Jurnal Ilmu Pertanian Indonesia (JIPI), October 2025
(Indonesian Journal of Agricultural Sciences)
ISSN 0853-4217 EISSN 2443-3462

Vol. 30 (4): 696–702
http://journal.ipb.ac.id/index.php/JIPI
DOI: 10.18343/jipi.30.4.696

The Potential of Bioactive Peptides from Trypsin-Hydrolyzed Egg White of IPB-D1 Chicken as Antioxidant and Antibacterial

Nova Aryanti, Zakiah Wulandari*, Cahyo Budiman

(Received December 2024/Accepted July 2025)

ABSTRACT

The IPB-D1 chicken is the outcome of crossbreeding native Indonesian chickens and can produce eggs high in protein, which is excellent for bioactive peptide manufacturing. Egg white hydrolysates contain bioactive peptides with antioxidant and antibacterial properties. The purpose of this work was to assess the antioxidant and antibacterial properties of bioactive peptides produced from IPB-D1 egg white hydrolyzed with trypsin. The research procedures included determining the Haugh unit (HU) value, protein content, degree of hydrolysis (DH), and antioxidant and antibacterial activity. The hydrolysate inhibited DPPH by 44.82% and had an antioxidant capacity of 23.16 mg EVC 100/g. The hydrolysate had greater inhibition zones against Gram-positive and Gram-negative bacteria than the non-hydrolyzed egg white, indicating its antibacterial action. Inhibition zone sizes ranged from 2.42 to 3.48 mm. The findings show that enzymatic hydrolysis of IPB-D1 egg white with trypsin significantly improves its antioxidant and antibacterial properties.

Keywords: antibacterial, antioxidant, bioactive peptide, IPB-D1 chicken, trypsin enzyme

INTRODUCTION

Eggs are known as a low-cost and conveniently available source of animal protein for the Indonesian population. Eggs include essential elements such as protein, vitamins, minerals, and fat (Purwati *et al.* 2015). The nutritious value of eggs supports their use in daily life, yet consumption of local chicken eggs is primarily limited to meeting dietary needs for animal protein. According to the Ministry of Agriculture (2022), Indonesians consumed 87.18% of commercial chicken eggs in 2021, while native or local chicken eggs were consumed at only 5.58%. This demonstrates a major opportunity for creating products from native hens as a viable source of table eggs. Apart from meeting nutritional requirements, eggs provide functional benefits as a source of bioactive compounds that are helpful to human health. Egg white is the most utilized component of functional meals. In general, egg white accounts for 58−60% of the weight of a whole egg (Agustina *et al*. 2013) and contains approximately 11% protein, with the primary components being ovalbumin, ovomucoid, ovotransferrin, ovomucin, and lysozyme (Ismoyowati 2020). Egg white's high protein content makes it a viable raw material for the manufacture of bioactive peptides with a wide range of actions, including antioxidants, antibacterials, and antihypertensives (Liao *et al.* 2018).

Antioxidants and antibacterial chemicals are significant in both the health and food industries. Antioxidants are commonly used as supplements to boost the immune system and lower the risk of diseases caused by free radicals, including cancer, diabetes, premature aging, and heart disease. Furthermore, these two chemicals are used as food preservatives to prevent fat oxidation and slow the growth of bacteria that cause food deterioration (Aditia *et al.* 2018). Several studies have indicated that hydrolysate of broiler and native chicken egg whites can be exploited as a source of bioactive peptides with antioxidant (Dewi 2023) and antibacterial activity (Suparmono 2019). Eggs from native Indonesian hens, such as IPB-D1 birds, have the ability to provide protein for the production of bioactive peptides that are antioxidants and antibacterials. The D1 chickens are a local chicken strain derived from a cross between *pelung*, *sentul*, *kampung*, and commercial broiler chickens, produced and legally acknowledged by the Republic of Indonesia's Ministry of Agriculture (Habib *et al*. 2020). This bird has the potential to produce extremely productive eggs because of its 25% contribution of commercial broiler chicken genetics (Habiburahman *et al*. 2020). As a freshly produced indigenous chicken, the use and marketing of IPB-D1 chicken products are limited. Therefore, it is necessary to investigate the potential of IPB-D1 egg white. as a source of bioactive peptides.

