ISSN 1682-8356
ansinet.org/ijps

INTERNATIONAL JOURNAL OF

POULTRY SCIENCE

ANSInet
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan
Mob: +92 300 3008585, Fax: +92 41 8815544
E-mail: editorijps@gmail.com

International Journal of Poultry Science 13 (1): 21-25, 2014
ISSN 1682-8356

Endogenous Antioxidant Activity in the Egg Whites of Various Types of Local Poultry Eggs in South Sulawesi, Indonesia

Nahariah[1,2], Anang Mohamad Legowo[1], Effendi Abustam[2], Antonius Hintono[1], Priyo Bintoro[1] and Yoyok Budi Pramono[1]
[1]Doctoral Study Program, Faculty of Animal and Agricultural Sciences, Diponegoro University, Semarang, Jl. Drh. Soejono Koesoemowardojo Tembalang, Semarang-50275, Indonesia
[2]Laboratory of Meat and Eggs, Faculty of Animal Husbandry, Hasanuddin University, Makassar, Jl. Perintis Kemerdekaan, Km 8, Makassar-90245, Indonesia

Abstract: Egg is a food that had been identified as a material containing the inhibitory activity of endogenous antioxidant against free radicals. This study aimed to evaluate the potential of endogenous antioxidant activity of egg white from different kinds of local South Sulawesi poultry eggs. The materials used are kampung chicken eggs, chicken eggs and duck eggs, which were obtained from the same farm. The variables measured were proximate analysis, antioxidant activity and polyphenol content of egg whites. The results showed that the average of endogenous antioxidant activity of eggs was 20.99% which higher than that of kampung chicken eggs (9.57%) and duck eggs (15.70%). There was a tendency that antioxidant activity of duck eggs higher than that of the kampung chicken eggs, but phenolic acid and flavonoid content varied in all three types of poultry eggs. The presence of antioxidant activity in egg whites provide a positive added value in eggs and the highest was found in egg white of chicken egg.

Key word: Egg white, endogenous antioxidants, local poultry eggs, south sulawesi

INTRODUCTION

The main types of poultry that are kept in South Sulawesi, Indonesia consist of kampung chicken, chicken and duck. The three types of poultry producing eggs as a source of protein-rich foods. In addition, it contains the antioxidant activity is good for health. Kampung chicken, chicken and duck are traditionally, poultry reared and are named according to their areas of origin, respectively.

The human body has an antioxidant compound known as endogenous antioxidants to neutralize free radical compounds. Free radicals are molecule that has one or more unpaired electrons, which are unstable and highly reactive so can result in damage to cellular components such as DNA, proteins and other macromolecules (Soeksmanto *et al*., 2007; Mine and D' Silva, 2008). Antioxidants are not balanced in the body, it can lead to degenerative diseases include cancer, heart disease, cataracts, diabetes and liver, because of there is no capability to neutralize the increase of free radicals in the body (Mine and D' Silva., 2008; Morelli *et al*., 2003).

Limitedness of antioxidants availability in the human body requiring the input from the outside through the daily food consumption. The exogenous antioxidants can be obtained through supplementation of vitamin E, vitamin C, vegetables, fruits and also from foods of animal origin include meat, milk and eggs (Settharaksa *et al*., 2012; Perry *et al*., 2009; Hernandez *et al*., 2003; Robbins, 2003; Watson, 2002).

In general, animal products such as meat, milk and eggs are recognized as a source of bioactive compounds, including antioxidant (Yu *et al*., 2011). The availability of antioxidants in eggs associated with the presence of several vitamins, including vitamin A, vitamin E, carotenoids, reduced glutathione, selenium and glutathione peroxidase that relates to embryonic development in the egg and the occurrence of stress during embryogenesis phase (Surai, 2003). Therefore, the different types of local South Sulawesi poultry will have different antioxidant characteristics that need to be studied. This study examined the potential of endogenous antioxidant activity of egg white from different kinds of poultry eggs.

MATERIALS AND METHODS

Materials: The materials used were 15 kampung chicken eggs, 15 chicken eggs and 15 duck eggs. Each type of eggs originating from the same farm in Makassar, South Sulawesi, Indonesia. First age for chicken, duck and kampung chicken were 6, 7-8 and 6-7 months, respectively. The egg white were separated from the yolk and stirred for 3 min without foam forming to obtain liquid egg whites that were ready to be analyzed.

