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## Cloning of dengue virus type 3 (Indonesian strain D3-1703) non structural-1 gene into pYES2/CT vector

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Abstrak

## <b>ABSTRACT</b><br>

Dengue is an infectious disease caused by dengue virus. Dengue endemic region includes America, Western Pacific.

Africa, East Mediterranian, and South East Asia including Indonesia. An early diagnostic system specific for Indonesia

is needed to control dengue in Indonesia. In this research, cloning of Non Structural 1 (NS1) gene from dengue virus

type 3 (Indonesian strain D3-1703) into pYES2/CT vector was performed. In the long run, NS1 recombinant protein

will be expressed in

Saccharomyces cerevisiae

for diagnostic materials. Polymerase Chain Reaction (PCR)

amplification of NS1 gene fragments were done with optimal annealing temperature at  $55\,^{\circ}$ C. NS1 gene fragment and

pYES2/CT were cut by Bam H I and Not I enzymes. The digested pYES2/CT was dephosphosrylated using Calf Intestine Alkaline Phospatase enzyme. Ligation with the vector:insert

ratio of 1:12 and 1:20 resulted in 6 and 5 recombinant colony candidates respectively. Restriction enzyme and PCR verifications showed that 5 recombinant plasmids contained NS1 gene. Sequencing of the first 600 bp of one recombinant plasmid was performed. The blastn

analysis showed that it had a 99% identity with dengue virus type 3 strain FW06. Finally, it was shown that NS1 clone

within pYES2/CT was in the correct Open Reading Frame and ready to be expressed in S. cerevisiae.