Protein hydrolysis can produce bioactive peptides; the hydrolysis process can be aided by the presence of

Department of Animal Production Science and Technology, Faculty of Animal, IPB University, IPB Darmaga Campus, Bogor 16880
* Corresponding Author:
Email: zakiahwu@apps.ipb.ac.id

enzymes (enzymatic hydrolysis) (Agyei *et al*. 2016). The enzyme selection is critical, as the effectiveness of the resultant peptides is heavily impacted by the protein supply and enzyme specificity (Kusumaningtyas *et al.* 2015). The hydrolysis process has been carried out using a variety of proteases, one of which is trypsin, which is particular for severing peptide bonds on the carboxyl side of L-arginine and L-lysine residues (Khirzin *et al*. 2020).Trypsin is widely used in many industries because to its stability in NaCl concentrations and capacity to work in harsh settings (Rahmawati 2023). In this investigation, the trypsin enzyme was employed to hydrolyze IPB-D1 chicken egg white protein. The hydrolysate is projected to exhibit strong antioxidant and antibacterial activity, paving the way for the future use of bioactive peptides from IPB-D1 chicken egg white as functional foods. The purpose of this work is to assess the potential antioxidant and antibacterial activities of bioactive peptides obtained by hydrolyzing IPB-D1 chicken egg white with trypsin.

METHODS

Place and Time of Research

The research was done from December 2023 to March 2024 at the Microbiology Laboratory and Integrated Laboratory, Division of Animal Product Technology, Department of Animal Production and Technology, Faculty of Animal Science, IPB University.

Tools and Materials

A spectrophotometer (Perkin Elmer, Indonesia), a centrifuge (Z323K, Germany), centrifugal bottles with ultrafiltration membranes (Sartorius, Germany), a laminar air flow cabinet (Esco, Singapore), petri dishes, and paper discs were utilized. This study's materials included IPB-D1 chicken egg white, trypsin (EC 3.4.21.4; containing 7500 BAEE units/mg solid) (Sigma Aldrich, China), 0.1 M Na_2HPO_4, NaH_2PO_4, buffer solution, ortho-phthaldialdehyde (OPA), 2,2-diphenyl-1-picrylhydrazyl (DPPH) (Sigma Aldrich, Germany), and bacterial strains *Escherichia coli*, *Salmonella*, *Bacillus cereus*, and *Staphylococcus aureus*.

Egg Sample Preparation

The eggs used were IPB-D1 chicken eggs obtained from Sinar Harapan Farm. The egg whites and yolks were carefully separated aseptically. The collected egg whites were stored in 50 mL centrifuge bottles. For testing, egg white samples were diluted with distilled water at a 1:10 ratio (Dewi 2023).

Haugh Unit (HU) Measurement

Weighed eggs were cracked into a clear container, and the thick albumen height was measured using a caliper (Purwati *et al.* 2015). HU measurement was calculated using the formula:

$$\mathrm{HU} = 100 \log (H + 7{,}75 - 1{,}7 \,.\, W^{0{,}37})$$

where
H = height of thick egg white (mm)
W = egg weight (g)

Protein Content Analysis

Standard solutions of bovine serum albumin (BSA) were produced at concentrations of 0, 2.5, 5, 12.5, 25, 50, 125, 250, and 500 µL. A total of 0.1 mL of standard or sample was mixed with 0.1 mL of 2N NaOH, homogenized, and heated to 100 °C for 10 min. Add 1 mL of Na_2CO_3 2%, $CuSO_4$ 1%, and NaK tartrate 2% in a ratio of 100:1:1 (v:v:v). The mixture was homogenized and allowed to sit for 10−15 min at room temperature. One mL of Folin Ciocalteu was added and left for 30 min. Waterborg and Mattews (2002) used a spectrophandmeter to detect absorbance at 550 nm.

$$\text{Protein content (mg/mL)} = \frac{(\frac{\text{Sample absorbance}}{\text{Slop}} \times \text{Dilution factor})}{1000}$$

Egg White Hydrolysis

IPB-D1 egg white samples were dissolved in hydrolysis buffer at a 1:50 (w/v) ratio and hydrolyzed with trypsin (EC 3.4.21.4) at three different enzyme concentrations (0, 50, and 100 units). The hydrolysis was carried out at 37 °C in 0.1 M $Na_2.HPO_4$-NaH_2PO_4 buffer (pH 8). The samples were cultured for 0, 6, 12 h. The reaction was inactivated by heating to 95 °C for 10 min before cooling to room temperature (Je *et al*. 2007).