Corresponding Author: Nahariah, Doctoral Study Program, Faculty of Animal and Agricultural Sciences Diponegoro University, Semarang, Jl. Drh. Soejono Koesoemowardojo Tembalang-Semarang 50275, Indonesia

Proximate analysis: Proximate analysis were performed to evaluate moisture, ash, protein, fat and carbohydrates content of liquid egg white (AOAC, 1984).

Antioxidant activity: Measurement of antioxidant activity by metode DPPH (1, 1-diphenyl-2-picrylhydrazyl) (Pajak *et al*., 2013) were slightly modified. An aliquot of 3.9 mL of 0.1 mM DPPH radical in methanol was mixed with 0.1 mL of methanol extract samples. The mixture of DPPH reagent was shaken vigorously and allowed to stand at ambient temperature in the dark for 3 h. The absorbance of the sample was measured with a UV-VIS spectrophotometer (Shimadzu brand) at 515 nm. DPPH Radical Scavenging Effect (%) = $[(A_{DPPH}-A_{Sample})/A_{DPPH}]x100$, A_{DPPH} is the absorbance of DPPH, A_{Sample} is the absorbance of the sample.

Determination of total phenolic content: Total phenolic content of the egg whites were analyzed using the Folin-Ciocalteau reagent method (Ratnayani *et al*., 2012) with slight modifications. Sample in dilute egg white with 0.4 deionized water and dissolved with 85% methanol in a 10 mL volumetric flask to mark boundaries. The mixture was filtered and the filtrate was pipetted 1.0 mL of Folin reagent was added to 0.8 mL, inserted in a 10 mL volumetric flask. After the mixture was shaken. Furthermore, Na_2CO_3 5% was added to mark boundaries, so that the total volume of the solution to 10 mL. The solution was allowed to stand for 60 min and absorbance was measured using a spectrophotometer Shimadzu brand at maximum wavelength. The total phenolic content expressed as mg Gallic Acid Equivalent (GAE) per g of dry matter (dm).

Determination of flavonoid compounds: Flavonoid content in liquid egg white were analyzed using the spectrophotometric method described by Pajak *et al*. (2013) with slight modifications. Four milliliters of deionized water and 0.3 mL of sodium nitrate (15 g/100 mL) was added to 1 mL of methanol, then 0.3 ml of aluminum chloride was added to a solution of methanol (10 g/100 mL) and 4 mL of sodium hydroxide solution (4 g/100 mL) was added to a solution of a mixture of methanol then was added water until it reaches a final volume of 10 mL. The mixture was stirred and allowed to stand for 15 minutes to form a yellow color. The absorbance was measured at 645 nm using spectrophotometer (Shimadzu Brand). The amount of flavonoids in the egg white was calculated by equation from a standard curve of quercetin solution and expressed as mg Quercetin Equivalent (QE) per g dm of egg white.

RESULTS AND DISCUSSION

Endogenous antioxidant activity of egg whites: Figure 1 showed the endogenous antioxidant activity in chicken eggs was 20.99% higher than that of the kampung chicken eggs (9.57%) and duck eggs (15.70%) and there was a tendency, these duck eggs had a higher antioxidant activity than that of the kampung chicken eggs. The existence of these differences were likely due to the ability of different types of poultry in converting feed containing antioxidant into products. Antioxidant activity in kampung chicken and duck eggs takes to prevent free radicals and stress on embryonic development (Surai, 2003; Biard, 2005) so that both have lower antioxidant activity. Laying hens produce eggs for consumption purposes and generally not fertilized so there is no process of embryogenesis and consequently relatively more stable antioxidant activity

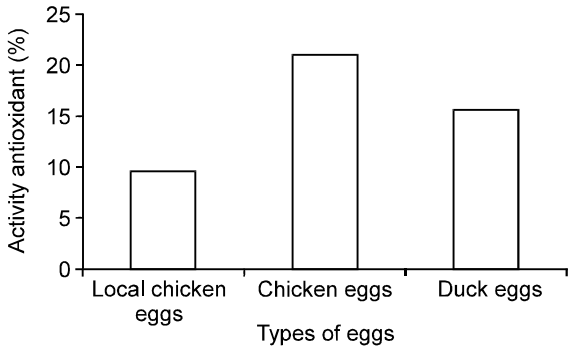

Fig. 1: Endogenous antioxidant activity of egg whites in different types of poultry

