

***Sargassum* EN LAS DIETAS DE GALLINAS RHODE ISLAND MEJORA LA CALIDAD DEL HUEVO Y SU FUNCIONALIDAD POR ENRIQUECIMIENTO CON IODO**

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RESUMEN. *Sargassum* es un alga muy abundante y de fácil acceso en las zonas costeras, su composición química permite utilizarla como suplemento en dietas para gallinas de postura. México es el quinto productor y el principal consumidor a nivel mundial de huevo, uno de los alimentos más nutritivos debido a la excelente calidad de su proteína y a sus propiedades multifuncionales. El objetivo de este estudio fue evaluar si la incorporación de *Sargassum* en la dieta de gallinas de postura Rhode Island mejora la calidad nutricional del huevo e incrementa el contenido de yodo. El estudio se realizó con 135 gallinas Rhode Island de 24 semanas de edad, distribuidas en 3 tratamientos: 0%, 3% y 6% de *Sargassum*. Diariamente se registró el consumo de alimento y producción de huevo, durante un periodo de 12 semanas. Al finalizar el experimento se evaluó en el huevo, el peso, la altura de albúmina, el color de la yema, el grosor del cascarón; y se cuantificó el contenido de colesterol, triglicéridos y yodo. El color de la yema se incrementó conforme aumentó la concentración de *Sargassum* en la dieta, obteniéndose con el abanico DSM un valor de 8.9 para el tratamiento con 6% del alga; asimismo, se incrementó el peso del huevo. La concentración de colesterol en el huevo del tratamiento con 6 % de *Sargassum* fue menor (226 mg/100 g), que en el resto de los tratamientos. No se encontró diferencia significativa en el sabor del huevo de las gallinas alimentadas con pienso enriquecido con *Sargassum*. Por lo que incorporar 6% de harina de *Sargassum* en la dieta de las gallinas ponedoras es adecuado para incrementar el peso del huevo y el contenido de yodo, reducir la concentración de colesterol y favorecer la coloración amarilla de la yema, sin afectar negativamente las variables productivas, calidad física y sabor del huevo, por lo tanto, se considera que el Sargazo tiene potencial para la industria avícola.

Palabras clave: Sargazo, gallinas ponedoras, color de la yema, colesterol, triglicéridos

***Sargassum* in the diet of Rhode Island hens improves egg quality and functionality by iodine enrichment**

ABSTRACT. *Sargassum* is an abundant seaweed that is easy to harvest in coastal areas; its chemical composition allows using it as a dietary supplement for laying hens. Eggs are among the most nutritious feed items due to their high protein quality and multifunctional properties. Mexico is the fifth largest producer and first consumer of eggs worldwide. The objective of the present study was to assess the incorporation of *Sargassum* meal into the diet of Rhode Island laying hens diet improves the nutritional quality of eggs and increases iodine content. The study was conducted with 135 Rhode Island hens distributed in 3 treatments: 0%, 3%, and 6% *Sargassum*. Daily feed consumption and egg production were recorded, for 12 weeks. At the end of the experiment, egg weight, albumen height, yolk color, and shell thickness were measured; and egg cholesterol, triglyceride, and iodine content were quantified. Yolk color increased with *Sargassum* concentration in the diet, reaching a value of 8.9 of the DSM yolk color fan with 6% *Sargassum* meal. Egg weight also increased. Egg cholesterol content with 6% *Sargassum* meal was lower (226 mg/100 g) than in the rest of the treatments. No significant differences were found in the flavor of eggs from hens fed *Sargassum*-enriched feed. These findings suggest that supplementing laying hen feed with 6% *Sargassum* meal increase the egg weight and iodine content; reduce egg cholesterol content, and enhance yolk color without adversely affecting the production parameters, egg physical quality and egg flavor. Therefore, *Sargassum* to have potential for the egg-production industry.

Keywords: Sargazo, laying hens, yolk color, cholesterol, triglycerides

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INTRODUCTION

The genus *Sargassum* comprises seaweeds of great ecological and economic importance for their abundant biomass in coastal areas worldwide (Graham & Wilcox, 2000). Its chemical composition includes polysaccharides (alginates, fucoidan, laminaran), secondary metabolites (terpenoids, fluorotannins, polyphenols, volatile hydrocarbons), and minerals that can be used as fertilizers, fodder, and raw materials for the phycocolloid, textile, and pharmaceutical industries (Mascheck & Baker, 2008).