Degree of Hydrolysis (DH) Measurement

To create the OPA reagent, we combined 25 mL of 100 mM sodium tetrahydroborate, 2.5 mL of 2% SDS, 40 mg of OPA (dissolved in 1 mL of methanol), and 100 µL of β-mercaptoethanol, then diluted to 50 mL with distilled water. Afterward, 10−50 µL of protein sample (5-100 µg) was mixed with 1 mL of OPA reagent. The mixture was incubated for 2 min at room temperature. To assess free amino groups, absorbance was measured at 340 nm and a standard curve was established with L-serine (0−4 mg/mL) (Mirzaei *et al*. 2016).

$$\mathrm{DH} = \frac{h}{h_{\mathrm{tot}}} \times 100\%$$

where
h = number of hydrolyzed peptide bonds (meqv serine-NH_2/g protein)
h_{tot} = total peptide bonds per equivalent protein (for egg = 7.67 meqv/g protein (Rao *et al.* 2020))

Hydrolyzed Peptide Filtration

The best-hydrolyzed peptides were filtered using Amicon Ultra-0.5 ultrafiltration membranes with a size of ≤ 3 kDa. The hydrolysate was centrifuged at 6000 rpm for 15 min at 4°C (Raghavan and Kristinsson 2009), with modifications to the speed and timing.

Antioxidant Activity

Vitamin C solutions were made at concentrations of 0.50, 1, 1.50, 2, and 2.50 mg/100 mL distilled water. A total of 0.15 mL of vitamin C solution or sample was mixed with 0.90 mL of DPPH in methanol and incubated at 37 °C for 30 min. The absorbance was measured at a wavelength of 517 nm. The antioxidant potential of IPB-D1 chicken egg white hydrolysate peptide was measured using a standard curve based on DPPH vitamin C inhibition (%) and represented in mg vitamin C equivalent (EVC) per 100 g IPB-D1 chicken egg white hydrolysate peptide (Pertiwi *et al.* 2022).

$$\text{DPPH inhibition}(\%) = \frac{(\text{OD standard} - \text{OD sample})}{\text{OD standard}} \times 100\%$$

where
OD = absorbance or optical *density*

$$\text{AO capacity} = \frac{\frac{\text{capacity in AO curve}}{100} \times \frac{0{,}90}{0{,}15} \times 10}{\text{W}} \times 100$$

where
AO capacity = antioxidant capacity (mg EVC/100 g)
AO capacity curve = antioxidant capacity curve (mg/100 mL)
W = sample weight (mL)

Antibacterial Activity

The disc diffusion method was used to investigate antibacterial activity against both Gram-positive (*B. cereus* and *S. aureus*) and Gram-negative bacteria (*S. typhi* and *E. coli*). Bacteria were cultivated on Nutrient Agar (NA) and incubated at 37ºC for 24 h. Bacterial suspensions were produced in a sterile 0.85% NaCl solution corresponding to 0.5 McFarland (10^8 cfu/mL) and diluted to 10^6 cfu/mL. A cotton brush was used to disperse the bacterial suspension equally across the MHA media (Mueller Hinton Agar). A disc of paper soaked in sample, positive control (amoxicillin 0.01%), and negative control (distilled water) was placed on MHA medium and incubated at 37°C for 24 h. The inhibitory zone was measured in mm with a caliper (Suherman *et al*. 2018).

Experimental Design and Data Analysis

This study used a completely randomized design (CRD) with a one-way pattern with two treatments, IPB-D1 chicken egg white without hydrolysis (P0) and IPB-D1 chicken egg white hydrolyzed with trypsin (P1), each with three replications. Antioxidant activity data were evaluated using the *T*-test, while antibacterial activity was analyzed descriptively.

RESULTS AND DISCUSSION

Haugh Unit and Protein Content

The Haugh unit (HU) in this study is consistent with Habiburrahman *et al*. (2020), who reported a HU of 87.45 for IPB-D1 G7 eggs (Table 1). The HU is modified by various factors, including storage temperature, relative humidity, and storage time. High HU values in eggs suggest higher protein content and thicker albumen. Albumen proteins include ovalbumin, ovotransferrin, lysozyme, and ovomucin (Herranz *et al.* 2024). Egg white protein is a source of bioactive peptides that can be used as raw materials in industrial, food processing, and medicine. As a result, high-quality albumen is essential for both the food and health industries (Obianwuna *et al*. 2022). The protein content of IPB-D1 egg white exceeded that reported by Dewi (2023) for kampung chicken eggs, which varied from 51.39 to 54.27 mg/mL. This distinction could be influenced by genetics, management techniques, egg preparation, and the method used. According to Jabalera *et al.* (2022), the total protein concentration in chicken egg white typically ranges from 85 mg/mL to 100 mg/mL. Iwashita e*t al.* (2015) suggested that egg white's high protein content (100 mg/mL) enhances functional food qualities such water retention, emulsification, foaming, and gelling.