Generally, many studies on antioxidant in egg yolk have shown that bioactive compounds transferred from feed to the egg yolk (Nirmalaratne *et al*., 2011), because the yolk function as a food source for prospective chicken embryos (Surai, 2003). Poultry that consume a lot of feed will produce eggs that contain high antioxidants. Poultry feed of laying hens or race comes from the high content of concentrate feed corn containing antioxidant-rich compounds phenolate (Nirmalaratne *et al*., 2011). This fact will be different compared to chicken and duck with a pattern of semi-intensive and extensive maintenance and obtained only from vegetable feed, household food, waste straw and other agricultural residues. The type of feed used was not a source of antioxidants so that the antioxidant activity of the egg was less available. Feeding grain mainly containing grains during germination will provide high antioxidant activity (Pajak *et al*., 2013). They states that the increase in antioxidant activity during germination values appear to be associated with an increased content of antioxidant compounds such as vitamins and polyphenols.

This study showed that the antioxidant activity in the egg white varied from 9.57 to 20.99% were lower than the antioxidants found in honey by 1.3%-25.61% (Gasic *et al*., 2014) and antioxidants in lettuce by 2.5-30% (Cruz *et al*., 2014) and meat laying rejected by 20.77% (Okarini *et al*., 2013). According to Surai (2003) the content of

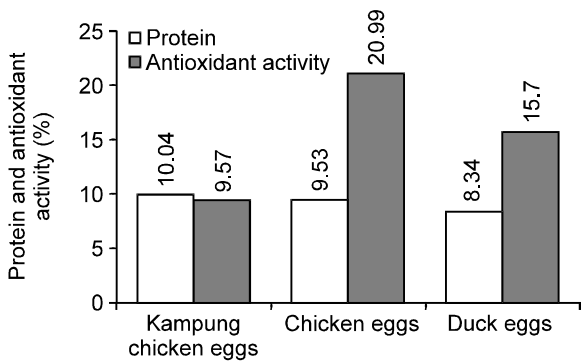

Fig. 2: Comparison value of protein and antioxidant activity in different types of poultry eggs whites

antioxidants in egg white by approximately 0.7 µg/mL will increase again by 3.0 µg/mL when the poultry were fed diets containing carotenoids.

Plants contain many phenol compounds, lipids, proteins and carbohydrates were known as natural antioxidants (Morelli *et al*., 2003; Balasuriya and Rupasinghe, 2011; Girgih *et al*., 2013). Antioxidants were also found in animal food products that contain high protein (Hernandez *et al*., 2003). Egg was a food that contains high protein with essential amino acid as a source of bioactive compounds, one of which was an antioxidant (Yu *et al*., 2011).

Figure 2 showed that there were differences in the levels of protein in the kampung chicken eggs (10.04%), chicken eggs (9.53%) and duck eggs (8.34%). These results showed that eggs had different antioxidant activity and there were a tendency that the high protein content were not followed by increased activity of antioxidants. The highest protein content was found in the kampung chicken egg but the highest antioxidant activity was found in chicken eggs. This difference was caused by the physiological conditions and the ability of each of the different types of poultry to convert feed into protein body (Ismoyowati and Sumarmono, 2011). Differences in content of proteins can also be caused by the composition of egg protein undergoes oxidation-inducing amino acids and reducing transition metals to form Reactive Oxygen Species (ROS) (Surai, 2003). Protein oxidation resulting in reduced activity of antioxidant available.

Protein oxidation in kampung chicken eggs was higher than other types of eggs that reduced antioxidant activity of protein breakdown that needs to produce a peptide having antioxidant activity. It was in accordance with the opinion of Mine and D' Silva (2008) that the hydrolysis of the egg white protein ovalbumin mainly produces potent antioxidant activity and inhibits free radical is the arrangement of the peptide Tyr-Ala-Glu-Glu-Arg-Tyr-Ile-Pro-Leu. Amino acids have a double bond or bonded with sulfur, can react directly with singlet oxygen (Raharjo, 2006). Hydrolysis of proteins in salmon and surimi by protease enzymes that produce peptide as an antioxidant by reducing oxidative stress and prolong shelf life by reduction of fat oxidation (Girgih *et al*., 2013; Wiriyaphan *et al*., 2012). Heating treatment can alter the structure of proteins that react Mailard and inhibitory effect of DPPH and fat oxidation (Jung *et al*., 2014; Sun *et al*., 2004). Heat treatment can break covalent bonds and release some phenolic compounds that increase its antioxidant capacity (Settharaksa *et al*., 2012).

