Molecular and immunological characteristics of Candida Secreted Aspartyl Proteinase 5

Alfin Titian Permata1,2, Afiono Agung Prasetyo2,3*, Ruben Dharmawan 2

1 Bioscience Department of Post Graduate Program, Universitas Sebelas Maret, Jl. Ir. Sutami 36A, Surakarta 57126, Indonesia
2 A-IGIC (A-Infection, Genomic, Immunology & Cancer) Research Group, Sumber, Banjarsari, Surakarta, 57138, Indonesia
3 Division of Virology, Department of Microbiology, Faculty of Medicine, Universitas Sebelas Maret, Jl. Ir. Sutami 36A, Surakarta, 57126, Indonesia

E-mail: alfintitian@gmail.com

Abstract. *Candida* sp. is the most common of yeasts causes fungal infections in human. One virulence factor of *Candida* sp. is the Secreted Aspartyl Proteinase (SAP) 5. However, the molecular and immunological characteristics of the Candida Secreted Aspartyl Proteinase 5 is still limited known. To study the molecular and immunological characteristics of the Candida Secreted Aspartyl Proteinase 5, all complete coding sequences of *Candida Secreted Aspartyl Proteinase 5* deposited in GenBank were retrieved and subjected to bioinformatic analysis. The sequences alignment was used to predict the evolutionary relationships among various *Candida* sp. based on the *Secreted Aspartyl Proteinase* 5 gene and to find out any possible genomic and proteomic variations. The molecular weight (Mw), signal peptide, protein charge, antigenicity, hydrophobicity, estimated half-life, and secondary structure of the predicted proteins also had been discussed. The results from this study would contribute information of molecular and immunological characteristics of the Candida Secreted Aspartyl Proteinase 5.

1. Introduction

*Candida* sp is a microflora found in an organism, almost 50% found in the body microflora. *Candida* sp can be a pathogen when the host is impaired in the immune system, example *Candida albican*. *Candida albicans* is an opportunistic pathogen of humans with the ability to switch between the yeast and filamentous growth forms [1] [2][3].

Secreted aspartic proteases (SAPs) are proteins produced by *Candida albican*. This protein has a role in virulence factor and has a specific role in infection. Secreted aspartic proteases (SAPs) produced by *Candida Albicans* have ten family, one of which is secreted aspartyl proteinase 5 (sap5)[4]. The different members of the SAP family of the human pathogenic yeast *Candida albicans* are proposed to play different roles during infection. Secreted aspartic proteinase 5 (SAP5) is one of the important virulence factors during *Candida albicans* mucosal or disseminated infections[5]. However, limited data is known about the protein, information of molecular and immunological characteristics of the *Candida* Secreted Aspartyl Proteinase 5.
2. Experimental Methods

To study the molecular and immunological characteristics of the Candida Secreted Aspartyl Proteinase 5, all complete coding sequences of Candida Secreted Aspartyl Proteinase 5 deposited in GenBank were retrieved and subjected to bioinformatic analysis. Multiple alignments of reference sequences were reconstructed using ClustalW as implemented in MEGA 6 software. Protein analysis was performed using CLC Main Workbench 8.0 software. The parameters for hydrophobicity plot was set by Kyte-Doolittle, Eisenberg, Hopp-Woods, Janin, Rose, respectively, with a number of residues - must be odd as 11. The blastp protein sequence analysis was performed using non-redundant protein sequences (nr) database with standard database genetic code. To create the antigenicity plot, both Welling and Kolaskar-Tongaonkar antigenic scale was used. The T Cell Epitope Prediction Tools was used to predict the Major histocompatibility complex-binding and immunogenicity prediction.

3. Results and Discussion

There were only three secreted aspartyl proteinase 5 (sap 5) Candida complete coding sequence in gene bank, accession number XP_719147.1, orf19.5585 and Z30191.1, and one sequences identity to secreted aspartyl proteinase 5 (sap 5) NC_032094.1. The sequence was confirmed as Candida albicans SAP5. There were have conserved nucleotide and amino acid at position 268-320 and 322-1524 (1611 nucleotide), 17 and 401 (418 amino acid) respectively. Based on genotyping and phylogenetic analysis of sap 5 gene, sequence on number orf 19.5585 closely related to Z30191.1, XP_719147.1, NC_032094.1 (Figure 1).

![Phylogenetic analysis](image)

**Figure 1.** Phylogenetic of analysis based on secreted aspartyl proteinase 5 (sap 5) from deposit genebank, accession number orf 19.5585, XP_719147.1, Z30191.1 and NC_032094.

The predicted protein had 418 amino acid sequence length, pH was 5. Beta strand secondary structures were frequent in our predicted SAP5 (75.67%,28 /37). Only seven alpha helix secondary structures were found, at aa (amino acids) 134-136, 143-145, 203-205, 217-223, 304-314, 352-355 and 384-387, respectively. We performed the immunogenicity prediction and major histocompatibility complex-binding for the Candida albicans SAP5 using the T Cell Epitope Prediction Tools from Immune Epitope Database (IEDB) Analysis Resource. In total, 14 epitopes were found had a high affinity (percentile rank 0.01) for Major histocompatibility complex II-binding (Table 1), consistent with the antigenicity plot (Figure 2).

**Table 1.** Major histocompatibility complex II-binding and immunogenicity prediction of Candida SAP5

| Allele                | Start | End  | Peptide          |
|-----------------------|-------|------|------------------|
| HLA-DPA1*01:03/DPB1*02:01 | 1     | 15   | MFLKNILSVLAFALL |
| HLA-DPA1*01:03/DPB1*02:01 | 2     | 16   | FLKNILSVLAFALLI |
| HLA-DPA1*01:03/DPB1*02:01 | 3     | 17   | LKNILSVLAFALLID |
| HLA-DPA1*01:03/DPB1*02:01 | 4     | 18   | KNILSVLAFALLIDA |
| HLA-DPA1*01:03/DPB1*02:01 | 1     | 15   | MFLKNILSVLAFALL |
| HLA-DPA1*01:03/DPB1*02:01 | 2     | 16   | FLKNILSVLAFALLI |
| HLA-DPA1*01:03/DPB1*02:01 | 3     | 17   | LKNILSVLAFALLID |
| HLA-DPA1*01:03/DPB1*02:01 | 4     | 18   | KNILSVLAFALLIDA |
Secreted aspartic proteases (SAP5) involved in the adhesion, invasion and tissue destruction by *Candida albicans*, also expressed in the biofilms produced by the fungal Secreted aspartic proteases (SAPs) 5 was expressed when after infection and its association with fungi morphological changes[6][7]. Changes in the morphology of the fungus to the hypa will damage the epithelial layer, and sap 5 will be expressed by biofilm formation [1][8]. Sap5 plays a role in cell colonization, penetration and mucosal infection found in *Candida albicans* [9]. In candidemia patients, vulvovaginal infections, and teeth caries child, sap5 is found in the biofilm layer [3]. Since SAP5 is one of the key virulence factors that play a central role in the pathogenesis of *Candida albicans*, therefore SAP5 is a nice target for designing potent antifungal agents[10][11][12]. In the present study, we found the amino acids at position 1 until 18 had a high affinity for Major histocompatibility complex II-binding with high antigenic property, therefore, may be potential as a target to improving diagnostic, therapeutic, or vaccine efficacy.

In conclusions, The present study will contribute information about our *Candida* SAP5 and benefits for further works willing to develop diagnostic and therapeutic strategies against the *Candida sp*.

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5. References
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