



## EFFECT OF TOTAL CHOLESTEROL AND FATTY ACIDS IN EGGS OF LAYING HENS BASED ON PERUVIAN ZAPALLO CREOLE MACREE FLOUR (*Cucurbita maxima Duch*).


Rojas Martel Gabriel\*

### Summary


To evaluate the effect of different levels with four concentrations of inclusion and a control (0), 4, 7 and 10 % Creole macree zapallo seed flour (*Cucurbita maxima duch*) on the total cholesterol and fatty acids of the eggs of laying hens, 120 Indian hens of the vanaraja species were located for 80 days, according to a completely random design. Simple classification analysis of variance was applied, except for the ethereal extract concentration, where factorial analysis was used with four treatments and 20 repetitions. The concentration of ethereal extract in the egg increased due to the effect as the evaluated weeks passed and the different concentrations of HDSdZMC inclusion (32.97 to 37.19). The oilseed enriched the egg in octadecanoic, oleic, linoleic, and  $\alpha$ -linolenic fatty acids; While it reduced the amount of arachidonic acid. The lowest ratio of saturated/polyunsaturated fatty acids and Om6/Om3 was observed, also the inclusion of this supplement decreased the total cholesterol in the egg with respect to the control. It is recommended to include 7 % to 10 % of Creole macree zapallo seed meal in the diets of laying hens to increase the ether extract, beneficial fatty acids, reduce total cholesterol and harmful fatty acids in the eggs of Creole laying hens, (fowl).

**Keywords:** Zapallo macree creole, cholesterol, fatty acids, sedes, laying hens, vanaraja.

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\*Graduated from the Faculty of Agrarian Sciences, from the professional career of agro-industrial engineering. Hermilio Valdizán National University: Av. Universitaria N° 601-607 Cayhuayna – Pillco Marca, Huánuco-Perú. Rojas Martel, Gabriel:  <https://orcid.org/0000-0001-9581-6659>  
Gmail: unhevalgab.edu@gmail.com

### \*Corresponding Author:

\*Graduated from the Faculty of Agrarian Sciences, from the professional career of agro-industrial engineering. Hermilio Valdizán National University: Av. Universitaria N° 601-607 Cayhuayna – Pillco Marca, Huánuco-Perú. Rojas Martel, Gabriel:  <https://orcid.org/0000-0001-9581-6659>  
Gmail: unhevalgab.edu@gmail.com

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## INTRODUCTION

The zapallo, scientifically known as *Cucurbita maxima mach*, belongs to the botanical family of Cucurbitaceae. It is a plant native to Central America and Mexico, but it is distributed throughout the world. Requires fertile, well-drained, moisture-rich soil, a sunny, wind-protected location (Sanchez et al., 2019). The pumpkin macre (*Cucurbita maxima* Duch) in Peru carried out various research works on the management of the cultivation of (ZM), currently its consumption has been increasing due to the great demand and acceptance by consumers both nationally and internationally due to the impact of flavor and nutritional value (Cerna and Reymundo, 2021). Zapallo (*Cucurbita maxima* Duch) has a high nutritional value of pulp and seeds such as carotenoids, lutein, Vitamin A, Vitamin C, starches, long-chain polyunsaturated oils; Becomes competitive and sustainable (Rodriguez et al, 2018). The same variety was cultivated in Huanchag -Panao region of Huánuco, around populated centers such as Cochatama among others, its production is normal in large quantities, it is accustomed to the use of furrows of wide length, however, the low availability of agricultural areas limits the execution of furrows, with widely separated distances. production is higher, it has a high nutritional value of the pulp and seeds such as carotenoids, lutein, Vitamin A, Vitamin C, starches, long-chain polyunsaturated oils; It becomes competitive and sustainable it has a high nutritional value of the pulp and seeds such as carotenoids, lutein, Vitamin A, Vitamin C, starches, long-chain polyunsaturated oils; Becomes competitive and sustainable (Claudia and rivera, 2022)

The egg has great nutritional value, since it is rich in proteins, fats, fat-soluble vitamins and minerals. Its inclusion in the daily diet is very important for human health. This is related to the detrimental effect of cholesterol and other lipids that it possesses (Simopoulos, 2002). Dietary supplementation of laying hens with organic selenium can lead to the production of value-added eggs. Increased the mineral content in the eggs, it has varied, from not being detected in the control periods of both trials, to 0.14 µg/g and 0.39 µg/g, during the supplementation period of each trial (Rodriguez-Alfaro, et al., 2019).