Antioxidant profile on the type of different poultry egg whites: Egg white components was consists mostly of proteins such as ovalbumin, conalbumin, ovomucoid, lysozyme, ovomucin, avidin, ovoglubolin, ovoinhibitor and flavoprotein. Two of which ovalbumin and lysozyme were identified having antioxidant activity (Powrie and Nakai, 1994; Mine and D'Silva, 2008).

Bioactive compounds in the egg white were a type of flavonoids and phenolic acid, both were types of phenols which have good antioxidant activity capacity (Shettharaksa *et al*., 2012). Bioactive compounds in the egg white were a type of flavonoids and phenolic acid, both were types of phenols which have good antioxidant activity capacity (Shettharaksa *et al*., 2012). Figure 3 showed that the levels of flavonoids of a kampung chicken egg, chicken egg and the duck egg was 0,011, 0,010 and 0.024 (QEmg/mL), respectively. Similarly, on Fig. 4 showed the levels of phenolic acid of a kampung chicken egg, chicken egg and duck egg was 0.022, 0.019 and 0.035 (GAEmg/mL), respectively.

Antioxidant profile consisting of phenolic acids and flavonoids having antioxidant activity which effectively counteract free radicals (Singer *et al*., 2011; Settharaksa *et al*., 2012). Levels of flavonoids and phenolic acids were indicated to have the highest antioxidant activity in duck eggs than other types of poultry eggs but the levels of both the kampung chicken eggs was slightly higher than chicken eggs. This suggests that differences in the type of livestock led to differences in the levels of antioxidant activity followed by different antioxidants profile. The difference is probably due to the composition of each constituent of the antioxidant compounds of different kinds of poultry eggs.

Flavonoids were compounds having side chain groups of glucose. Glucose chains can affect the interaction of carbohydrates with amino acids produces antioxidant activity on different types of poultry egg whites, according to (Jin *et al*., 2004). which states that the reaction between lysine and sugar will provide antioxidant effects. Lysine was an amino acid in a protein and sugar is a type of carbohydrate. Figure 5 shows the differences in the levels of protein and carbohydrates for the three types of poultry eggs, where as duck eggs had carbohydrate levels higher than kampung chicken eggs and chicken eggs. There was a tendency on foods that

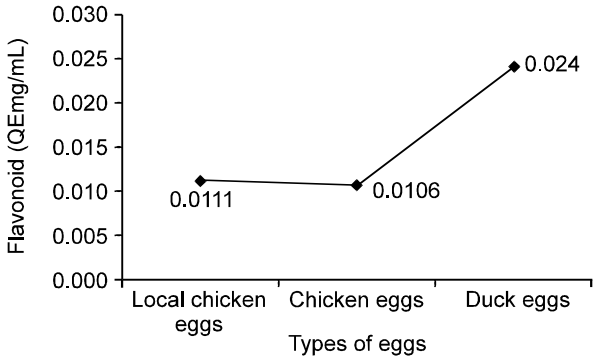

Fig. 3: Flavonoid content of poultry egg whites in different types

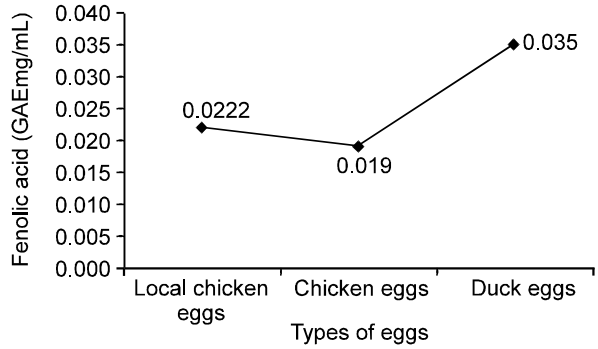

Fig. 4: Content of phenolic acid poultry egg whites in different types

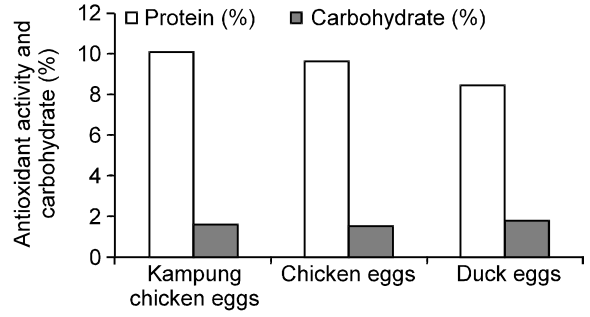

Fig. 5: Comparison value of protein and carbohydrate in different types of poultry eggs whites

contain high carbohydrates can produce high concentrations of antioxidants, but also resulted in lower antioxidant activity. It showed antioxidant activity at low concentrations to work effectively. In accordance with Kim *et al.* (2002) that the phenol group and the double bond determines the resulting concentration of antioxidants, lower concentrations of antioxidants resulted in higher antioxidant activity. Thing was different according to Singer *et al.* (2011) that the binding ability of peroxyl radicals were not correlated with the level of flavonoid compounds because of the possibility of the

