Abanico Veterinario. January-December 2020; 10:1-12. http://dx.doi.org/10.21929/abavet2020.14
Original Article. Received: 26/02/2020. Accepted: 17/06/2020. Published: 05/07/2020. Code: 2020-19.

Giant Squid meal (*Dosidicus gigas*) inclusion, as a source of protein in laying hens' diet

Inclusión de harina de calamar gigante *Dosidicus gigas* como fuente de proteína en dietas para gallinas ponedoras

Carranco-Jáuregui María[1] ID, Fuente-Martínez Benjamín[2*] ID, Ramírez-Poblano Miriam[3], Calvo-Carrillo María[1] ID, Ávila-González Ernesto [2] ID

[1]Departamento de Nutrición Animal Dr. Fernando Pérez-Gil Romo, Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán. Ciudad de México. México. [2]Centro de Enseñanza, Investigación y Extensión en Producción Avícola, Facultad de Medicina Veterinaria y Zootecnia, Universidad Nacional Autónoma de México. Ciudad de México. México. [3]Alumna de la Facultad de Química, Universidad Nacional Autónoma de México. Ciudad de México. México. * Responsible author and for correspondence: Fuente-Martínez Benjamín. Manuel M. López S/N, Colonia Santiago Zapotitlán, Alcaldía Tláhuac, 13209, Ciudad de México. México. rexprimero@hotmail.com, benjaminfuente@yahoo.com.mx, concepcion_calvo1@hotmail.com, avilaernesto@yahoo.com.

ABSTRACT

The giant squid *Dosidicus gigas* has a great potential to elaborate human consumption products and balanced food due to its high protein content. The objective of this work was to include 10 and 20 % of giant squid meal (GSM) as a protein source in diets for laying hens. One hundred thirty-five Bovans White hens were distributed as follows: control (T), 10 % (T1), and 20 % (T2), in a 6-week trial. The Productive variables measured were the physical quality of the egg, crude protein, amino acid profile, and sensory evaluation (taste), Raw GSM protein (77.76 %), amino acids (g aa/100 protein): methionine + cysteine (3.76), lysine (10.16), isoleucine (4.26), leucine (6.56), phenylalanine (4.56) and tryptophan (2.0). The Productive variables measured were ($P < 0.05$) in posture (%); egg weight (g); food conversion (kg: kg) and egg mass (bird/day/g), and without difference ($P > 0.05$) consumption (bird/day/g); physical quality of the egg with differences ($P < 0.05$) in egg weight (g), albumin height (mm) and Haugh units (UH). In egg raw protein and amino acid differences ($P < 0.05$), Sensory evaluation ($P > 0.05$), rating the 3 treatments in 4 "likes". According to the results, it is concluded that giant squid flour not exceeding 10 % can be an alternative as a source of protein for feeding posture hens.
Keywords: Giant squid meal, chemical composition, laying hens, egg.

RESUMEN

El calamar gigante *Dosidicus gigas* presenta un potencial para elaborar productos para consumo humano y alimentos balanceados por su alto contenido de proteína. El objetivo de este trabajo fue incluir 10 y 20 % de harina de calamar gigante (HCG) como fuente de proteína en dietas para gallinas ponedoras. Ciento treinta y cinco gallinas Bovans White se distribuyeron en testigo (T), 10 % (T1) y 20 % (T2) en ensayo de 6 semanas. Se midieron variables productivas, calidad física del huevo, proteína cruda, perfil de aminoácidos y evaluación sensorial (sabor). Proteína cruda en HCG (77.76 %), aminoácidos (g aa/100 de proteína): metionina+cisteína (3.76), lisina (10.16), isoleusina (4.26), leucina (6.56), fenilalanina (4.56) y triptófano (2.0). Variables productivas ($P < 0.05$) en postura (%); peso del huevo (g); conversión alimentaria (kg:kg) y masa de huevo (ave/día/g), y sin diferencia ($P > 0.05$) consumo (ave/día/g). Calidad física del huevo con diferencias ($P < 0.05$) en peso de huevo (g); altura de albúmina (mm) y unidades Haugh (UH). En huevo proteína cruda y aminoácidos diferencias ($P < 0.05$). Evaluación sensorial ($P > 0.05$), calificando los 3 tratamientos en 4 "gusta". Se concluye que la harina de calamar gigante puede ser una alternativa como fuente de proteína para la alimentación de gallinas de postura no mayor al 10 %.
Palabras clave: Harina de calamar gigante, composición química, gallinas ponedoras y huevo.