Degree of Hydrolysis (DH)

The degree of hydrolysis (DH) of IPB-D1 egg white increased with increasing incubation time and enzyme concentration. The highest DH (28.29%) was obtained after 12 h of hydrolysis with 100 units of trypsin and was chosen for further antioxidant and antibacterial testing (Table 2). This is congruent with the findings of Jin et al. (2017), who found that egg white hydrolyzed with alcalase had a DH of 23.79%, compared to 18.46% in untreated samples. DH is affected by enzyme type, concentration, temperature, pH, and hydrolysis time (Restiani 2016). The increase in DH with enzyme concentration indicates efficient hydrolysis, most likely due to the synthesis of more peptides and soluble amino acids as peptide bonds are severed during the process (Baehaki *et al*. 2015).

These findings are higher than those reported by Singh and Ramaswamy (2013), that the process of hydrolyzing egg white using trypsin enzyme for 90 min can enhance the percentage of the degree of hydrolysis while also increasing enzyme concentration and hydrolysis time. The percentages of DH were 2.78% without hydrolysis and 6.35% after trypsin enzyme treatment. Increasing the hydrolysis duration causes more peptide bonds in the substrate to break

during the hydrolysis process. Proteolysis reactions are critical in this process because they can disrupt peptide bonds, resulting in more free amino acids, carboxyl groups, and short-chain peptides with low molecular weight (Rozali *et al*. 2023).

Antioxidant Activity

IPB-D1 egg white hydrolyzed with trypsin showed considerably higher antioxidant activity and DPPH inhibition than untreated samples (p<0.05) (Figure 1). Lee *et al*. (2017) reported similar findings, where ovotransferrin hydrolyzed with Promod 2780 and thermolysin reduced DPPH by 47−62%, substantially greater than untreated ovotransferrin (< 20%). DPPH inhibition is affected by enzyme type, peptide size, amino acid makeup, and DPPH test circumstances. This work accords with Yuan *et al*. (2019): egg white without hydrolysis has an antioxidant capacity of 14 µmol AAE/g and increases to 33 AAE/g on egg whites that are hydrolyzed with numerous enzymes and go through an ultrafiltration stage.

Peptides' biological activity is regulated by their molecular weight. Low molecular weight peptides (<6 kDa) containing 2−20 amino acids outperform polypeptides or big proteins. Low molecular weight peptides isolated by ultrafiltration have been found to have the strongest antioxidant activity (Yuan *et al*. 2020). Antioxidant activity is determined by the amino acid content and certain sequence locations. Hydrophilic amino acids such as arginine may reduce antioxidant activity (Chen *et al*. 2022). Bioactive peptides' antioxidant activity is linked to the presence of amino acids like tyrosine (Tyr), tryptophan (Trp), methionine (Met), cysteine (Cys), and lysine (Lys). These amino acids can reduce Fe^{3+} ions to Fe^{2+} and chelate Fe^{2+} and Cu^{2+} ions. DPPH neutralization mechanism involves electron donation from peptides to 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical, reducing them to the stable form 1,1-diphenyl-2-picrylhydrazine (DPPH-H) (Aditia *et al.* 2018).

Antibacterial Activity

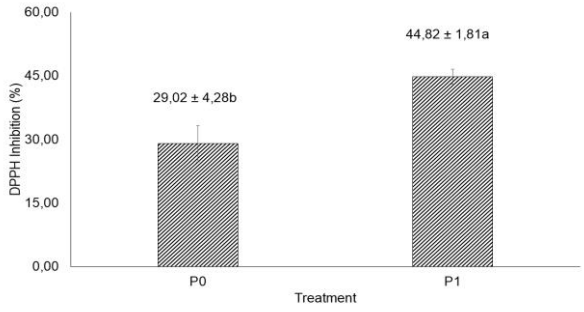
Antibacterial activity in trypsin-hydrolyzed IPB-D1 egg white increased significantly when compared to untreated samples and the negative control, although it remained lower than in the positive control. This was confirmed by the inhibition zone diameters against both Gram-positive and Gram-negative bacteria (Table 3). Hydrolysis can increase peptides' potential to break bacterial cell walls and membranes. Enzymatic hydrolysis can produce bioactive peptides with hydrophobic residues like Pro (P), Lys (K), or Arg (R), which function as antimicrobial agents (Mohammadi *et al*. 2020). These results are comparable with those of Mohammadi *et al*. (2020), who found that hydrolyzed egg white inhibited *B. cereus* (14 mm), *S. aureus* (33 mm), *S. typhi* (21 mm), and *E. coli* (13 mm) more effectively than untreated samples. The positive control (amoxicillin 0.01%), a broad-spectrum antibacterial penicillin derivative, has the biggest inhibition zones