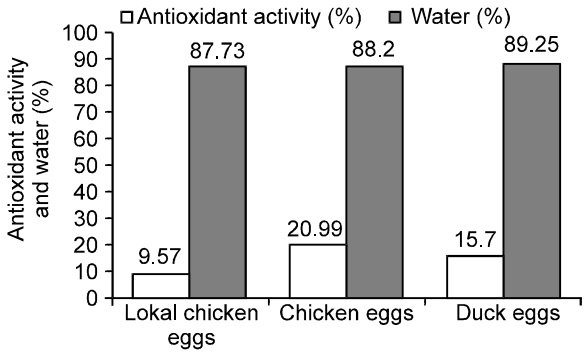

Fig. 6: Comparison value of water and antioxidant activity in different types of poultry eggs whites

formation of the phenoxyl radical in the compounds, although in general the total phenolic compounds have antioxidant activity.

Flavonoids were water soluble type of antioxidant, Fig. 6 showed that the water content of duck eggs (89.25%) was higher than in chicken eggs (88.20%) and kampung chicken eggs was 87.73%. The high water content in duck egg indicated that antioxidant compounds can dissolved in water and can decreased the activity of antioxidant.

Conclusion: Egg whites from three different kinds of local Indonesian poultry egg have been identified endogenous antioxidant activity and highest potential found on chicken eggs by 20.99% with total phenolic content was lower than other types of eggs was 0.029 (mg/mL).

ACKNOWLEDGEMENTS

The authors thank the Directorate of Research and Community Service, Directorate General of Higher Education, Ministry of Education and Culture which funded this research through the research grants Graduate Program 2013 with contract number 154c-1/UN7.5/PG/2013. DIPA Undip no 023.04.2.189815/ 2013. Also thanks to the wisdom of M. Ali and Ferliem for much helping in processing and reading of data.

REFERENCES

AOAC, 1984. Official Methods of Analysis. 12th. ed. Association of Official Analysis Chemist. Washington D.C.

Balasuriya, B.W.N. and H.P.V. Rupasinghe, 2011. Plant flavonoid as angiotensin converting enzyme inhibitory in regulation of hypertension. Functional Food in Healthy and Dis., 5: 172-188.

Biard, C., P.F. Surai and A.P. Moller, 2005. Effect of carotenoid availability during laying on reproduction in the blue tit. Oecologia, 144: 32-44.

Cruz, R., T. Gomes, A. Ferreira, E. Mendes, P. Baptista, S. Cunha, J. A. Pereira, E. Ramalhosa and S. Casal, 2014. Antioksidant activity and bioactive compounds off lettuce improved by espresso coffee residues. Food Chem., 145: 95-101.

Gasic, U., S. Keckes, D. Dabic, J. Trifkovic, D.M. Opsenica, M. Natie and Z. Tesic, 2014. Phenolic profile and antioxidant activity of Serbian polyfloral honeys. Food Chem., 145: 599-607.

Girgih, A.T., C.C. Udenigwe, F.M. Hasan, T.A. Gill and R.E. Aluko, 2013. Antioksidant properties of salmon (*salmo salar*) protein hydrolysate and peptide fractions isolated by reverse-phase HPLC. Food Res. Int., 52: 315-322.

Hernandez, L. Zomeno, B. Arino and Blasco, 2003. Antioxidant, lipolitic and proteolitic enzyme activities in pork meat from different genotypes. Meat Sci., 66: 525-529.

Ismoyowati and J. Sumarmono, 2011. Fat and cholesterol contents of local duck (*Anas platyrhynchos platyrhynchos*) meat fed mash, paste and crumble pasta. Asian J. Poult. Sci., 5: 150-154.

Jin, H. and D.D. Kitts, 2004. Antioksidant activity of sugar-lysine mailard reaction products in cel free and cell culture systems. Arch. Biochem. and Biophy., 429: 154-163.