INTRODUCTION

Mexico is one of the main countries in the world in the giant squid fishery (*Dosidicus gigas* D'Orbigny 1835) (Montaño *et al*., 2015). It is an oceanic and migratory species from the Eastern Pacific Ocean, it is distributed from Monterey, California, USA to the north of Chile. Of the squid species, *Dosidicus gigas* is exploited commercially in Mexico, its catch is officially registered in the Gulf of California and it is unloaded in the ports of Mazatlán, Sinaloa; Santa Rosalía, Baja California Sur and Guaymas, Sonora (Luna *et al*., 2006).

The capture of the giant squid in recent years has been abundant and important, as a resource within the fishing production system in Mexico. The catch of 40,878.02 kg in Guaymas, Sonora was reported for 2014 (CONAPESCA, 2017). In general, 75% of the squid is used without viscera, and as it is a food of marine origin, its nutritional value is considered good. It stands out the content of proteins (53%) of easy digestion (digestibility = 94%), non-assimilable carbohydrates, vitamins A, D and complex B, low fat and caloric content (Martínez-Vega *et al*., 2000; Luna *et al*., 2006; Toyes, 2016).

There is information on the ecology, biology, reproduction and distribution of the giant squid (*Dosidicus gigas*); as well as the chemical composition of fresh whole squid with a humidity of 82.23%, crude protein 15.32%, ash 1.31% and fat 0.87%; in flour a content of crude protein 77.76%, ash 8.54% and fat 6.33% is reported (Abugoch *et al*., 1999; Alegre *et al*., 2014; Calvo *et al*., 2016).

On the other hand, poultry activity contributes significantly to the food production system in the world, which is why a search is necessary to improve every day each of the aspects involved in the production process. Acquiring the right feed to obtain optimal results in poultry farming is an important aspect where the feed represents the highest percentage of the investment made in the production cycle; protein being the ingredient with the highest cost in the production of balanced foods. Therefore, research in the poultry area focuses on the possibility of modifying the chemical composition of its products; for example, reducing the level of cholesterol and saturated fat and enriching them with unsaturated fatty acids, vitamins, minerals, antioxidant pigments and proteins; both in chicken meat and in eggs (Morales *et al*., 2013; Martínez *et al*., 2016).

The use of giant squid meal (GSM) in bioassays applied to laying hens will allow to know the optimal level of inclusion in the diet to take advantage of this protein resource in the poultry industry. GSM has been used as feed in shrimp farms, but there is no scientific literature that reports its use as feed for birds, as has been done with other products of marine origin such as fish oils and meals, crustaceans and seaweed; as sources of protein, n-3 and n-6 fatty acids and pigments.

The objective of this research was to determine the effect of the inclusion of giant squid meal, as a protein source in diets for laying hens, and its effect on the productive variables and egg quality.

MATERIAL AND METHODS

Obtaining, receiving and storing giant squid meal (GSM)

The GSM (mantle, tentacles, viscera, feather and mouth) was provided by a marine products processing plant in Guaymas, Sonora, Mexico, it was transported to the Dr. Fernando Pérez-Gil Romo Department of Animal Nutrition of the National Institute of Medical Sciences and Nutrition Salvador Zubirán, and they were kept frozen (-20 °C) until their analysis and use.

Chemical analysis of giant squid meal (GSM)

It was analyzed by the standardized methods described by AOAC (2005) (6 repetitions): humidity by drying oven (method 934.01), ashes by calcination (method 942.05), crude protein by Kjeldahl (Nx6.25) (method 976.05), extract ethereal by Soxhlet (method 2003.06) and extract free of nitrogen and minerals by atomic absorption spectrophotometry. Gross energy using Calorimetric Pump (Parr Instrument Company, Inc., Moline Illinois). Amino acid profile by HPLC (Método Waters, 1993). Hydrolyzed Amino Acid Standards, Pierce Brand, Catalog NCI 0180.

