

ABSTRAK

Susu sapi merupakan susu dengan kandungan protein tinggi sehingga berpotensi sebagai sumber penghasil peptida bioaktif seperti antioksidan. Peptida bioaktif susu sapi dapat diperoleh melalui hidrolisis enzimatis terhadap protein susu menggunakan enzim protease yang berasal dari mikroorganisme seperti *Bacillus subtilis* B211. Tujuan penelitian ini untuk mengetahui potensi enzim protease isolat *B. subtilis* B211 dalam menghidrolisis protein susu sapi berupa kasein dan whey untuk menghasilkan protein hidrolisat dengan aktivitas antioksidan. Penelitian ini, dilakukan produksi dan fraksinasi enzim protease isolat *B. subtilis* B211 menggunakan garam amonium sulfat tingkat kejemuhan 0-70% untuk mendapatkan enzim yang lebih murni serta karakterisasi enzim meliputi pengaruh suhu dan pH. Hidrolisis kasein dan whey dilakukan pada kondisi optimum enzim dengan lama waktu inkubasi 10, 20, 30, 40, 50, dan 60 menit. Aktivitas antioksidan protein hidrolisat ditentukan dengan metode DPPH dan dilakukan uji hemolisis. Hasil penelitian menunjukkan bahwa aktivitas spesifik enzim protease F70 hasil fraksinasi sebesar 0,0082 U/mg dengan faktor purifikasi 0,371 kali serta optimum bekerja pada suhu 40 °C dan pH kisaran 7 – 8. Hidrolisis tertinggi diperoleh pada waktu inkubasi 60 menit dengan nilai derajat hidrolisis kasein sebesar 82,62% dan whey sebesar 28,0%. Persentase inhibisi tertinggi terhadap radikal DPPH ditunjukkan oleh protein hidrolisat kasein dan whey yang diinkubasi selama 10 menit sebesar 76% dan 82%. Berdasarkan nilai AAI, aktivitas antioksidan kasein dan whey hidrolisat masih tergolong antioksidan lemah karena nilai AAI < 0,5 yaitu sebesar $8,7 \times 10^{-4}$ dan $7,2 \times 10^{-4}$. Hasil uji hemolisis menunjukkan bahwa protein hidrolisat kasein dan whey menyebabkan lisis terhadap sel darah merah dengan persentase sebesar 8% dan 4%.

Kata kunci: antioksidan, *B. subtilis*, enzim protease, fraksinasi, protein hidrolisat

ABSTRACT

Cow's milk is milk with high protein content so it has the potential as a source of peptides producing various bioactive properties such as antioxidants. Cow's milk bioactive peptides can be obtained through enzymatic protein hydrolysis of milk proteins using protease enzymes derived from microorganisms such as *Bacillus subtilis* B211. The purpose of this study was to determine the potential of the protease enzyme isolate *B. subtilis* B211 to hydrolyzing cow's milk protein in the form of casein and whey to produce hydrolyzed protein with antioxidant activity. The production and fractionation of the protease enzyme isolate *B. subtilis* B211 was carried out using ammonium sulfate salt with a saturation level of 0-70% to obtain a purer enzyme as well as the characterization of the enzyme including the effect of temperature and pH. Hydrolysis of casein and whey was carried out under optimum enzyme conditions with incubation times of 10, 20, 30, 40, 50, and 60 minutes. The antioxidant activity of protein hydrolysate was determined by DPPH method and hemolysis test was performed. The results showed that the specific activity of the fractionated protease F70 enzyme was 0.0082 U/mg with a purification factor of 0.371 times and the optimum worked at a temperature of 40 °C and pH range 7 – 8. The highest hydrolysis was obtained at an incubation time of 60 minutes with a casein hydrolysis degree of 82.62% and whey by 28.0%. The percentage of inhibitory of DPPH radicals was shown by casein and whey protein hydrolysate which were incubated for 10 minutes by 76% dan 82%.. Based on the AAI value, the antioxidant activity of casein and whey hydrolysate were classified as weak antioxidants because the AAI value was < 0.5, namely 8.7×10^{-4} and 7.2×10^{-4} . The hemolysis percentage showed that casein and whey hydrolysate caused lysis of red blood cells with a percentage of 8% and 4%.

Keywords: antioxidant, *B. subtilis*, fractionation, hydrolysate proteins, protease enzyme