Table 1 Haugh unit values and protein content of IPB-D1 chicken egg whites

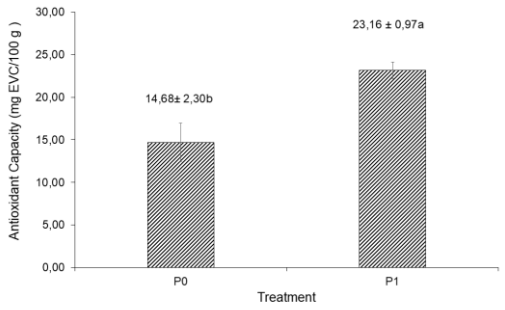
Parameter	Value
Haugh unit	74.12±11.10
Protein content (mg/mL)	103.43±1.76

Table 2 Degree of hydrolysis of IPB-D1 chicken egg white

Trypsin enzyme concentration (unit)	Degree of hydrolysis (%)		
	Incubation time (h)		
	0	6	12
0	14.00±1.77	16.09±2.11	17.04±1.69
50	14.49±3.30	16.20±4.17	18.67±0.98
100	18.14±0.75	22.33±6.38	28.29±8.82



(a)



(b)

Figure 1 Antioxidant activity of IPB-D1 chicken egg white. (a) DPPH inhibition, (b) Antioxidant capacity. Numbers followed by different letters indicate significantly different *T*-test results (p<0,05). P0: IPB-D1 chicken egg white without hydrolysis, P1: IPB-D1 chicken egg white resulting from hydrolysis with trypsin enzyme.

Table 3 Antibacterial activity of IPB-D1 chicken egg white against test bacteria

Treatment	Inhibition zone diameter (mm)			
	Gram positive		Gram negative	
	B. cereus	*S. aureus*	*B. cereus*	*E. coli*
Control (+)	8.23±0.71	6.10±2.20	7.31±1.10	6.04±1.10
Control (−)	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
P0	1.53±0.28	1.69±0.37	1.90±0.40	0.93±0.07
P1	3.48±0.68	2.90±0.24	3.42±0.50	2.42±0.40

Remarks: Control (+): amoxicillin 0.01%, Control (-): distilled water, P0: IPB-D1 chicken egg white without hydrolysis, P1: IPB-D1 chicken egg white resulting from hydrolysis using trypsin enzyme.

due to its ability to block bacterial cell wall formation. Because of the lack of antibacterial peptides, the negative control (distilled water) demonstrated no antibacterial effect (Azizah and Artanti 2019). Factors influencing inhibition zone dimensions include molecule size, diffusion rate, bacterial growth rate, incubation conditions, and antibacterial chemical concentration (Anjani *et al*. 2024).

Bioactive peptides in egg white lyse cell walls and chelate iron (Fe) from bacteria (Oles *et al*. 2016). Iron is required by bacteria to decrease ribonucleotide precursors in the DNA synthesis process, which is critical for bacterial growth and reproduction (Fikayuniar *et al*. 2022). Egg white is high in positively charged (cationic) amino acids such as Lys, His, and Arg (Carrillo *et al*. 2018). The presence of arginine in the peptide sequence is critical for antibacterial activity because it enhances the contact between the peptide and the bacterial cell membrane. This occurs because the outer surface of the bacterial cell wall is made up of negatively charged phospholipids, allowing for electrostatic interactions with positively charged peptides (Tamam *et al.* 2018). Antimicrobial peptide mechanisms rely on amino acid composition, amphipathicity, hydrophobicity, and cationic charge to allow membrane binding via barrel-stave, carpet, or toroidal-pore mechanisms (Mohammadi *et al*. 2020).

CONCLUSIONS

The results of this study show that enzymatic hydrolysis of IPB-D1 chicken egg white with trypsin greatly improves its antioxidant and antibacterial properties when compared to untreated egg white. Further research into peptide fractionation and characterization, as well as amino acid composition analysis, is required to better understand the mechanism of action and expand the potential applications of these peptides in the health and functional food industries.