The sample was hydrolyzed with phenol and 6N HCl, to later derivatize it with a phosphate buffer and 6-aminoquilonyl-N-hydroxysuccimonolyl carbamate (derivatizing reagent AccQ-tag fluor), converting the primary and secondary amino acids into stable derivatives of ureas that strongly fluoresce at 395 nm. The standards are derivatized in the same way as the sample.

HPLC conditions (Waters model 2475): a 4 µm Nova-Pak C18 high efficiency AccQ-Tag column was used, mobile phase with eluent A: WATERS AccQ-TAG buffer; eluent B: acetonitrile and eluent C: MILLI-Q HPLC grade water, 60 min. run time, Waters 470 nm fluorescence detector, column temperature 37 ° C and 5 µL injection volume, and continue with the analytical procedure described in the Waters Operating Manual for this column (Waters 1993).

Preparation of diets and behavior of birds

The study was carried out at the Center for Teaching, Research and Extension in Poultry Production (CEIEPAv) of the Faculty of Veterinary Medicine and Zootechnics of the National Autonomous University of Mexico, Mexico City, with a height of 2,250 m above sea level sea, temperate-humid climate, average annual temperature of 16 °C and average annual rainfall of 747 mm (CONAGUA, 2019). The procedure followed in the handling of the birds complied with NOM-062-ZOO-1999, technical specifications for the production, care and use of laboratory animals.

Table 1. Experimental diets supplemented with giant squid meal *Dosidicus gigas*

Ingredient	Control (T)	T + 10 % GSM	T + 20 % GSM
Sorghum	564.823	621.553	652.180
Soybean paste	269.096	197.115	149.015
Calcium carbonate	99.593	99.902	100.164
Vegetable oil	38.212	28.320	21.560
Giant Squid Meal (GSM)	0.000	26.910	53.820
Orthophosphate 1820	16.490	14.768	12.940
Salt (NaCl)	4.649	4.671	4.688
DL-Methionine 99%	1.768	1.566	1.134
Vitamin premix[1]	1.000	1.000	1.000
Mineral premix[2]	0.500	0.500	0.500
Toxisorb[3]	1.000	1.000	1.000
Avelut powder[4]	1.000	1.000	1.000
L-Lysine HCl 78.8%	0.870	0.693	0.000
Choline Chloride 60%	0.500	0.500	0.500
Vegetable red pigment[5]	0.200	0.200	0.200
Antioxidant[6]	0.150	0.150	0.150
Furafeed[7]	0.150	0.150	0.150
Total	1000.0	1000.0	1000.0
Price (National Currency)	120.59	109.51	99.52
		Nutrient analysis	
Metabolizable energy, kcal / kg	2.871	2.850	2.850
Crude protein, %	18.334	17.274	17.169
Methionine + total cystine,%	0.747	0.743	0.741
Total lysine,%	0.963	0.961	0.984
Total threonine,%	0.686	0.702	0.756
Total tryptophan,%	0.228	0.186	0.157
Total calcium,%	4.001	4.001	4.001
Available phosphorus,%	0.440	0.440	0.440
Sodium,%	0.190	0.190	0.190

[1]Vitamin content per kg: A; 4.0 MUI: D_3; 666,666.7 IU: RovomixHyD; 5 kg: K_3; 1.67 g: B_1; 0.83 g: B_2; 2.33 g: B_6; 1.17 g: B_{12}; 6,666.67 mg: Niacin; 10 g: D-Pantothenic Acid; 3.33 g: folic acid; 0.33 g: Biotin; 33.33 mg: Choline; 100 g. [2]Mineral content per kg: Iron; 20 g: Zinc; 26.67 g: Manganese; 36.67 g: Copper; 5 g: Iodine; 0.33 g: Selenium; 0.1 g. [3]Mycotoxin sequestrant. [4]Source of natural yellow xanthophylls. [5]Avired: 5 g/kg (minimum) of *Capsicum* spp. Fruit xanthophylls. [6]BHA; 1.2%: BHT; 9.0%: Ethoxyquin; 4.8%: Chelating agents; 10.0%. [7]Antifungal.

