase, bacteria actively divide and absorb large amounts of nutrients from the medium. When nutrient availability begins to decline, bacteria enter the stationary phase, in which the synthesis of bioactive compounds begins, including anticandida metabolites (Wibowo et al., 2020). The inhibition zones produced by bacterial pellet after 24 hours of incubation were measured using a digital caliper, and the results are presented in Table 3. The antibacterial activity of each isolate was classified according to the inhibition zone diameter categories proposed by Ouchari et al., (2019).

**Table 3.** Results of of Anticandida Activity Test using Bacterial Pellet

No	Isolate Code	Disc Diameter (mm)	Disc + Inhibition Zone Diameter (mm) ± SD	Active Inhibition Zone Diameter (mm) ± SD	Inhibition Category
1	SVP 1	6.00	14.25 ± 5.08	8.25 ± 0.05	Medium
2	SVP 6	6.00	21.50 ± 8.28	15.50 ± 0.10	Strong
3	SVP 15	6.00	11.50 ± 4.03	5.50 ± 0.10	Medium
4	TVL 6	6.00	21.40 ± 8.20	15.40 ± 0.20	Strong
5	TVL 11	6.00	22.65 ± 8.71	16.65 ± 0.45	Strong
6	TVL 13	6.00	19.58 ± 7.31	13.58 ± 0.32	Strong

Based on the test results, the anticandida activities of bacterial cultures and bacterial pellets showed differences in their ability to inhibit the growth of *C. albicans* among the isolates. Most isolates produced larger inhibition zone diameters when tested using bacterial pellets compared to bacterial cultures. Isolate TVL 11 showed an increase in inhibition zone diameter from 10.98 mm in the culture assay to 16.65 mm in the pellet assay, whereas isolate SVP 6 increased from 8.53 mm to 15.50 mm. These findings indicate that bacterial pellets exhibited higher anticandida activity than bacterial cultures in most of the isolates tested.

According to Sunny (2016), bioactive compounds produced by bacteria may be present in the culture medium as extracellular metabolites or may accumulate within or on the surface of bacterial cells as intracellular metabolites associated with bacterial biomass. Therefore, centrifugation was performed to separate the bacterial cells from the culture medium, resulting in a pellet containing a higher concentration of bacterial biomass. The greater anticandida activity observed in the bacterial pellets suggests that some of the bioactive compounds responsible for inhibiting the growth of *C. albicans* may accumulate within the bacterial biomass and are not entirely released into the culture medium.

Anticandida compounds are synthesized as a response to the interaction between symbiotic bacterial isolates and the test fungus. The presence of the test fungus acts as a competitor for space and nutrients, prompting the symbiotic bacteria to synthesize anticandida compounds to inhibit the fungal growth. The anticandida

compounds produced by the symbiotic bacteria are secondary metabolites. The formation of secondary metabolites is influenced by several factors, including nutrient availability, decreased growth rate, and enzyme inactivation. Secondary metabolites are typically synthesized when bacteria are in a stressed condition (Judianti et al., 2015).

Furthermore, differences in inhibitory activity between bacterial cultures and bacterial pellets may also be influenced by variations in the distribution of anticandida metabolites produced by each isolate. These findings indicate that the production and distribution characteristics of anticandida metabolites can vary among gut symbiotic bacterial isolates from sea urchins, thereby affecting the level of anticandida activity exhibited by each isolate.

## CONCLUSION

The pellet method exhibited higher anticandida activity than the culture method in most of the tested symbiotic bacterial isolates. All symbiotic bacterial isolates obtained from the digestive tracts of *S. variolaris* and *T. ventricosus* collected from Kaur Regency, Bengkulu, were able to inhibit the growth of *C. albicans*, as indicated by the formation of inhibition zones in both culture and pellet based assays. Isolate TVL 6 exhibited the highest anticandida activity in the culture assay, with an inhibition zone diameter of 11.58 mm, whereas isolate TVL 11 showed the highest activity in the pellet assay, with an inhibition zone diameter of 16.65 mm. These findings suggest that gut symbiotic bacteria of sea urchins represent a potential source of anticandida bioactive compounds, with the active metabolites likely being not only secreted into the culture medium but also accumulated within the bacterial biomass. This study contributes to the growing knowledge of sea urchin gut symbiotic bacteria as a potential source of marine derived anticandida agents, particularly from the coastal region of Kaur Regency, Bengkulu, Indonesia. The findings provide a scientific basis for future studies focusing on the identification of promising bacterial isolates, characterization of their bioactive metabolites, and investigation of their anticandida mechanisms of action, thereby supporting the development of alternative anticandida agents. Nevertheless, this study was limited to the evaluation of anticandida activity using the inhibition-zone assay and did not identify the specific bioactive compounds responsible for the observed activity or their mechanisms of action. Therefore, further studies involving molecular identification of the bacterial isolates, characterization of secondary metabolites, and determination of Minimum Inhibitory Concentration (MIC) and Minimum Fungicidal Concentration (MFC) values are required to validate and further explore the anticandida potential of these isolates.

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