*Cucurbita maxima* Also known as zapallo macre in Peruvian, it is one of the most consumed vegetables in the world. The subcontinent has been mentioned in the traditional system of medicine, in Indian for

the treatment of various diseases (Kistler et al. 2015; Rajasree et al. 2016). Zapallo has been reported by several biologics. Activities as antioxidant due to the high content of β-carotene, which, in turn, improves immunity and helps decrease the incidence of heart disease and cancer. In addition, it is cultivated globally for many purposes, ranging from livestock feed to consumption and industrial and decorative with the shell as; Mates or dishes for family use and animals. The largest producers are India. United States, Canada, China and Peruvian (Badr et al. 2011; Kistler et al. 2015; Rajasree et al. 2016). *Cucurbita maxima* is very popular in the Peruvian and Indian system of medicine such as Ayurveda, Siddha, Unani and Homeopathy. Traditionally, different parts of the plant. Of *Caesalpinia bunduc* are used as an anthelmintic, febrifuge, tonic and vesicant. The bark is anti-periodic, rubefacient and to counteract toothache. Various classes of phytochemicals, such as flavonoids, diterpenes, and steroids, have been isolated from different species of the genus *Caesalpinia* (Badami et al., 2003).

In the proximal analysis obtained from the seeds, the amount of proteins ( $29.79 \pm 0.66$  to  $39.56 \pm 0.78$  % ms) and lipids ( $30.4 \pm 0.8$  to  $40.8 \pm 2, 5$  % ms), presenting levels of carbohydrates that oscillate between  $5.91 \pm 0.94$  to  $7.15 \pm 0.71$  % ms and with saturated fatty acids in a range between 19.63 to 25.0 % and unsaturated between 38.8 to 52.395 %. Among the latter, palmitic C16:0 (13.04 to 15.30 %), stearic C18:0 (6.49 to 9.81 %) stand out; n9 oleic C18:2 (27.16 to 38.30 %), linoleic C18:2 (37.84 to 52.59%) and arachidic C20:0 (0.53 to 0.78%). The researchers conclude that these dried seeds provide an important quantity of proteins and lipids rich in unsaturated fatty acids (Bloeck et al., 2012). The nutritional status of experimental animals at the end of the recovery stage (from 16 days to 32 days) showed an effect, but it was not significant for body weight and length, however, for tail length there was a significant difference ( $p=0.028$ ), and also for liver weight ( $p=0.016$ ). The consumption of pumpkin seed meal (*Cucurbita maxima*) on the nutritional status of the experimental units subjected to malnutrition had an effect, evidenced by the morphometric and biochemical indicators (basurco, 2019).

*Cucurbita maxima*, It is one of the vegetables belonging to the Cucurbitaceae family and has a relatively high economic importance (F. Gemrot et al., 2006; M. During et al., 2014). The seeds have

around 22 to 64 % lipids in their composition (YM Younis et al., 2000; J Jiao et al., 2014). These lipids being rich in monounsaturated and polyunsaturated fatty acids, such as oleic, linoleic and linolenic acids (M Doring et al., 2014).