135 first cycle Bovans White hens, 18 weeks old, housed in battery cages with 3 hens each were used; They were distributed with a completely randomized design in 3 treatments with 5 repetitions with 9 birds each: control diet, 10 and 20 % of protein from GSM. The Allix2 computer program was used. See 5.37.1 to formulate the experimental diets supplemented with GSM. The procedure used consists of including the data from the proximal chemical analysis of the ingredients. The calculation was made based on the protein contribution, where the GSM protein partially replaced that of the soybean paste, which met the nutritional needs of the lineage according to the production phase (table 1).

Quantification of crude protein and amino acid profile in the egg

At the end of the evaluation of the physical quality of the egg, of these, 5 of each repetition were taken at random and mixed with a hand mixer, and the crude protein quantification was carried out by the Kjeldahl method (AOAC, 2005). The quantification of the amino acid profile was carried out by HPLC, using the AccQ-TAG Waters 1993 method (Manual No. WATO52874, Waters, 1993).

Sensory evaluation

10 eggs were sampled from each treatment (2 per repetition), they were cooked in scrambled egg form without oil and without salt. A taste level test was carried out, and a 5-point scale was established (1 = dislike very much; 2 = dislike; 3 = neither like nor dislike; 4 = like and 5 = like very much). Thirty untrained judges participated, but they were habitual egg consumers (Anzaldúa, 2014).

Statistical analysis

For all the variables studied, an analysis of variance (ANDEVA) was carried out with 95% confidence, and the difference between means by the Tukey test, through the statistical package of Statistical Analysis System (SAS Inst. Inc., 2003). The results of the sensory evaluation were analyzed using Friedman's non-parametric test ($P < 0.05$) (Anzaldúa, 2014).

RESULTS AND DISCUSSION

Chemical analysis of GSM and diets

In table 2, the chemical composition of the giant squid meal (GSM) is presented, where the crude protein value stands out (77.76% BH and 82.82% BS). Toyes (2016) reports a crude protein content in squid viscera meal of the same species of 53%, observing a difference of 28% lower than in GSM. However, Ezquerra *et al.* (2007) reported that of the total crude protein of various species of squid, non-nitrogenous elements (trimethylamine oxide and other amines, free amino acids and octopine, arginine, glycine, betaine, alanine and nucleotides), constitute 37%; although these authors do not specify if this value corresponds to whole squid or only edible parts, and if it is fresh or in flour; however, Maza *et al*. (2003), mention in their study that the non-protein nitrogen content in fresh mantle of giant squid is 39.5%. On the other hand, in GSM the ashes (8.54%), are mainly made up of iron and sodium (0.19 and 0.16 mg/100g), respectively. As part of the process to obtain the GSM, a pressing is carried out that generates a fluid formed by water and oil, for which the fat content in the flour was 6.33%. For this reason, the energy intake is less than 4.03 Kcal/g, being similar to that published in *Illex illecebrosus* squid flour (4.13 Kcal / g) (Calvo *et al*., 2016).

It should be noted that the difference reported in the results of this study with reference to other authors could be due to the squid species, harvest season, parts of the squid analyzed, handling and preparation of the samples for analysis (fresh or in flour).

Table 2. Chemical composition, amino acid and mineral profile in giant squid meal (GSM) *Dosidicus gigas*

	GSM /wet base	GSM/dry base
Proximal analysis (g/100g)[1]		
Humidity	3.46 ± 0.002	----
Crude protein	77.76 ± 0.04	80.54
Ethereal extract	6.33 ± 0.007	6.55
Ashes	8.54 ± 0.002	8.84
Nitrogen-free extract[2]	3.91	4.07
Gross Energy (Kcal / g)	4.03 ± 0.02	4.17
Amino acids	(g aa/100 g of protein)	
Methionine *	1.64 ± 0.01	1.69
Cysteine	2.12 ± 0.03	2.19
Methionine + Cystine	3.76	3.89
Lysine *	10.16 ± 0.03	10.52
Threonine *	3.86 ± 0.02	3.99
Aspartic acid	9.53 ± 0.01	9.87
Glutamic acid	14.53 ± 0.02	15.05
Proline	5.16 ± 0.03	5.34
Wisteria	7.57 ± 0.03	7.84
Alanine	6.79 ± 0.03	7.03
Valine *	5.40 ± 0.03	5.59
Isoleucine *	4.26 ± 0.01	4.41
Leucine *	6.56 ± 0.02	6.79
Serine	3.42 ± 0.02	3.54
Phenylalanine *	4.56 ± 0.02	4.72
Arginine	3.86 ± 0.02	3.99
Histidine *	6.89 ± 0.03	7.13
Tryptophan *	2.0 ± 0.03	2.07
Minerals	(mg/100g)	
Calcium	0.15 ± 0.001	0.15
Sodium	0.16 ± 0.005	0.16
Potassium	0.14 ± 0.002	0.14
Magnesium	0.08 ± 0.005	0.08
Iron	0.19 ± 0.009	0.09