Within the context of animal nutrition, the use of seaweeds as feedstuff has been a common practice for many centuries. For instance, on the Scottish Island of North Ronaldsay, in the Orkney archipelago, sheep have grazed on a nearly exclusively seaweed diet since the 19th century (The Orkney Sheep, 2022). In the 20th century, farm animals in Iceland were fed with dried seaweeds, during the winter. During the Second World War, they were used as feedstuff in Europe due to the scarcity of other sources (Makkar *et al.*, 2016). As a result, seaweeds were given renewed attention recently (Ribeiro *et al.*, 2021).

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The research in the use of *Sargassum* in animal nutrition, such as ruminants (e.g. Choi *et al.*, 2020a,b), sheep (Ellamie *et al.*, 2020), laying hens (Al-Harathi & El-Deek, 2011), broilers (Erum *et al.*, 2017), amongst others, continues to address its uses and benefits as a supplementary element in their diets. However, in México this type of research has been scarce (Marín-Álvarez *et al.*, 2003, 2009; Casas-Valdez *et al.*, 2006 a,b), particularly in the region of the Gulf of California where the resource is abundant (see Casas-Valdez *et al.*, 2016). While there is also a great amount of research in the addition and enrichment of eggs with supplementary elements around the world (Lesnierowski & Stangiers, 2018; Sarlak *et al.*, 2020 and citations therein); there is a lack of studies on the enrichment of eggs with microelements and their functionality in México and the Gulf of California.

Among its mineral composition, iodine content (I) is particularly high, given that these seaweeds can easily assimilate it from seawater (Ribeiro *et al.*, 2021). As a trace element, iodine is essential in human nutrition, mostly due to its role in thyroid hormone synthesis, and its deficiency constitutes a global issue that affects around 2 billion people worldwide (Krela-Kazmierczak *et al.*, 2021).

The egg white is an excellent natural source of high-quality protein, which is rich in essential amino acids. In addition, the yolk is a source of antioxidants, aromatic amino acids, carotenoids, vitamins, phospholipids, and proteins that provide nutritional value to the egg and act as health promoters in its consumers (Lesnierowski & Stangierski, 2018). Chicken eggs are natural sources of iodine (9.5 to 58 µg/100g) (Krela-Kazmierczak *et al.*, 2021). Furthermore, Abeyrathne & Ahn (2015) have pointed out that eggs are rich in anti-bacterial, anti-carcinogenic, and immunostimulatory compounds.

Rodríguez (2018) found that meal from the brown algae (*Macrocystis pyrifera*) supplemented in the diet of laying hens increased iodine content in eggs. Therefore, this author considers that iodine-fortified egg may be an alternative for populations in areas with iodine deficiency that suffer diseases known as iodine deficiency disorders (IDD) such as goiter, among others (Arrizabalaga *et al.*, 2015).

Particularly, when *Sargassum* meal is used as a supplement in the diet of laying hens, it improves egg quality and reduces its cholesterol and triglyceride content (Al-Harathi & El-Deek, 2012; Carrillo *et al.*, 2012a,b). These components are important because high contents produce dyslipidemias associated with cardiovascular diseases — the main cause of death in the Mexican population (INEGI, 2016).

From the economic and food-production perspectives, the poultry industry is of great importance worldwide and is growing steadily. In 2018, Mexico ranked first in egg consumption worldwide, with 23 kg/year consumed per capita (INA, 2018). There is a

growing demand for poultry feed worldwide, which currently accounts for about 47% of total farming consumption (Kulshreshtha *et al.*, 2020). For this reason, the identification of new feed resources would play a central role in the sustainable development of the livestock industry (Makkar *et al.*, 2016).

Given the importance of poultry production and its high consumption, coupled with the high abundance of *Sargassum* in coastal areas, its potential use as a supplement to the diet of Rhode Island laying hens was explored to improve the nutritional quality of eggs and increase its iodine content for further human consumption.

MATERIALS AND METHODS

The brown algae *Sargassum* spp. was harvested in Piedras Coloradas (24°20'51.32" N, 110°40'24.94" W) La Paz Bay, Baja California Sur, Mexico. Samples were collected manually in the intertidal zone; then, they were spread over a cement plate, dried by sunlight exposure for 48 hours, and ground in a hammer mill until the meal was obtained (400 kg). One kilogram of meal was used to perform chemical analysis in triplicate.

