

Isolasi Gen *pall* yang Mengkode Sukrosa Isomerase dari Bakteri Isolat T1j

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Abstrak

Sukrosa, yang digunakan secara luas sebagai pemanis makanan dan minuman memiliki dampak negatif bagi kesehatan, seperti menyebabkan kerusakan gigi dan berbahaya bagi penderita penyakit diabetes. Berbeda dengan sukrosa, isomaltulosa yang merupakan isomer dari sukrosa bersifat rendah kalori, non-kariogenik, non-karsinogenik, dan aman bagi penderita diabetes, sehingga sangat ideal digunakan sebagai gula pengganti sukrosa, meskipun kadar kemanisannya hanya setengah dari sukrosa. Beberapa spesies bakteri telah diketahui memiliki gen *pall*, yaitu gen yang mengkode enzim sukrosa isomerase pengonversi sukrosa menjadi isomaltulosa. Bakteri isolat T1j diisolasi dari batang tebu pabrik gula Djatiroto. Bakteri ini diketahui memiliki gen *pall* karena menunjukkan hasil positif ketika dihibridisasi dengan gen *pall* dari bakteri *Erwinia rhapontici*. Penelitian ini bertujuan mengisolasi gen *pall* dari bakteri isolat T1j, dan memperbanyak gen tersebut dalam vektor plasmid pGEM[®]-T Easy. Isolasi gen *pall* dari bakteri isolat T1j dengan menggunakan metode *nested PCR* menghasilkan fragmen berukuran 1211 bp. Fragmen DNA tersebut telah berhasil diklon ke dalam vektor plasmid pGEM[®]-T Easy. Analisis sekuensing yang dilakukan terhadap fragmen DNA yang diisolasi dari bakteri isolat T1j menunjukkan kemiripan urutan nukleotida sebesar 97% dengan data sekuen nukleotida gen *pall* *Erwinia rhapontici* yang terdapat di GenBank dengan nomor akses AY223550. Hal tersebut membuktikan bahwa fragmen DNA berukuran 1211 bp yang diklon ke dalam plasmid pGEM[®]-T Easy merupakan fragmen gen *pall*. Dilakukan juga analisis sekuensing terhadap fragmen-fragmen lain yang teramplifikasi dari bakteri isolat T1j ini untuk memprediksi identitas bakteri tersebut. Dari hasil sekuen fragmen-fragmen tersebut, dapat disimpulkan sementara bahwa bakteri T1j kemungkinan besar berasal dari famili yang sama dengan *Erwinia rhapontici*, yaitu Enterobacteriaceae.

Kata kunci: Isomaltulosa, Gen *pall*, Sukrosa isomerase.

Cloning *pall* Gene Encoding Sucrose Isomerase from Bacterial Isolate T1j

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Abstract

Sucrose, which is widely used in foods and drinks, has many disadvantages such as causing tooth decay and not suitable for diabetic patients. On the other hand isomaltulose, which is an isomer of sucrose has different properties than sucrose: low calorie, noncariogenic, noncarcinogenic, and will not increase the blood glucose significantly in diabetic patient, although the sweetness of isomaltulose is only half compared with sucrose. Several bacteria are known to have *pall* gene, the gene encoding sucrose isomerase enzyme that converts sucrose into its isomers (including isomaltulose). T1j bacteria, which is isolated from sugarcane waste product from Djatiroto sugar factory, has *pall* gene because it gave positive result when hybridized with *pall* gene from *Erwinia rhapontici*. The aim of this research is to isolate the *pall* gene from isolate T1j bacteria and clone the gene into pGEM[®]-T Easy vector plasmid. The *pall* was cloned from isolate T1j bacteria by nested PCR and the DNA fragment was cloned successfully into pGEM[®]-T Easy vector plasmid. The length of DNA fragment confirmed by sequencing is 1211 bp. Sequence analysis of the DNA fragment has 97% similarity to the *pall* gene from *Erwinia rhapontici* (accession number AY223550). It can be concluded that the 1211 bp DNA fragment which has been successfully cloned into pGEM[®]-T Easy was *pall* gene. Sequence analysis was also done to other DNA fragments that was amplified from isolate T1j bacteria to predict the identity of the bacteria. Based on those other sequences, the T1j bacteria is from the same family as *Erwinia rhapontici*, called Enterobacteriaceae.

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