[1] n=12. [2]Por diferencia; *Aminoácidos esenciales [1] n = 12. [2]By difference; *Essential amino acids

The quality of the protein is in accordance with its profile of essential amino acids, linked to the efficiency in protein conversion (EPC). In this GSM it was found that the values of lysine, histidine, sulfur and aromatic amino acids were similar to the amino acids of milk and eggs, with the exception of lysine and histidine, present in higher amounts in milk and higher valine and isoleucine in eggs (Calvo *et al*., 2016).

Productive variables and physical quality of the egg

In table 3, it is observed that there were significant differences (P <0.05) between the control, 10 and 20 % of GSM in the% of laying (91.42, 94.23 and 89.33), egg weight (53.61, 52.82 and 52.35 g), feed conversion (1.96, 1.95 and 2.05 kg/kg) and egg mass (49.11, 49.93 and 46.91 bird/day/g) and without difference (P> 0.05), feed consumption (95.49, 96.16 and 94.07 g) respectively between the three diets. At the end of the study, the hens were 24 weeks old, and according to the data published in the Bovans White Management Guide (ISA, 2009) the egg weight 54.6 g, egg mass 49.4 g, feed consumption g/day 100 and feed conversion 2.02 kg/kg; therefore both groups of data were similar.

The results reported in this study agree with the Bovans White Management Guide (ISA, 2009), which mentions that birds between 30-35 weeks of age reach a good production development, where they report 60-61 egg weight g, egg mass of 25.5 g, feed consumption 106 g/day and feed conversion of 2.08 kg/kg.

Regarding the physical quality of the egg, the Mexican Standard (NMX-FF-127-SCFI-2016) mentions 5 categories for fresh eggs determined by weight and size (Extra-large ≥ 64, Large 60 - 64, Medium 55 - 60, Small 50 - 55 and Marble ≥ 50). In this study, the egg weight was lower with 20% GSM (52.35 g) compared to the control (53.61 g), so they are within the boy classification, justified this result by the age of the hens.

For this same Mexican standard, there is another classification for fresh eggs for dishes: extra, category I, category II and out of classification, and it mainly refers to the appearance of the shell (normal, intact and clean); air chamber (normal and not exceed 3.2 mm), clear or albumen and Haugh Units (slimy, clean and firm), and yolk (round, in the center, with visible germ disc and color between 9 and 13 on the fan scale Roche colorimetric). According to this classification, the physical quality of the studied egg falls on extra, since, although there were significant differences in HU (98.11 for 10% and 95.86 for 20% of GSM and control of 97.20 HU) and albumin height (9.42 mm and 8.90 mm for 10 and 20% GSM respectively and 9.27 mm for control), are within this standard. These variables indicate the freshness of the egg, where the albumin must be viscous (colloidal), the one that surrounds the yolk, and to distinguish 3 layers (two dense and 1 aqueous) that as the laying time passes CO_2 is lost and increases pH 7.6 to 9.7; as well as loss of moisture in the form of water vapor that will denature the proteins, and this will cause the albumin to lose its structure, making it more liquid and this leads to a lower value of albumin height and HU, as well as the capacity to keep the yolk in the center; modification that occurs when the egg has been laying for several days. Possibly the small increase in protein in the diet at 10% GSM was reflected in the physical characteristics of albumin.