Pumpkin seed oil has been widely used as an edible oil and contains several bioactive compounds, such as phytosterols (M. Nishimura et al., 2014; BB Rabrenovic et al., 2011), phenolic (L. Rezig et al., 2012), antioxidants, (L. Rezig et al., 2012), tocopherols (M. Nishimura et al., 2014; G. Procida et al., 2013) and small levels of carotenoids (M. Nishimura et al., 2014; X. Wang et al., 2017; G. Procida et al., 2013). On the other hand, the peel of *Cucurbita maxima*, which is often not used, contains significant amounts of carotenoids (M. Kreck et al., 2006; MY Kim et al., 2012). Carotenoids and tocopherols are lipid soluble compounds, vitamin precursors.

The egg is a basic product of the family basket that is above other food products, due to its cost and its high nutritional value, since it contains calcium, iron, vitamins A and D (Gomez, 2013; Leguizamon, 2014), are very useful for people's health, which is why it is one of the most widely consumed products in the country. As far as quality is concerned, there are some problems associated with egg production such as Salmonellosis, which is present in chickens and eggs, making it very difficult to eliminate, it is only possible through ultraviolet processes in some of the individuals (Garcia, 2016). On the other hand, there are problems of cracks that affect the product due to the entry of material foreign to it. There are also problems related to deficiencies in the nutrients of the egg, double yolk, eggs without yolk, with traces of blood or meat material inside it, all this related to the mechanisms of raising and maintaining animals in confined places without the due compliance with regulations (Boerjan, 2015).

The rice powder incorporated up to 18 % in the diet of hens in production adding multienzymatic complex does not affect the productive performance. The use of saponified marigold at levels of 20 ppm in diets with a high content of rice powder improves the pigmentation of the egg yolk. The best pigmentation of egg yolk was obtained with 40 ppm of the saponified marigold pigment, regardless of the levels of rice powder (basurco, 2019). The reduction of cholesterol in the egg is a complex task, since this substance is essential for the development of the embryo. Various drugs that cause adverse reactions in birds have been used to

reduce harmful lipids in the egg (Yin et al. 2008). However, the most objective and healthy solution is the incorporation of functional foods into the diet, rich in phytosterols, dietary fiber and essential fatty acids that also provide omega 3 and omega 6 fatty acids.

(Ayerza and Coates, 2000) have managed to lower total cholesterol and enrich eggs with omega 3 and omega 6 essential fatty acids, by including levels of oilseeds such as rapeseed (*Brassica napus*) and chia (*Salvia hispanica*), respectively.

Pumpkin is a cucurbit that has seeds rich in protein, polyunsaturated fats, phytosterols and dietary fiber (Martinez, 2009). In laying hens, the reduction of harmful lipids and an increase in serum essential fatty acids are observed when including four levels of HSC in the diets, without affecting the main productive indicators (Martinez et al., 2010).

The objective of this study was to evaluate the effect of the different concentrations (4 levels) of inclusion (0, 4, 7 and 10 %) of Creole macre squash seed flour (HDSdZMC) on the total cholesterol and fatty acid profile of eggs from Indian laying hens.

## MATERIALS AND METHODS.

The experiment was carried out in the small town of Cochatama at an altitude of 2685 m asl, Huácar district, Ambo province, Huánuco region - Perú. The average relative humidity was 68 %, the average minimum temperature was 18 °C and the average maximum temperature was 22 °C. The pumpkins of the macree creole variety were collected from the farm, then they were transferred and stored in open fields for their subsequent extraction of the seeds, then the samples of whole pumpkin seeds from different batches of species (*Cucurbita maxima*) were mixed shower).

### Composition and contribution of the diets.

The diets were formulated according to the requirements to reduce costs and benefits in feeding and inclusion levels (Martinez et al., 2012). The treatments consisted of four diets, with a control (0), 4, 7 and 10 % seed meal of the Creole macree zapallo variety (HDSdZMC). The ingredients used to prepare each diet are shown in Table 1.

