ANALISIS PENGHAMBATAN N-PHOSPHORYL-L-LEUCINAMIDE PADA PROTEASE NETRAL (NPr) TERMOSTABIL DARI *Geobacillus* sp. DS3 SECARA *IN SILICO*

INTISARI

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Protease adalah enzim yang dapat menghidrolisis ikatan peptida pada molekul protein. Dalam industri pangan, protease banyak berperan dalam pembuatan keju, produk susu, pengempukan daging, dan meningkatkan nilai gizi roti. *Geobacillus* sp. DS3 yang diisolasi dari Kawah Sikidang Dieng dapat menghasilkan enzim protease yang memiliki aktivitas optimum pada suhu 70^{o}C dan pH 9,6. Pada penelitian sebelumnya telah dilakukan pemodelan struktur 3D protease netral (NPr) dari *Geobacillus* sp. DS3 dan analisis penghambatan enzim terhadap inhibitor phosphoramidon. Tujuan penelitian ini adalah melakukan analisis penghambatan oleh inhibitor N-phosphoryl-L-leucinamide secara *in silico* untuk mengetahui mekanisme dan tipe penghambatan N-phosphoryl-L-leucinamide, serta prediksi residu pada sisi aktif NPr *Geobacillus* sp. DS3. Bahan penelitian dipreparasi menggunakan Autodock Tools, *molecular docking* dilakukan menggunakan program Autodock Vina, serta divisualisasikan menggunakan Discovery Studio Visualizer dan PyMOL. Terdapat interaksi antara ion zink pada sisi aktif NPr dengan inhibitor sehingga terjadi penghambatan aktivitas enzim. Beberapa residu katalitik seperti Glu139 dan His227, serta *binding residue* seperti Arg199 dan Asn108 juga dihambat oleh N-phosphoryl-L-leucinamide. Hasil analisis menunjukkan bahwa N-phosphoryl-L-leucinamide diduga merupakan inhibitor kompetitif. Akan tetapi, perlu diteliti lebih lanjut terkait penghambatan ini dengan melakukan *X-ray crystallography*.

Kata Kunci: **protease netral termostabil (NPr), *Geobacillus* sp. DS3, *molecular docking*, inhibitor, N-phosphoryl-L-leucinamide**

IN SILICO INHIBITION ANALYSIS OF N-PHOSPHORYL-L-LEUCINAMIDE ON THERMOSTABLE NEUTRAL PROTEASE (NPr) FROM *Geobacillus* sp. DS3

ABSTRACT

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Protease is an enzyme which can hydrolyze peptide bonds in protein molecules. In the food industry, proteases are used in the manufacture of cheese, dairy products, tenderizing meat, and increasing the nutritional value of bread. *Geobacillus* sp. DS3 from Sikidang Crater Dieng Plateau could produce protease enzymes which had optimum activity at 70^{o}C and pH 9.6. Previous study has carried out neutral protease (NPr) three-dimensional structure and analysis of enzyme inhibition against phosphoramidone inhibitors. In this study, NPr inhibition by N-phosphoryl-L-leucinamide has been done by molecular docking method. The purpose of this study was to determine the mechanism and type of N-phosphoryl-L-leucinamide inhibition, and predict the residue on the active site of NPr *Geobacillus* sp. DS3. Materials were prepared using Autodock Tools, molecular docking was performed using the Autodock Vina program, and the result were visualized using Discovery Studio Visualizer and PyMOL. His138, His142, and Glu162 were likely zinc binding residue of NPr. There was an interaction between Zn^{2+} on the active site and inhibitors resulting in inhibition of enzyme activity. Besides, several catalytic residues such as Glu139 and His227, also binding residues on the active site such as Arg199 and Asn108 were inhibited by N-phosphoryl-L-leucinamide. The results showed that N-phosphoryl-L-leucinamide was predicted to be a competitive inhibitor. However, further research is needed regarding this inhibition by performing X-ray crystallography.

Keywords: **Thermostable neutral protease (NPr), *Geobacillus* sp. DS3, molecular docking, inhibitor, N-phosphoryl-L-leucinamide**