Table 3. Average of the productive variables in Bovans Blanca hens and physical quality of the egg with different inclusion percentages of giant squid meal (GSM) *Dosidicus gigas*

	Productive variables		
	Control	10 % GSM	20 % GSM
Posture (%)	91.42 ± 5.08[b]	94.23 ± 6.31[a]	89.33 ± 10.37[b]
Egg Weight (g)	53.61 ± 2.62[a]	52.86 ± 2.36[b]	52.35 ± 2.56[b]
Consumption, bird/day (g)	95.49 ± 8.24	96.16 ± 8.80	94.07 ± 8.37
Feed Conversion (kg/kg)	1.96 ± 0.27[b]	1.95 ± 0.28[b]	2.05 ± 0.35[a]
Egg mass, bird/day (g)	49.11 ± 4.95[a]	49.93 ± 5.44[a]	46.91 ± 7.27[b]
	Physical quality of the egg		
Egg Weight (g)	53.55 ± 3.78[a]	52.58 ± 4.90[b]	51.83 ± 3.91[b]
Albumin height (mm)	9.27 ± 0.96[a]	9.42 ± 1.11[a]	8.90 ± 1.05[b]
Haugh Units (HU)	97.20 ± 4.49[a]	98.11 ± 5.43[a]	95.86 ± 4.99[b]
Egg classification according to Haugh Units in Mexico[1]	México Extra	México Extra	México Extra

Productive variables n = 135. Physical quality of the egg n = 300. [a, b] Different letter in the same row shows statistically different values (P <0.05). [1] Albumen height greater than 5.5 mm or in Haugh Units greater than 70 (NMX-FF-079-SCFI, 2004)

Crude protein quantification and amino acid profile in egg

In table 4, it is observed that between crude egg protein with inclusion of 10% (12.58%) and control (12.34%) there was no statistical difference (P> 0.05), but with 20% of GSM (10.79%) there was there was a difference (P <0.05).

In relation to the amino acids reported, there was a difference (P <0.05) between the control and the samples with 10 and 20% of GSM. Studies such as Naber (1979) who classified the chemical components of the egg based on changes in the diet of the hens, concluded that the nutrients proteins, amino acids, total fat and macrominerals show little variation when modifying the diet; not so with microminerals, vitamins and fatty acids that are more influenced by changes in diet, all depending on the nutrient being studied. Likewise, Stadelman and Pratt (1989) evaluated the factors that modify the composition of the egg and mention that the protein level in it increases slightly with increasing protein and energy in the diet, that the total amount of albumen will depend on the balance of amino acids of the diet, and that a deficiency in lysine or methionine will reduce the weight of the albumen and decrease the concentration of all free amino acids.

Table 4. Protein content and amino acid profile in eggs of hens fed with different inclusions of giant squid meal (GSM) *Dosidicus gigas*

	Control	10 % GSM	20 % GSM
Crude protein (%)	12.34 ± 0.56[a]	12.58 ± 0.32[a]	10.79 ± 0.67[b]
Amino acid profile (g aa / 100g protein)			
Isoleucine *	5.26 ± 0.05[a]	5.18 ± 0.03[a]	4.92 ± 0.09[b]
Leucine *	8.46 ± 0.06[a]	8.26 ± 0.06[b]	8.35 ± 0.03[b]
Lysine *	7.28 ± 0.07[a]	6.93 ± 0.02[b]	7.17 ± 0.05[a]
Methionine + Cystine *	3.91 ± 0.05[a]	3.71 ± 0.10[b]	3.55 ± 0.03[b]
Phenylalanine *	5.79 ± 0.21[a]	5.66 ± 0.06[b]	5.60 ± 0.04[b]
Threonine *	4.87 ± 0.11[a]	4.68 ± 0.07[a]	4.40 ± 0.05[b]
Valine *	6.52 ± 0.07[a]	6.32 ± 0.07[b]	6.11 ± 0.05[c]
Tyrosine	4.40 ± 0.07[a]	4.21 ± 0.09[b]	4.12 ± 0.06[b]
Arginine	6.74 ± 0.03[a]	6.52 ± 0.06[b]	6.45 ± 0.13[b]

n = 10. [a, b, c] Different letter in the same row shows statistically different values (P <0.05). *Essential amino acids

Sensory evaluation

The results obtained from the egg taste evaluation had a score of 4, which corresponds to the level of "like". In the comments of this evaluation, he did not refer to unpleasant flavors (table 5).