### experimental procedure.

The experiment was carried out in a rectangular-shaped house made of rustic material (wall) with a 6m x 9m tin roof, with four divisions, where three hens were housed separately. The birds received

100 g of food/hen/day. Water was supplied by three drinkers. Which three hens were evaluated, feeding with 4, 7 and 10 % of Creole macree zapallo seed

**analytical determinations.**

From week \*25 to week \*30, 21 eggs/treatment were collected. The buds were extracted from the albumen and weighed in blocks. Next, they were frozen at -20 °C until their subsequent analysis in the instrumentation analysis laboratory.

The ethereal extract of the collected egg yolks was determined according to the AOAC (nineteen ninety five). To avoid the formation of trans fatty acids, anhydrous ethyl ether was considered as a

meal. The chickens did not receive medicines or veterinary care during the investigation carried out

solvent due to its lower boiling point (Martinez, 2009). To eliminate the residual anhydrous ethyl ether in the samples, a buchi rotary evaporator was used with a speed of 90 rpm, with T° regulated at 38 °C.

Once the samples were mixed in 1:50v/v anhydrous ethyl ether, in week \*30 the total cholesterol in the yolk was determined using enzymatic kits. A biotech SL 400 X 266 UV/VIS spectrophotometer was used. Total, AG was quantified according to the procedure of (AOAC, 2002).

**Table 1.** Nutritional value of diets in % (wet basis)

Raw material in (%)	*Different concentrations of zapallo Macree Creole seed flour (%)			
	0	4	7	10
yellow cornmeal	58.90	58.40	57.99	56.37
Macre pumpkin seed flour	0.00	4.00	7.00	10.00
Dicalcium phosphate	1.32	1.32	1.32	1.33
calcium carbon	9.65	9.63	9.60	9.60
BTH (Antioxidant)	0.01	0.01	0.01	0.01
DL-Methionine	0.21	0.21	0.22	0.23
L-lysine	0.06	0.07	0.08	0.08
premix	1.00	1.00	1.00	1.00
Zeolite	1.22	0.89	0.76	1.50
Contributions calculated in (%)				
ME (MJ/kg DM)	12.60	12.60	12.63	12.76
Crude protein (bp)	19.00	19.00	19.00	19.00
Lysine (Lys)	0.96	0.96	0.96	0.96
methionine	0.72	0.72	0.74	0.75
methionine + cystine	0.83	0.83	0.83	0.83
Calcium	4.22	4.22	4.22	4.22
Available Phosphorus (P)d	0.65	0.65	0.65	0.65
ethereal extract	2.50	3.60	4.70	6.60
Crude fiber (Cf)	3.50	3.80	4.30	4.50

Table 1 shows the amount of vitamins in zapallo Macree Creole flour (HDSdZMC) in kg.

To carry out the analysis of the diets, a GC chromatograpu Gas Chromatograph – 2010 was used. The equipment was controlled by a GC Chemstation data operator, version A.09.03. Fatty acids in the form of methyl esters were separated on a HP-23 cis\trans capillary column (60 m x 250 µm ID x 0.25 µm film thickness). The transformation of the fats into their respective methyl esters was verified in thin layer chromatography (AOAC, 2002). The determinations were made in the laboratories of the University of the Instrumentation Analysis Research Center; The preparation of the samples was carried out at the same university. The analyzes of ethereal extract, ethereal and profile of fatty acids in the yolks were carried out tenfold; Total cholesterol was determined quintuplicate therein.

A completely randomized design was carried out with simple classification analysis of variance, except for the ethereal extract variable, for which it was used (ANOVA) for 6 x 4 treatments (six weeks and treatment four) of (HDSdZMC). Where necessary, Duncan (2015) was used to determine the differences between means; All statistical analysis data was processed using IBM SPSS statistical software, version 22 for analyses.

**RESULTS AND DISCUSSIONS.**

Table 2 shows the relationship (P < 0.05) of the weeks under study with the inclusion concentrations of (HDSdZMC) of the ethereal extract in the yolk of the eggs of Indian laying hens. The concentration level was more notorious with

the inclusion of (HDSdZMC) and with the increase of the weeks studied.