ACKNOWLEDGEMENTS

The author wishes to convey his respect and admiration to Prof. Dr. Cece Sumantri, M.Sc., the primary researcher on IPB-D1 chicken breeding. Thank you to Dirjen Dikti for awarding a research grant (18828/IT.D10/PT.01.03/P/B/2023) for Regular Fundamental Research (PFR) entitled Characterization of Bioactive Peptides from IPB-D1 Local Chicken Meat and Eggs to Improve Health and Support SDGs.

REFERENCES

Aditia RP, Desniar D, Trilaksani W. 2018. Aktivitas antioksidan dan antibakteri hidrolisat protein hasil fermentasi telur ikan cakalang. *Jurnal Pengolahan Hasil Perikanan Indonesia*. 21(1): 1–12. https://doi.org/10.17844/jphpi.v21i1.21256

Agustina N, Thohari I, Rosyidi D. 2013. Evaluasi sifat putih telur ayam pasteurisasi ditinjau dari pH, kadar air, sifat emulsi dan daya kembang angel cake. *Jurnal Ilmu Produksi dan Teknologi Hasil Peternakan*. 23(2): 6–13.

Agyei D, Ongkudon CM, Wei CY, Chan AS, Danquah MK. 2016. Bioprocess challenges the isolation and purification of bioactive peptides. *Food and Bioproducts Processing*. 98: 244–256. https://doi.org/10.1016/j.fbp.2016.02.003

Anjani R, Sahputri J, Novalia V. 2024. Uji efektivitas antibakteri ekstrak etanol umbi lapis bawang merah (*Allium cepa* L.) terhadap pertumbuhan bakteri *Staphylococcus aureus*. *Jurnal Ilmiah Mahasiswa Kedokteran*. 7(2): 267–276. https://doi.org/10.31850/makes.v7i2.2918

Azizah R, Antarti AN. 2019. Uji aktivitas antibakteri ekstrak dan getah pelepah serta bonggol pisang kepok kuning (*Musa paradisiaca* Linn.) terhadap bakteri *Pseudomonas aeruginosa* dan *Klebsiella pneumoniae* dengan metode difusi agar. *Journal of Pharmaceutical Science and Clinical Research.*4(1): 29-38. https://doi.org/10.20961/jpscr.v4i1.26544

Baehaki A, Lestari SD, Romadhoni AR. 2015. Protein hydrolysis from catfish prepared by papain enzyme and antioxidant activity of hydrolyzate. *Jurnal*

Pengolahan Hasil Perikanan Indonesia. 18(3): 230–239. https://doi.org/10.17844/jphpi.v18i3.11208

Carrillo W, Lucio A, Gaibor J, Morales D, Vásquez G. 2018. Isolation of antibacterial hydrolysates from hen egg white lysozyme and identification of antibacterial peptides. *Journal of Microbiology and Food*. 21(8): 808–818. https://doi.org/10.1089/jmf.2017.0134

Chen Y, Han P, Ma B, Wang X, Ma M, Qiu N, Fu X. 2022. Effect of thermal treatment on the antioxidant activity of egg white hydrolysate and the preparation of novel antioxidant peptides. *International Journal of Food Science and Technology*. 57(5): 2590–2599. https://doi.org/10.1111/ijfs.15036

Dewi RL. 2023. Aktivitas antioksidan dari putih telur ayam kampung dan itik alabio dengan penambahan ekstrak nanas [Undergraduate thesis]. Bogor (ID): Institut Pertanian Bogor.

Fikayuniar L, Waldani DP, Lidia I, Wahyuningsih ES. 2022. Uji aktivitas antibakteri pada ekstrak biji kurma ajwa (*Phoenix dactylifera* L.) terhadap bakteri *Staphylococcus aureus*. *Journal of Biochemical Food*. 2(1): 4–7. https://doi.org/10.36465/jop.v5i2.899

Habib MFA, Murtini S, Cyrilla L, Arief II, Mutia R, Sumantri C. 2020. Performa pertumbuhan ayam IPB-D1 pada perlakuan pakan dan manajemen pemeliharaan yang berbeda. *Jurnal Agripet*. 20(2): 177–186. https://doi.org/10.17969/agripet.v20i2.16375

Habiburahman R, Darwati S, Sumantri C. 2020. Produksi telur dan kualitas telur ayam IPB D-1 G7 serta pendugaan nilai ripitabilitasnya. *Jurnal Ilmu Peternakan dan Teknologi Hasil Peternakan*. 8(2): 97–101. https://doi.org/10.29244/jipthp.8.2.97-101