Finally, it is worth mentioning that the chemical composition of the egg in this study in comparison with the control egg and other reports will depend on the age of the hen, the lineage and type of management; however, the most important factor will be diet.

Table 5. Results of the evaluation of the taste of eggs from hens fed with different inclusions of giant squid meal (GSM) *Dosidicus gigas*

	Control	10 % GSM	20 % GSM
Taste	4.16 ± 1.02	4.09 ± 0.86	4.06 ± 1.01

n = 30. 5-point scale (1 = dislike very much; 2 = dislike; 3 = neither like nor dislike; 4 = like and 5 = like very much)

CONCLUSION

The results obtained in this study can be concluded that the giant squid meal *Dosidicus gigas* can be used as a source of protein in diets for laying hens up to 10% inclusion, without affecting the productive variables, the taste of the egg and with a slight increase in the protein content of the egg; Therefore, the use of this flour in the poultry industry can be an alternative.

CITED LITERATURE

ABUGOCH L, Guarda A, Pérez LM, Paredes MP. 1999. Determination of proximal chemical composition of squid (*Dosidicus gigas*) and development of gel products. *Archivos Latinoamericanos de Nutricion*. 49(2):156-161. https://europepmc.org/article/med/10488395. PMID: 10488395.

ALEGRE Ana, Frédéric Ménard, Ricardo Tafur, Pepe Espinoza, Juan Argüelles, Víctor Maehara, Oswaldo Flores, Monique Simier, Arnaud Bertrand. 2014. Comprehensive Model of Jumbo Squid *Dosidicus gigas* Trophic Ecology in the Northern Humboldt Current System. *PLoS ONE*. 9(1): e85919. https://doi.org/10.1371/journal.pone.0085919.

ANZALDÚA MA. 2014. Evaluación sensorial de los alimentos en la teoría y la práctica. Ed. Acribia S.A. Pp. 214. ISBN: 978-84-200-0767-0. https://www.editorialacribia.com/libro/la-evaluacion-sensorial-de-los-alimentos-en-la-teoria-y-la-practica_53649/

AOAC. 2005. Official methods of analysis of AOAC International. 18 ed. Arlington, VA, USA. ISBN 0-935584-77-3. http://www.eoma.aoac.org/

CALVO CMC, Carranco JME, Salinas CA, Carrillo DS. 2016. Composición química de harina de calamar gigante *Dosidicus gigas*. *Archivos Latinoamericanos de Nutrición.* 66(1): 74-81. ISSN 0004-0622. www.alanrevista.org.

CONAGUA. 2019. Resúmenes mensuales de temperaturas y lluvia. https://smn.conagua.gob.mx/es/climatologia/temperaturas-y-lluvias/resumenes-mensuales-de-temperaturas-y-lluvias.

CONAPESCA. 2017. Estadísticas pesqueras en México. https://www.gob.mx/conapesca/documentos/estadistica-pesquera-y-acuicola-de-mexico.

EZQUERRA-Brauer JM, Díaz AC, Fenucci JL. 2007. Harina de Calamar. Manual de Ingredientes Proteicos y Aditivos en la Formulación de Alimentos Balanceados para Camarones. (García Galano T, Villarreal-Colmenares H, Fenucci JL. Eds). EUDEM. Argentina. ISBN 978-987-1371-02-0. 2007. 41-55. http://retos-aaa.edu.umh.es/wp-content/uploads/sites/299/2017/06/CV-Marina-J.-Ezquerra.pdf

ISA. 2009. Bovans White. Guía de manejo Sistemas de producción en jaula. Institut de Sélection Animale BV. The Netherlands-EU. Disponible en: https://www.bovans.com/documents/261/HGL_nutrition_management_guide_L7121-2.pdf.