By including the concentration of 10 % of (HDSdZMC) in the diets of the hens, it contributed a greater amount of ethereal extract to the diets (6.60 %) (table 1). In such a way it increased by effect after spending the weeks of (32.97 to 37.19)

According to Martínez et al., 2012 mentions that, by including a higher concentration (HSC) it demonstrated the increase of the ethereal extract in the yolk from (31.90 to 36.15 %). In polyunsaturated fatty acids in the seed zapallo macree creole, it was possible to determine a greater incorporation of ethereal extract in the yolk. In this way, the total serum lipids in the hens did not show statistical differences between treatments ( $P < 0.05$ ), the presumed suspicion is the high concentration of unsaponifiable material in the pumpkin seed carried out in the company. San Antonio de los Banos Cuba (Martinez et al., 2011). Silva (2018) refers to The inclusion of 1.5 % sacha inchi oil in the diet of laying hens influences a higher content of omega-3 fatty acids in the egg. According to Gable (2016) mentions that the omega 3 and omega 6 fatty acids present in flaxseed and sunflower seed with 13.5 % of diets do not alter the quality of the eggs of laying hens, considering these as a beneficial product for feeding human beings.

As the weeks go by, the hens determine a higher concentration of ethereal extract in the yolk, with respect to the increase in egg size, weight and size of the yolk (Martinez 2009 and Silva 2018). In this way, the maintenance of the bird with greater body weight, in the transport of fat in very low density lipoproteins increases with greater incorporation of lipids into the yolk of the eggs studied (Grobas et al., 2001).

Total cholesterol in egg yolk from Indian hens was higher ( $P < 0.05$ ) for the control (0) compared to 4, 7 and 10 % inclusion of HDSdZMC. The mg/egg and mg/g of cholesterol have decreased by including HDSdZMC as a fed supplement (table 3).

Feeding laying hens with the *Arthrospira maxima* cyanobacteria supplement increased egg weight, yolk color, proteins, minerals, bioactive compounds such as carotenoids, also decreased fat and cholesterol, and increased protein and mineral levels such as potassium, iron and phosphorus (Parra et al., 2017).

In the physicochemical analyzes of the squash flour (HZ) they obtained; fibers 2.44 %, ash 6.59 %, protein 4.63 %, fats 1.82 %, humidity 13.24 %, energy 320.02 kcal/100mg, total carotenoids 76.4 mg/Kg and carbohydrates 71.28 % (Rivadeneira et al., 2019).

**Table 2.** Different concentrations of inclusion of HDSdZMC and the weeks that the concentration of ethereal extract of the eggs of the laying hens were evaluated during six weeks studied are shown.

Different concentrations of zapallo macree creole seed meal (%)					
WEEKS	CONTROL	4	7	10	SE ±
*25	32.86aa*1	34.93a*2	35.86a*3	37.01a*4	0.11***
*26	32.97bb*1	34.92b*2	34.86b*3	36.72bb*4	
*27	32.89cc*1	35.07cc*2	34.79c*3	36.97cc*4	
*28	32.12d*1	34.09d*2	35.60d*3	37.19d*4	
*29	33.33e*1	34.36e*2	34.99e*3	35.94ee*4	
*30	33.32f*1	34.36f*2	34.98f*3	35.88f*4	

With duplicate letters it is observed that they differ  $P < 0.05$  (Duncan 2015) \*\*\*  $P < 0.0011$   
 Evaluated weeks = \*25, \*26, \*27, \*28, \*29, \*30.

Control and different concentrations of the HDSdZMC = 4, 7 and 10.

With different letters they are averages = yy\*1, bb\*1, cc\*2, dd\*1, bb\*4.

**Table 3.** Maximum amount of cholesterol in the yolk of the chicken eggs under study fed with different concentrations of HDSdZMC.