Chemical composition of *Sargassum*. Standardized AOAC methods (2000) were followed for bromatological and mineral analysis: moisture content (60° drying oven to constant weight, method 976.05), ash (calcination at 550°C in muffle, method 923.03), crude fiber (Soxhlet apparatus, method 962.09), ether extract (Soxhelt apparatus, method 920.39), nitrogen content (micro-Kjeldahl, method 976.05); protein content was calculated applying a conversion factor of 6.25. Ca, Mg, Na, K, Fe, Zn, and Cu were determined by acid digestion, followed by atomic absorption spectrophotometry analysis (method 969.08). Phosphorus concentration was determined by colorimetry (method 999.11). For iodine determination, method 925.26 of the AOAC (2007) was used. Gross energy was determined using a Parr calorimetric pump.

Experimental Assay. The experiment was carried out at the “El Platanito” commercial farm, located in La Paz, B.C.S., during summer-autumn (September to December 2019), when the climate is dry warm with rainfall. During the experiment, the temperature of the coop where hens were housed was recorded daily with a digital thermometer (VWR Cat. No. 620-2023). We used 135 in-production laying hens (Rustic Rhode Island) aged 24 weeks, randomly distributed across three treatments: control (0%) and diets supplemented with 3% and 6% of *Sargassum* meal. Each treatment included 45 hens with 5 replicates of 9 hens each, housed in cages. Feed and water were provided *ad libitum* for 12 weeks.

Diets were formulated using the Nutrition program (version 3.2) for laying hens, according to the NRC (1994), to cover their nutritional requirements and partially replace corn, soybean, and calcium carbonate with *Sargassum* meal. The experimental diets were

subjected to bromatological analysis following the methodology described above. In addition, diets were similar in protein and energy. Feed intake and egg production were recorded daily and summarized per week according to Quintana (2011), obtaining average values for these parameters. The feed conversion ratio was calculated.

Physical quality of the egg. At the end of the experiment, six eggs per replica, 30 per treatment, were randomly selected to determine egg weight, yolk color, albumen height, and then calculate the Haugh units. Albumen height was measured with a micrometer (Baxlo Haugh Micrometer); yolk color was determined with a DMS Color Fan against a white background and under illumination with artificial white light. Finally, Haugh units were calculated considering albumen height corrected by egg weight, using the following formula:

$$UH = \frac{100 \log H - (\sqrt{G(30W^{0.37} - 100)} + 1.9)}{100}$$

Where: H = height of the albumen in mm, W = weight of the egg in grams, G = constant 32.2, LogH = natural logarithm of the height of the albumen.

Determination of egg cholesterol, triglycerides, and iodine content. To quantify cholesterol, triglyceride, and iodine contents, at the end of the experiment six eggs per replicate were randomly selected for each treatment (30 per treatment). The shell was removed, the white and yolk were mixed into a beaker and homogenized with a Braun blender for 60 seconds approximately. Next, the contents of each sample were poured into glass flasks with a plastic cap, then freeze-dried in a USIFROID, SMH-50 Lyophilizer; afterward, three samples were obtained from each for analysis. Cholesterol was determined in triplicate by an enzymatic-colorimetric method (CHOP-PAP, RANDOX Biochemical Kit); triglycerides, by the enzymatic-colorimetric method (GPO-PAP, RANDOX Biochemical Kit); and iodine was determined using the method 992.22 of the AOAC (2007).

Acceptance test. According to Watts *et al.* (1992), a hedonic acceptance test was used at the end of the experiment. Ten eggs per treatment were cooked in a Teflon pan at low heat with a bit of oil, without adding salt. Samples of each treatment were placed on a white plate (each sample was identified with a three-digit code); a portion of bread was placed on each plate, and a glass of natural water was on one side of the plate. The test involved 25 non-trained panelists of both sexes who were regular egg consumers. They were instructed to consume a portion of a sample, choose one of the options in the acceptance questionnaire, then consume a portion of bread and some water before evaluating the following sample, to eliminate any residual taste. Three acceptance grades were included: like, neither like nor dislike, dislike.

Statistical analysis. Data from productive parameters, egg physical quality, and egg chemical composition were analyzed through a one-way analysis of variance (ANOVA) with the Statistical 8.0 program; the Tukey's test ($p < 0.05$) was used for the multiple comparisons of means (Zar, 2010). A correlation analysis was conducted between the ambient temperature recorded in hen coops and feed intake per hen per week, as well as in relation to egg production, using the Excel 2016 statistical package.

RESULTS

Chemical composition. The chemical composition of *Sargassum* meal is shown in Table 1. Table 2 shows the ingredients and chemical composition of experimental diets. The amount of *Sargassum* meal varies according to the inclusion percentage in feed, and the amount of the remaining ingredients is proportional to reaching 100%. All diets were similar in protein and energy.