LUNA RMC, Urciaga GJI, Salinas ZCA, Cisneros MMA, Beltrán MLF. 2006. Diagnóstico del consumo del calamar gigante en México y Sonora. *Economía, Sociedad y Territorio*. 6(22): 535-560. ISSN: 1405-8421. https://doi.org/10.22136/est002006267

MARTÍNEZ-Vega JA, Cruz-Suárez LE, Ricque-Marie D. 2000. Composición corporal y proceso de secado del calamar gigante Dosidicus gigas. *Ciencia y Mar*. 4(11):35-38. http://www.umar.mx/revistas/

MARTÍNEZ Aispuro JA, González Alcorta MJ, Miranda Romero LA, Carrillo Domínguez S, Castillo Domínguez RM. 2016. Sustitución de aceite de soya por aceite de atún en la dieta de pollos como alternativa para enriquecer la carne con ácidos grasos omega-3. *Interciencia.* 41(12):851-856. https://www.redalyc.org/articulo.oa?id=33948806009.

MAZA S, Rosales M, Castro R. 2003. Efecto de un proceso de lixiviación ácida salina sobre la calidad del surimi de *Dosidicus gigas* "Pota". *Boletín de Investigaciones Ins. Tec. Pes. Perú*. 5: 81-88. ISSN: 1023-7070. http://repositorio.itp.gob.pe/handle/ITP/103.

MONTAÑO MIE, Hernández GLA, Lomelí MH, Mesías DFJ, Ávila AA. 2015. Caracterización del consumidor de calamar gigante en Baja California Sur, México. *Ciencia y Tecnología Agropecuaria. 16*(1):41-50. ISSN: 0122-8706. http://www.scielo.org.co/scielo.php?script=sci_arttext&pid=S0122-87062015000100004&lng=en&tlng=es

MORALES BJE, Gonzáles AMJ, Castillo DRM, Prado ROF, Hernández VX, Menconi A, Téllez G, Marshal HB, Carrillo DS. 2013. Fatty Acid Deposition on Broiler Meat in Chickens Supplemented with Tuna Oil. *Food and Nutrition Sciences*. 4(9A1):5. ID:36099. https://doi.org/10.4236/fns.2013.49A1003

NABER EC. 1979. The effect of nutrition on the composition of eggs. *Poultry Science*. 58: 518-528. ISSN: 0032-5791. https://doi.org/10.3382/ps.0580518

NORMA Mexicana NMX-FF-079-SCFI-2004. Para: Productos avícolas – Huevo fresco de gallina – Especificaciones y Métodos de Prueba (Cancela a la NMX-FF-079-1991). Secretaría de Economía, Ciudad de México, México. Publicada en el Diario Oficial de la Federación del 19 de octubre de 2004.Disponible en: http://dof.gob.mx/nota_detalle_popup.php?codigo=5049464

NORMA Mexicana NMX-FF-127-SCFI-2016. Para: Productos avícolas – Huevo fresco de gallina – Especificaciones y Métodos de Prueba. Secretaría de Economía, Ciudad de México, México. Publicada en el Diario Oficial de la Federación del 24 de octubre de 2016. Disponible en: www.dof.gob.mx.

NORMA Oficial Mexicana NOM-062-ZOO-1999. Especificaciones Técnicas para la Producción, Cuidado y Uso de los Animales de Laboratorio. Publicada en el Diario Oficial de la Federación del 18 de junio de 2001. http://www.ibt.unam.mx/computo/pdfs/bioterio.NOM-062.pdf.

SAS Institute Inc. 2004. SAS/STAT ® 9.1 User's Guide. Cary, NC: SAS Institute Inc. ISBN: 1-59047-243-8. https://www.sas.com/es_mx/home.html

STADELMAN WJ, Pratt DE. 1989. Factors influencing composition of the hen's egg. World's. *Poultry Science Journal*. 45(3): 247-266. https://agris.fao.org/agris-search/search.do?recordID=GB9103232

TOYES VE. 2016. Aprovechamiento de subproductos marinos para la alimentación de camarón de cultivo y gallinas ponedoras. Tesis Doctoral. México, Centro de Investigaciones Biológicas del Noreste, Baja California, México. http://cibnor.repositorioinstitucional.mx/jspui/handle/1001/215.

WATERS. Water AccQ-Tag Chemistry Package. 1993. Manual Number WAT052874. Millipore Corporation, Milford, MA, USA. www.waters.com.