Maximum amount of cholesterol in yolk.	Different concentrations of zapallo macree creole is (%)				
	0	4	7	10	SE ± sig
mg/g egg	279.00a1*	250.00b2*	247.00b2*	253.00b2*	0.47***
mg/g yolk	16.95a1*	14.22b2*	14.29b2*	14.67b2*	0.18***



a1\*, b2\* With letters, number and asterisk means difference in the same row differ at  $P < 0.05$

By including HDSdZMC, cholesterol reduction is achieved in the eggs of the laying hen studied. The reduction of cholesterol in the yolk could be determined by the presence of unsaturated fatty acids in the HDSdZMC (Martinez et al., 2010). determined that linoleic (13894.00 mg/100 g) and oleic (8616.98 mg/100 g) fatty acids in dry pumpkin seed flour reduced the serum circulation of low-density lipoproteins (LBD), as well as total cholesterol; Perhaps it could influence the lower amount of total cholesterol in the yolk of the eggs of the laying hens evaluated.

of the company San Antonio de los Baños Cuba, pumpkin seeds of the *Cucurbita maxima* variety were collected, they were taken from five different batches, which C-88 is rich in phytosterols (233.08 mg/100 g) demonstrated in said investigation by (Martinez et al., 2011). Betasitosterol and campesterol have a structure similar to that of cholesterol, in such a way, in dry pumpkin flour it contributes to the reduction of total serum cholesterol in chickens, it is for this reason a possible suspicion in its reducible effect in the chicken egg using the food supplement (Martinez et al., 2010; Liu et al., 2010), The reduction of the waxy substance (cholesterol) in the egg is visualized from the sixth week of experimentation with high levels of phytosterols. Thus, it demonstrates the hypocholesterolemic effectiveness of the sterols found in the *Cucurbita bicifolia* seed. Elkin and Lorenz (2009) mentions a low incorporation of sterols in the egg yolk, also the reduction of cholesterol in the yolk, esterification by the enzyme 3-hydroxy-3-methylglutaryl-CoA reductase in the inhibition.

According to (Martinez, 2009), dietary fiber in pumpkin seed flour 41.4 % could lower cholesterol in egg yolk. (Ayerza and Coates 2000), The inclusion of chia seed diets of laying hens up to 28 %, it was possible to visualize the decrease in cholesterol in the yolk, which is related to dietary fiber and the high percentages of unsaturated fatty acids in the aforementioned diets. The hypocholesterolemic characteristics of dried pumpkin seed flour, the total cholesterol in the yolk was not gradually reduced (Ayerza and Coates, 2000; Elkin and Lorenz, 2009; Liu et al., 2010). Mention similar or similar versions in their results. Cholesterol is the third type of essential lipid for birds, cholesterol synthesis accumulates in the liver, apparently to create homeostasis, and it is

(Duncan 2015) \*\*\*  $P < 0.001$  in the identified table.

Deposited in the egg for the embryo, that is, where the embryo sac develops (Yin et al., 2008). It is worth mentioning that it also increases in the ethereal extract in the yolk of eggs with HSC levels increases lipids (total cholesterol).

Table 4 shows the AG profile in the yolk of the eggs of laying hens fed with different concentrations of HDSdZMC. Palmitic (HZ33) is the most representative saturated fatty acid in egg yolk, which increased when HDSdZMC concentrations were included. Also the octadecanoic (HZ35) showed high values in concentrations such as: control 0 % (g), 4 % (e) a greater amount of difference is observed, then it decreases 7 % (f) and progressively rises from 10 % HDSdZMC.

The oleic fatty acid (Z16:1n-9), the most abundant monounsaturated in the yolk, presented values higher than the control (0), that is, it increased progressively when including up to 10 % of HDSdZMC. Among the PUFA, linoleic acid (HZ16:2n-6) was progressively increased in laying hens' yolk. From 0 % to 4 % a great difference is observed, therefore, from 7 to 10 % the  $\alpha$ -linolenic polyunsaturated fatty acid (HZ16:3n-3) slowly increases. According to table N° 4, more detail is observed with significant differences ( $P < 0.05$ ) compared to the control (0).

