P12

Cloning and characterization of a gene for fibrinolytic enzyme from *Bacillus subtilis* BB-1 isolated from black bean *chung-kuk*.

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Five fibrinolytic isozymes producing bacterium was isolated from black bean chung kuk. The bacterium was identified to *Bacillus subtilis* BB-1 by 16s rDNA sequencing and sequence homology search.

One gene of five fibrinolytic genes from the *Bacillus subtilis* BB-1 was cloned by shot-gun method.

A *Cla 1* fragment of chromosomal DNA of *B. subtilis* BB-1 was cloned to pBluescript II SK(-) and showed the fibrinolytic activity in bacterial cells. The *Cla 1* fragment was sequenced by autometic DNA sequencer and the sequence homology was not detected in any protease or fibrinolytic enzyme of the other organisms.

The Cla 1 fragment was reduced to 2577 bp by activity-guided PCR cloning method. The optimum pH and temperature of the enzyme are 5.0 and 35°C, respectively.

Substrate specificity of the fibrinolytic enzyme was detected in skim milk, casein, gelatin and blood agar plates. Any activity of the enzyme was not detected in these substrates. Taken together, this enzyme is a new fibrinolytic enzyme and may be used to prevent thrombosis and arteriosclerosis.