Herranz B, Romero C, Sánchez-Román I, López-Torres M, Viveros A, Arija I, Pascual-Teresa S, Chamorro S. 2024. Enriching eggs with bioactive compounds through the inclusion of grape by-product in laying hens diet: Effect on internal and external egg quality parameters. *Foods*. 13(10): 1–18. https://doi.org/10.3390/foods13101553

Ismoyowati. 2020. Potensi telur sebagai immunomodulatory food di masa new normal pasca pandemi COVID-19. Prosiding Seminar Teknologi dan Agribisnis Peternakan VII. Fakultas Peternakan Universitas Jenderal Soedirman, Purwokerto (ID), 27 Jun 2020.

Iwashita K, Inoue N, Handa A, Shiraki K. 2015. Thermal aggregation of hen egg white proteins in the presence of salts. *Protein Journal*. 34: 212–219. https://doi.org/10.1007/s10930-015-9612-3

Jabalera Y, Dominguez-Gasca N, Muñoz A, Hincke M, Jimenez-Lopez C, Rodriguez-Navarro AB. 2022. Antimicrobial defenses of table eggs: Importance of antibacterial proteins in egg white as a function of hen age in an extended production cycle. *Food Microbiology*. 107: 1–7. https://doi.org/10.1016/j.fm.2022.104068

Je JY, Qian ZJ, Byun HG, Kim SK. 2007. Purification and characterization of an antioxidant peptide obtained from tuna backbone protein by enzymatic hydrolysis. *Process Biochemistry*. 42(5): 840–846. https://doi.org/10.1016/j.procbio.2007.02.006

Jin Y, Liang R, Liu J, Lin S, Yu Y, Cheng S. 2017. Effect of structure changes on hydrolysis degree, moisture state, and thermal denaturation of egg white protein treated by electron beam irradiation. LWT *Food Science and Technology*. 77: 134–141. https://doi.org/10.1016/j.lwt.2016.11.025

[Kementan] Kementerian Pertanian. 2022. *Statistik Ketahanan Pangan Tahun 2022*. Jakarta (ID): Kementerian Pertanian

Khirzin MH, Hilmi M, Prastujati AU, Mawardi N, Rahayu R. 2020. Karakteristik hidrolisat gelatin tulang itik dengan enzim tripsin sebagai penghambat alfa amilase. *Jurnal Ilmiah Inovasi*. 20(3): 55–60. https://doi.org/10.25047/jii.v20i3.2403

Kusumaningtyas E, Widiastuti R, Kusumaningrum HD, Suhartono MT. 2015. Aktivitas antibakteri dan antioksidan hidrolisat hasil hidrolisis protein susu kambing dengan ekstrak kasar bromelin. *Jurnal Teknologi dan Industri Pangan*. 26(2): 179–188. https://doi.org/10.6066/jtip.2015.26.2.179

Lee JH, Moon SH, Kim HS, Park E, Ahn DU, Paik HD. 2017. Antioxidant and anticancer effects of functional peptides from ovotransferrin hydrolysates. *Journal of the Science of Food and Agriculture*. 97(14): 4857–4864. https://doi.org/10.1002/jsfa.8356

Liao W, Jahandideh F, Fan H, Son M, Wu J. 2018. Egg protein-derived bioactive peptides: Preparation, efficacy, and absorption. *Advances in Food and Nutrition Research*. 85: 1–58. https://doi.org/10.1016/bs.afnr.2018.02.001

Mirzaei M, Aminlari M, Hosseini E. 2016. Antioxidant, ACE-inhibitory and antimicrobial activities of *Kluyveromyces marxianus* protein hydrolysates and their peptide fractions. *Functional Foods in Health and Disease*. 6(7): 425–439. https://doi.org/10.31989/ffhd.v6i7.250

Mohammadi ARA, Osman A, Enan G, Abdel-Shafi S, El-Nemer M, Sitohy M, Taha MA. 2020. Powerful antibacterial peptides from egg albumin hydrolysates. *Antibiotics*. 9(12): 1–21. https://doi.org/10.3390/antibiotics9120901

Nurjanah N, Nurhayati T, Latifah A, Hidayat T. 2021. Aktivitas antioksidan dan komponen bioaktif hidrolisat protein jeroan ikan kakap putih (*Lates calcarifer*). *Indonesian Journal of Industrial Research*. 38(1): 70–78. https://doi.org/10.32765/wartaihp.v38i1.6444

Obianwuna UE, Oleforuh-Okoleh VU, Wang J, Zhang HJ, Qi GH, Qiu K, Wu SG. 2022. Natural products of plants and animal origin improve albumen quality of chicken eggs. *Frontiers in Nutrition*. 9: 1–19. https://doi.org/10.3389/fnut.2022.875270

Oles FB, Hidayati PI, Yulianti DL. 2016. Pengaruh daya antibakteri albumin telur unggas terhadap beberapa spesies bakteri Staphylococcus sp. *Jurnal Ilmiah Kesehatan Media Husada*. 5(2): 1–11.