Jung, W.K., P.J. Park, C.B. Ahn and J.Y. Je, 2014. Preparation and antioksidant potential of mailard reaction product from(MPRS) chitooligomer. Food Chem., 145: 173-178.

Kim, H.J., J.C. Eun, H.C. Sung, K.C. Shin, D.P. Heu and W.C. Sang, 2002. Antioxidative activity of resveratrol and its derivatives isolated from seed of *Paeonia lactiflora*. Biosci. Biotech. Biochem., 66: 1990-1993.

Mine, Y. and I. D'Silva, 2008. Bioactive component in egg white. In: Egg Bioscience and Biotechnology. Y. Mine. A John Willey and Sons, Inc. Publication. Canada.

Morelli, R., S.R. Volpe, N. Bruno and R.L. Scalzo, 2003. Fenton-dependent damage to carbohydrates: Free radical scavenging activity of some simple sugars. J. Agri. Food Chem.

Nirmalaratne, C., D.L. Lutz, A. Schieber and J. Wu, 2011. Free aromatic amino acids in egg yolk show antioksidan properties. Food Chem., 129: 155-161.

Okarini, I.A., H. Purnomo, Aulanni'am and L.E. Radiati, 2013. Proximate, total phenolic, antioxidant activity and amino acids profile of bali indigeneous chicken, spent laying hen and broiler breast fillet. Int. J. Poult. Sci., 12: 415-420.

Pajak, P., R. Socha, D. Galkowska, J. Roznowski and T. Fortuna, 2013. Phenolic profile and antioxidant activity in selected seeds and sprouts. Food Chem., 143: 300-306.

Perry, A., H. Rasmussen and E.J. Johnson, 2009. Xanthophyll (*lutein, zeaxanthin*) content in fruits, vegetables and corn and egg product. J. Food Composition and Analysis, 22: 9-15.

Powrie, W.D. and S. Nakai, 1994. In: Egg Science and Technology. Ed. Stadelman, W.J and O.J. Cotterill. The Chemistry of Egg and Egg Product. Food Product Press An Imprint of The Haworth Press, Inc. New York. London.

Ratnayani, K., M. Laksmiwati and N.P.I. Septin, 2012. Kadar total senyawa fenolat pada madu randu dan madu kelengkeng serta uji aktivitas antiradical bebas dengan metode DPPH (Difenilpikril Hidrazil). J. Kimia, 6: 163-168.

Raharjo, S., 2006. Kerusakan Oksidatif pada Makanan. Gadjah Mada University Press Yogyakarta. Yogyakarta. ISBN: 979-420-620-2.

Robbins, R.J., 2003. Phenolic Acids in Food: An Overview of analytical methodology. J. Agri. Food Chem., 51: 2866-2887.

Settharaksa, S., A. Jongjareonrak, P. Hmadhlu, W. Chansuwan and S. Siripongvutikorn, 2012. Flavonoid, phenolic contents and antioxidant properties of thai hot curry paste extract and its ingredients as affected of pH, solvent types and high temperature. Int. Food Res. J., 19: 1581-1587.

Soeksmanto, A., Y. Hapsari and P. Simanjuntak, 2007. Kandungan antioksidan pada beberapa bagian tanaman mahkota dewa, *Phaleria macrocarpa* (Scheff) Boerl. (Thymelaceae). Biodiversitas, 8: 92-95.

Sun, Y., S. Hayakawa and K. Izumori, 2004. Modification of ovalbumin with a rare ketohexose through the mailard reaction: Effect on protein structure and gel properties. J. Agri. Food Chem., 52: 1293-1299.

Singer, A., J. Czubinski, P. Kachlicki, K. Dwiecki, E.L. Szczapa and M.N. Kalucka, 2011. Antioxidant activity and phenolic content in three lupin species. J. Food Composition and Analysis, 25: 190-197.

Surai, P.F., 2003. Natural Antioksidans In Avian Nutrition And Reproduction. Nottingham University Press. Nottingham. ISBN: 1-897676-95-6.

Wiriyaphan, C., B. Chitsomboon and J. Yongsawadigul, 2012. Antioxidant activity of protein hydrolysates derived from threadfin bream surimi by products. Food Chem., 132: 104-111.

Yu, Z., Y. Yin, W. Zhao, Y. Yu, B. Liu, J. Liu and F. Chen, 2011. Novel peptide derived from egg white protein inhibiting alpha-glucosidase. Food Chem., 129: 1376-1382.