Productive parameters. The daily feed intake in the control treatment ranged between 75 and 104 g/hen, in the 3% *Sargassum* treatment it was greater than 100 g/hen, except in weeks 4 and 6, and in the 6% treatment it ranged between 74 and 116 g/hen. No significant differences were found among treatments ($p > 0.05$) (Table 3). However, at weeks 4 and 6, there was a general reduction in consumption per hen.

Weekly egg production (%) fluctuated from 62% to 74% in the control treatment, from 64% to 75% with the 3% *Sargassum* feed, and from 61% to 71% with the 6% *Sargassum* feed. Concerning egg weight, the 6% *Sargassum* feed was associated with the highest egg weight ($p < 0.05$) compared with the other treatments, with a difference of 2.6 g. No significant differences were found between treatments (0%, 3%, 6%) in feed intake (consumption/hen/day), egg production, and feed conversion (Table 3).

The temperature recorded during the experiment varied from 16°C to 36°C. The most significant difference between the lower and the higher temperature

Table 1. Chemical composition of *Sargassum* spp. meal

Chemical component	<i>Sargassum</i> meal
Moisture (%)	8.37
Crude protein (%)	6.26
Ash (%)	33.64
Ether extract (%)	0.58
Crude fiber (%)	8.88
Nitrogen-free extract (NFE) (%)	42.28
Minerals	
Calcium (g/100 g)	5.12
Phosphorus (g/100 g)	0.35
Chlorine (g/100 g)	8.65
Sodium (g/100 g)	1.68
Potassium (g/100 g)	3.80
Manganese (ppm)	56.27
Zinc (ppm)	14.22
Copper (ppm)	2.86
Selenium (ppm)	0.025
Iodine (ppm)	0.028

Table 2. Experimental diets supplemented with *Sargassum* meal and chemical composition (%).

Ingredients	Control	3% <i>Sargassum</i> meal	6% <i>Sargassum</i> meal
Ground corn	63.9	59.6	54.1
Soybean	20.5	20.9	22.1
<i>Sargassum</i> meal	0	3	6
Calcium carbonated	9.6	9.2	8.8
Premix Poultry Vitamins and minerals	4	4	4
Bovine Tallow	2	3.3	5
Composition			
Moisture	10.7	10.7	10.4
Crude protein	15.9	15.2	15.8
Ash	10.5	10.8	10.7
Ether extract	3.7	3.7	7.1
Crude fiber	3.5	3.9	4.3
Nitrogen free extracts	55.7	55.6	51.8
ME (kcal/kg)	2854	2826	2982

during the day was observed at week 5, with minimum and maximum temperatures of 17°C and 33°C, respectively. The mean temperature increased at weeks 2 and 3 (33°C) and subsequently decreased steadily to 24°C, at week 10.

The correlations of temperature with daily feed consumption/hen for weeks 1 to 12 (Fig. 1) and weekly egg production (Fig. 2) showed that temperature is a central factor influencing the variables mentioned above since a correlation coefficient of $R^2 = 0.79$ was obtained in relation to daily feed intake per hen and of $R^2 = 0.92$ in relation to egg production; these correlations were inverse.

Egg physical quality. No significant differences were found in albumen height and Haugh units between the *Sargassum*-enriched feed and the control diet. However, yolk color increased significantly ($p < 0.05$) with higher *Sargassum* content in the feed, with a value of 8.9 read in the DSM Fan for 6% *Sargassum* treatment (Table 3).

Egg chemical quality. Regarding the egg chemical composition, no significant differences were found in triglyceride concentration between the three treatments. However, cholesterol concentration was lower ($p < 0.05$) in eggs of hens fed 6% *Sargassum* feed, while egg iodine concentration increased along with *Sargassum* concentration in feed; these differences were statistically significant ($p < 0.05$) (Table 4).

Acceptance test. The results of the evaluation by panelists regarding the degree of acceptance of egg taste of the three treatments (control, 3% and 6% *Sargassum*) showed no significant differences ($p > 0.05$).

DISCUSSION

Sargassum chemical composition. The fat content of algae is considered to be similar to that of cereals (rice and wheat) and legumins (rice beans, mung bean and alfalfa) since they contain less than 3% (Heuzé *et al.*, 2016; Yang *et al.*, 2022), which is a favorable characteristic, since because of this it poses no rancidity issues and can be preserved for a long time.