Pertiwi AF, Taufik E, Arief II. 2022. Karakteristik kefir susu sapi dengan penambahan ekstrak bunga telang (*Clitoria ternatea). Jurnal Ilmu Produksi dan Teknologi Hasil Peternakan.* 28(1): 34–45. https://doi.org/10.18343/jipi.28.1.34

Purwati D, Djaelani MA, Yuniwarti EYW. 2015. Indeks kuning telur (IKT), haugh unit (HU), dan bobot telur pada berbagai itik lokal di Jawa Tengah. *Jurnal Agribisnis Peternakan.* 4(2): 1–9.

Raghavan S, Kristinsson HG. 2009. ACE-inhibitory activity of tilapia protein hydrolysates. *Food Chemistry*. 117(4): 582–588. https://doi.org/10.1016/j.foodchem.2009.04.058

Rahmawati S. 2023. Karakteristik dan stabilitas enzim tripsin dalam NaCl dari usus ikan dengan kebiasaan makan yang berbeda [Undergraduate thesis]. Bogor (ID): Institut Pertanian Bogor.

Rao PS, Nolasco E, Handa A, Naldrett MJ, Alvarez S, Majumder K. 2020. Effect of pH and heat treatment on the antioxidant activity of egg white protein-derived peptides after simulated in-vitro gastrointestinal digestion. *Antioxidants*. 9(11): 1–14. https://doi.org/10.3390/antiox9111114

Restiani R. 2016. Hidrolisis secara enzimatis protein bungkil biji nyamplung (*Calophyllum inophyllum*) menggunakan bromelain. *Jurnal Ilmu-Ilmu Hayati*. 1(3): 103–110. https://doi.org/10.24002/biota.v1i3.1226

Rozali ZF, Zaidiyah Z, Lubis YM. 2023. Hidrolisis protein beras oleh ekstrak kasar enzim bromelin. *Jurnal Bioleuser*. 7(1): 11–14.

Singh A, Ramaswamy HS. 2014. Effect of high-pressure treatment on trypsin hydrolysis and antioxidant activity of egg white proteins. *International Journal of Food Science and Technology*. 49(1): 269–279. https://doi.org/10.1111/ijfs.12443

Suherman B, Latif M, Dewi STR. 2018. Potensi kitosan kulit udang vannamei (*Litopenaeus vannamei*) sebagai antibakteri terhadap *Staphylococcus epidermidis*, *Pseudomonas aeruginosa*, *Propionibacterium acnes*, dan *Escherichia coli* dengan metode difusi cakram kertas. *Media Farmasi.* 14(1): 132–143. https://doi.org/10.32382/mf.v14i1.145

Suparmono MA. 2019. Aktivitas antibakteri lisozim kering beku telur unggas lokal terhadap bakteri gram positif dan gram negatif [Undergraduate thesis]. Bogor (ID): Institut Pertanian Bogor.

Tamam B, Syah D, Lioe HN, Suhartono MT, Kusuma WA. 2018. Beberapa penciri berbasis sekuens untuk mengenali sifat fungsional peptida bioaktif: Studi eksplorasi. *Jurnal Teknologi dan Industri Pangan*. 29(1): 1–9. https://doi.org/10.6066/jtip.2018.29.1.1

Yuan J, Zheng Y, Wu Y, Chen H, Tong P, Gao J. 2020. Double enzyme hydrolysis for producing antioxidant peptide from egg white: Optimization, evaluation, and potential allergenicity. *Journal of Food Biochemistry*. 44(2): 1–12. https://doi.org/10.1111/jfbc.13113

Waterborg JH, Matthews HR. 2002. *The Lowry method for protein quantitation*. In: Walker JM, editor. *The Protein Protocols Handbook*. New Jersey (US): Humana Press. p. 7–10. https://doi.org/10.1385/1-59259-169-8:7