Thus, arachidonic acid decreased with different inclusion concentrations of HDSdZMC macree pumpkin seed meal. The important relationship Om6/Om3 and AGS/PUFA (table 4) was lower ( $P < 0.05$ ) with 4, 7 and 10% of HDSdZMC with respect to the control (0).

The *Cucurbita maxima* seed is privileged in a group of oilseeds that have essential fatty acids, which are considered in omega 3 and omega 6, depending on the species and varieties of pumpkins, squashes (Martinez, 2009).

The eggs of Indian hens (birds) fed with 10 % HDSdZMC were enriched in 619 mg of linoleic acid/100 g of yolk with respect to the control (0) (table 4). Caston and Leeson (1990) They acquired similar results in their study carried out on omega-6 in the yolk of eggs, using feed supplements for hens with flax and rapeseed, it was effectively effective. In such a way, it demonstrates that the concentrations of inclusion of omega 6 in the diets

influences the percentage incorporated in the feeding of the hens.

According to Pilco and Natali in (2015), it says that the inclusion of two levels of flaxseed on the productive and quality parameters of chicken eggs, of two treatments 4 % (T2) and 8 % (T3), by including flaxseed increased the protein, dry matter, weight and ether extract, T3 presented the best results, with the inclusion of flaxseed the nutritional quality of the eggs of Indian laying hens was improved, from the economic point of view the production of eggs with the inclusion of flaxseed in the diet is viable up to 8 % presented maximum results in protein and ethereal extract.

The arachidonic fatty acid content (ZH17:4n-6) in the egg yolk began to decrease with the addition of 4% (0.46), in such a more noticeable and effective way, it is observed with the addition of 7 and 10 % HDSdZMC (0.38 and 0.38 g/100 g of yolk) compared to the control (0.47g/100 g of yolk) (table 4).

(Martinez et al., 2010) He demonstrated in his research the reduction of arachidonic acid in the blood serum of chickens, by including up to 10 % dry pumpkin flour (HSC). In addition, the inclusion of chia and flax seed in laying hens showed similar results by decreasing the arachidonic content in the egg (Ayerza and Coates, 2000).

**Table 4.** Fatty acids are observed in the yolk of the eggs of laying hens under study, fed with different concentrations of HDSdZMC (mg/100g).

Fatty acids	Different concentrations of zapallo macree criole seed (%)				SE ± sig
	0	4	7	10	
HZ30	4.88±0.31e	4.65±0.30ef	4.94±0.21e	3.96±0.21f	(*1)
HZ31	121.29g	128.70f	134.77f	141.63e	0.27 (*2)
HZ32	25.26	26.69	24.99	25.05	1.05
HZ33	8347.00f	8592.00ef	8757.00e	8788.00e	43.84 (*1)
HZ34	65.38	76.88e	74.84e	79.75e	2.89 (*1)
HZ35	3012.00g	3463.00e	3161.00f	3297.00ef	91.09 (*1)
HZ24	29.93ab±2.92	23.92b±3.69	27.12ab±2.91	36.85a±3.12	(*2)
HZ10:1	23.26f	22.17f	25.64e	24.29e	0.92 (*1)
HZ11:1n-7	891.92	997.63	975.00	910.00	32.28
HZ16:1n-9	13010.00g	14298.00f	14480.00f	15027.00e	193.80 (*3)
HZ16:1n-5	602.07	629.18	648.59	557.98	13.52
HZ19:1	81.57g	88.91af	91.51e	95.56f	1.44 (*3)
HZ16:2n-6	6122.00f	6146.00f	6537.00e	6795.00e	103,100 (*3)
HZ16:3n-3	347.70g	804.69f	970.00e	998.00e	88.79 (*3)
HZ17:4n-6	469.19e	464.15e	383.51f	376.39f	18.93 (*1)
HZ21:6n-3	116.00	119.00	130.00	133.00	16.49
Σ MUFA	13234.00g	14569.00f	14589.00f	15224.00e	212.00 (*2)
Σ AGS	11503.00f	12616.00e	12008.00ef	12089.00ef	239.23 (*2)
Σ PUFA	7112.00g	7598.00f	8042.00ef	8370.00e	164.12 (*3)
AGS/PUFA	1.61ef	1.66e	1.49f	1.50ef	0.05 (*1)
ΣOm-6	6379.00f	6412.00f	6727.00ef	6992.00e	119.88 (*2)
ΣOm-3	399.12f	852.65e	980.08e	996.08e	90.33 (*3)
Om6/Om3	13.82e	7.52f	6.86f	7.01f	1.39 (*3)