It should be noted that ash content (inorganic matter) is high, particularly in brown and green algae (Prian *et al.*, 2020). In this case, a 33.6% value was obtained, higher than in alfalfa and oat hay, which have approximately 7-10% ash content (Benitez, 2022; Zhou *et al.*, 2018) and wheat straw (11%) (Katoch, 2022), which are used as ingredients in animal feed.

The nitrogen-free extract was found in a very high percentage, 42.3%, which comprises mainly polysaccharides such as alginates, laminaran, fucosa and mannitol (Makkar *et al.*, 2106; Vijay *et al.*, 2017), that are an important part of the animal nutrition. The value obtained was similar than that reported by Choi *et al.* (2020 b) for *S. fusiforme* (43%), but higher than Vázquez-Delfin *et al.* (2021) for pelagic *Sargassum* spp. (15%).

Calcium content was 5.1%, a lower percentage than the values of 8.1% for *S. fulvellum* and 8.9 % in *S. fusiforme* reported by Choi *et al.* (2020a,b). Calcium is important for shell formation, for which *Sargassum* is a suitable alternative to enrich hen feed. Sodium content was 1.7%, below the reported values of 3.2% and 3.4% for *S. sinicola* and *S. herporizum*

Table 3. Productive parameters and physical quality of eggs from Rhode Island laying hens fed with different levels of *Sargassum* meal for 12 weeks.

	Control 0%	3% <i>Sargassum</i>	6% <i>Sargassum</i>
Feed intake (g hen ⁻¹ , d ⁻¹)	97.2 ± 7.65	103.1 ± 7.69	104.1 ± 8.04
Egg production (%)	69.5 ± 3.39	69.6 ± 3.80	66.4 ± 3.75
Egg weight (g)	62.5 ± 0.55 ^a	65.3 ± 0.23 ^{ab}	67.1 ± 2.63 ^b
Feed conversion ratio (g/g)	2.4 ± 0.15	2.4 ± 0.10	2.5 ± 0.29
Albumen height (mm)	8.6 ± 0.51	8.2 ± 0.29	8.9 ± 0.44
Haugh units (HU)	94.9 ± 3.36	94.4 ± 2.33	92.8 ± 2.40
Yolk color	7 ± 0.0 ^a	8 ± 0.0 ^b	8.9 ± 0.14 ^c

a,b,c Different letters in each column indicate a significant differences ($p < 0.05$).

by Carrillo *et al.* (2008); and 27 g/kg⁻¹ of *S. fulvellum* and 34.4 g/kg⁻¹ for *S. fusiforme* (Choi *et al.*, 2020a, b). Phosphorus concentration was 0.35%, greater than that reported by Benitez (2022) and Zhou *et al.* (2018) for alfalfa (0.20%) and oat hay (0.18%). Therefore, this element can also be added to the list of ingredients in laying hen feed that enrich the egg.

Regarding iodine, this study recorded a value of 2.8 µg/100g, while Oyesiku & Egunyomi (2014) reported 0.04 mg/100g for a mix of pelagic *S. natans* and *S. fluitans*, Choi *et al.* (2020) reported 151.5 mg/kg in *S. fusiformis*. Other brown algae have higher iodine concentrations, such as *Laminaria japonica* (3.6 µg/100 g), and *Macrocystis pyrifera* (8.3 µg/100 g) (Rodríguez, 2018). Seaweeds can concentrate iodine up to 30 000 times of seawater (Zava & Zava, 2011). Smyth (2021) mention that brown algae contain higher concentrations of iodine (2500-10000 µg/g DW), than red (20-200 µg/g DW) and green (30-185 µg/g DW) algae. As a trace element, iodine is essential in human nutrition, mostly due to its role in the thyroid hormone synthesis (O'Kane *et al.*, 2018).

Productive parameters. No significant differences in feed intake and egg production were found among the different treatments in this study, in contrast with the information reported by others authors (Carrillo *et al.*, 2008, 2012b) who used *Sargassum* spp. meal. No significant differences were found in the dietary conversion factor, showing that the inclusion of *Sargassum* in the diet does not affect this variable, consistent with the findings reported by Carrillo *et al.* (2008, 2012a).

The weight of the egg increased as the concentration of *Sargassum* in the diet increased, finding significant differences in the treatments 3% and 6% with respect to the control. Choi *et al.* (2018) also found that the egg weight was higher when a brown algae *Undaria pinatifida* was added to the diet of laying hens. This may be due to the fact that the algae favored a higher protein availability, water and minerals, as well as a higher retention and absorption of food in the gastrointestinal tract, as suggested by Kulshreshtha *et al.* (2014).

