



## ABSTRAK

Koro pedang putih (*Canavalia ensiformis* (L.) D.C.) merupakan legum yang kaya akan protein. Pada fermentasi tempe koro pedang putih, jamur tempe menghasilkan protease yang dapat menghidrolisis protein menjadi peptida-peptida dan asam amino. Tujuan dari penelitian ini adalah: 1) mempelajari potensi koro pedang putih sebagai sumber peptida inhibitor ACE melalui fermentasi tempe; 2) mempelajari degradasi peptida hasil fermentasi tempe oleh enzim-enzim pencernaan secara *in vitro* dan pengaruhnya terhadap aktivitas penghambatan ACE; dan 3) melakukan uji penyerapan peptida menggunakan kantong usus terbalik yang dibagi menjadi tiga segmen usus (duodenum, jejunum, dan ileum) dan mengkarakterisasi peptida yang terserap (aktivitas penghambatan ACE, pola penghambatan ACE, berat molekul dan sekuen asam amino). Penelitian ini meliputi fermentasi tempe koro pedang putih untuk menghasilkan peptida yang memiliki aktivitas penghambatan ACE, hidrolisis peptida tempe menggunakan enzim pepsin selama 120 menit dilanjutkan dengan pankreatin selama 120 menit, dan pengujian penyerapan peptida tempe menggunakan metode kantong usus terbalik. Hasil penelitian menunjukkan bahwa fermentasi tempe mampu meningkatkan aktivitas penghambatan ACE. Peningkatannya selama fermentasi berbanding lurus dengan peningkatan aktivitas proteolitik dan derajat hidrolisis hingga tempe difermentasi selama 72 jam, namun setelah itu derajat hidrolisis yang tinggi tidak berkorelasi positif dengan aktivitas penghambatan ACE. Aktivitas penghambatan ACE tertinggi diperoleh pada tempe yang difermentasi selama 72 jam dengan aktivitas penghambatan sebesar 60,04% ( $IC_{50}=1,03$  mg/mL). Simulasi pencernaan *in vitro* menggunakan enzim pepsin dilanjutkan pankreatin menunjukkan bahwa koro pedang rebus, tempe 48 jam, dan 72 jam memiliki derajat hidrolisis yang tinggi yaitu masing-masing 67,90; 74,52; dan 68,04 %. Peptida yang dihasilkan selama simulasi pencernaan *in vitro* mengalami peningkatan aktivitas penghambatan ACE yang signifikan. Aktivitas penghambatan ACE tertinggi diperoleh pada hidrolisat tempe 72 jam dengan penghambatan sebesar 88,20 % ( $IC_{50} = 0,60$  mg/mL). Uji penyerapan menggunakan kantong usus terbalik menunjukkan bahwa peptida tempe 72 jam memiliki persentase penyerapan peptida yang lebih tinggi daripada koro pedang rebus, dengan penyerapan tertinggi di jejunum. Peptida tempe yang terserap tersusun atas tujuh peptida (DLGKAPIN, GKGRFVYG, PFMRWR, DKDHAEI, LAHLYEPS, KIKHPEVK, dan LLRDTCK), memiliki aktivitas penghambatan ACE sebesar 80,60 %, BM. 826-978 Da, bersifat sebagai *pro-drug inhibitor* dan memiliki pola penghambatan campuran.

Kata kunci: ACE, peptida, tempe, koro pedang putih, fermentasi, simulasi pencernaan *in vitro*



## ABSTRACT

Jack bean (*Canavalia ensiformis* (L.) D.C.) is a protein-rich legume, which the composition of hydrophobic amino acids and positively charged amino acids of the seeds achieved of 43.5% and 12.4%. Fermentation of jack bean tempe involved the mold, produced proteases that can hydrolyze proteins into peptides and amino acids. The objectives of this research were: 1) to investigate the potential of jack bean as a source of ACE inhibitory peptide through tempe fermentation; 2) to investigate the degradation of peptides resulting from tempe fermentation by digestive enzymes *in vitro* and their effect on ACE inhibitory activity; and 3) to determine peptide absorption and characterize peptides absorbed by the small intestine. The research included the fermentation of jack bean tempe to produce peptides with the ACE inhibitory activities, hydrolysis of resulting peptide from the fermentation using pepsin pancreatin, sequentially, and determination of peptides absorption using the method of everted intestinal sacs. The results demonstrated that jack bean possessed the ability to produce ACE inhibitory peptide during tempe fermentation. Even, the boiled jack bean (unfermented) already had ACE inhibitory activity, with the value of 13.89%. Tempe fermentation was found to improve the ACE inhibitory activity of resulting peptides. Their increases were in line with the increase of proteolytic activity and degree of hydrolysis until 72 h of tempe fermentation, but after that, a high degree of hydrolysis did not positively correlate with ACE inhibitory activity. The highest ACE inhibitory activity was obtained in tempe fermented for 72 h with a value of 60.04% ( $IC_{50} = 1.03$  mg / mL). *In vitro* digestion simulation using pepsin pancreatin demonstrated that boiled jack bean, tempe 48 h, and 72 h possessed a high degree of hydrolysis, with a value of 67.90; 74.52; and 68.04%, respectively. The produced peptides during *in vitro* digestion simulation exhibited a significant increase of ACE inhibitory activity. The highest ACE inhibitory activity was obtained at 72 h tempe hydrolyzates (HF72) with the inhibition of 88.20 % ( $IC_{50} = 0.60$  mg / mL). The most significant increase was found in boiled jack bean hydrolyzates (HF0) with an increase of 13.89 to 75.10%. The hydrolysis of jack bean protein using pepsin pancreatin sequentially effectively generated the peptides with high inhibitory activity. The absorption experiments using everted small intestinal sacs demonstrated that the tempe, which fermented for 72 h, possessed a higher percentage of peptide absorption than boiled jack bean. The absorbed peptides were composed of seven peptides (DLGKAPIN, GKGRFVYVG, PFMRWR, DKDHAEI, LAHLYEPS, KIKHPEVK, and LLRDTCK), with ACE inhibition of 80.60 %, MW of 826-978 Da, classified as a pro-drug inhibitor with the mixed inhibition pattern.

Keywords: ACE, peptides, tempe, jack bean, fermentation, *in vitro* digestion simulation