<sup>e,f,g</sup> Alphabets in the same row that differ at P < 0.05 are means (Duncan 2015)(\*1)P < 0.05, (\*2)P < 0.01, (\*3)P < 0.001.

HDSdZMC: seed flour of zapallo Macree Creole

AG: fatty acid

SFA: saturated fatty acids

MUFA: monounsaturated fatty acids

PUFA: Polyunsaturated fatty acids; Om-6: Omega 6 ; Om-3: Omega 3.

The different concentrations increased in the inclusion of HDSdZMC progressively of the omega-3 in the egg, from 399.12 mg/100 g of yolk for the control to 996.08 mg/100 g of yolk for the

inclusion of 10 % of HDSdZMC (table 4). In the antecedents of the reports about foods rich in alpha-linolenic fatty acids in diets of laying hens, the tendency to enrich the egg with omega 3 is

mentioned (Caston and Leeson 1990; Ayerza and Coates 2000; Rowghani et al. 2007). The sum of the MUFA and PUFA in the Indian egg yolk (table 4) increased with respect to the level of inclusion of HDSdZMC, due to the concentration of oleic, linoleic and alpha-linolenic fatty acids in the HDSdZMC, and to the serum circulation. and its incorporation into the eggs of laying hens (Martinez et al., 2010).

Thus, docosahexaenoic fatty acid (DHA) does not increase in the yolk when HDSdZMC concentrations increase, as shown in (table 4). Despite the fact that its higher concentration in the blood serum of laying hens with the concentrations of HDSdZMC (Martinez et al., 2010). According to (Chauca, 2017) investigated probiotic uses and amino acids and identified that it does not necessarily generate satisfactory results in breeding domestic birds due to their physiological characteristics, the incorporation of fatty acids resulting from elongation and desaturation is poor. (Ayerza and Coates, 2000) carrying out more studies found an increase in DHA when using up to 28% of chia seeds when feeding it. The high concentration of omega 3 in the chia seed (65 %) an inclusion level could influence related research.

Eggs from laying hens, fed up to 10 % HSC, have approximately 186 mg of omega 3. This makes it a functional food that can contribute to meeting the omega 3 requirements in humans (Martinez et al., 2012)

When comparing and relating omega6/omega3 (n6/n3) it decreased as the concentration of HDSdZMC increased. It was observed that it decreased from 13.82 for the control to 6.86 for the diet added with 7 % of HDSdZMC (table 4). World Health Organization (2008) On saturated fats and oils in human nutrition, in his report, he recommends a ratio of n-6/n-3 of 5-10/1 to prevent atherosclerosis and cardiovascular risk in the future. The values presented in the (table 4) are within the proposed reference range. Simopoulos (2002) recommends ratios of 2.1/1 to enhance the increase in omegas.

## CONCLUSION.

When including flour of seed of zapallo macree (HDSdZMC) cholesterol decreased with respect to the control. It is recommended to include 10 % Creole macree zapallo seed meal in the diets of Indian laying hens to increase ether extract and beneficial fatty acids, reduce total cholesterol and harmful fatty acids in poultry eggs. By including up

to 10 % of Creole zapallo seed meal in the diets of Vanarajas laying hens, the ethereal extract increased with interaction as the weeks passed and the beneficial fatty acids. Thus, total cholesterol and harmful fatty acids in poultry eggs were reduced.

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