The results of the correlation analysis between ambient temperature and feed intake and egg production, suggested that lower temperatures favor an increase in both parameters within the temperature limits of the experiment. The optimal temperature for the performance of adult laying hens ranges between 19 and 22°C; temperatures above or below this range would need thermoregulation (Lin *et al.*, 2006) since temperature may produce heat stress, affect performance, and blood chemical alterations leading to death (Kilic & Simsek, 2013; Barret *et al.*, 2019). Jacob *et al.* (2000) mention that good egg quality requires avoiding prolonged periods with temperatures above 30°C.

Egg physical quality. Several authors (Carrillo *et al.*, 2008, 2012b; Al-Harathi & El-Deek, 2012; Abu *et al.*, 2019) indicate that the inclusion of brown algae (*Sargassum sinicola*, *S. cinereum*, *S. dentifolium* and *S. fusiforme*) in the diet of hens improve albumen height and yolk color. However, no significant differences were observed in albumen height between treatments in the present study. The values obtained with 3% and 6% *Sargassum* (8.2, 8.9 mm) were similar to those obtained when using *Sargassum* concentrations of 2%, 4%, 6%, and 8% in another study (8.9, 8.8, 8.7, and 8.8 mm, respectively) (Carrillo *et al.*, 2012a).

According to Menezes *et al.* (2012), high temperatures deteriorate albumen quality. In the experiment reported here, the mean temperature was 28.4°C; but during weeks 1 to 6, the mean temperature was 31.3°C, whereas in weeks 7 to 12, it was 25.5°C. However, albumen height values obtained in this experiment showed no adverse effect, most likely due to the management given to hens since at higher temperatures, fans were turned on to improve air circulation, and water was sprayed with a sprinkler.

Yolk color increased with higher *Sargassum* concentration in the diet, with DSM Fan values of 8 and 8.9 for the 3% and 6% *Sargassum* treatments, respectively. The color of the egg yolk is one of the main criteria used by the consumer to judge its quality, a more intense orange color is more attractive to them. Our results showed higher values than those obtained

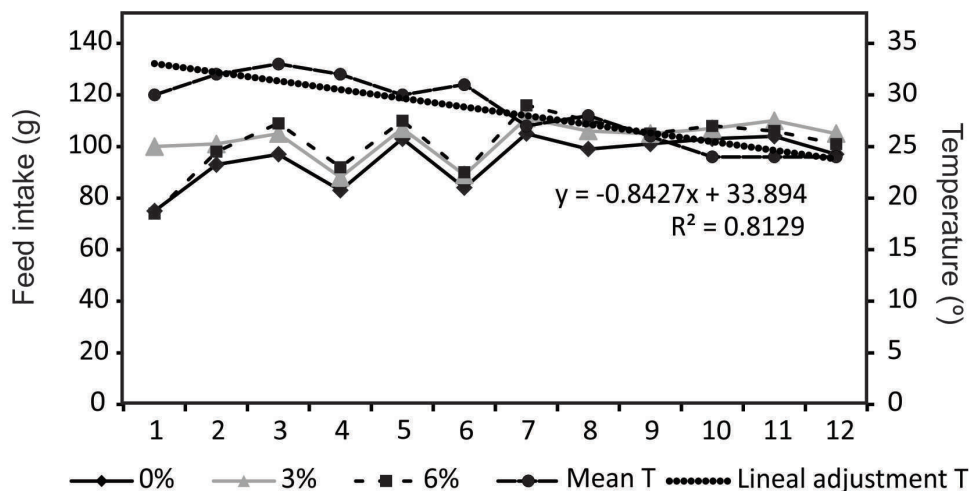


Figure 1. Correlation of feed intake per hen/day for weeks 1 to 12 with mean temperature: Control (0%), *Sargassum* (3%), and *Sargassum* (6%).

ned by Carrillo *et al.* (2012a) (7.4 to 7.9) when using *Sargassum* concentrations of 2% to 8% in hen diets. Al-Harthi and El-Deek (2012) found that carotene and lutein plus zeaxanthin increase in egg yolk when 3% and 6% of *S. dentifebium* was added to the diets of laying hens. Also, Abu *et al.* (2019) mentioned that brown seaweeds (*S. cinereum*) improve the yolk color of laying quails. According to Carrillo *et al.* (2012b) the increase in yolk color is due to carotenoid composition, mainly xanthophylls and fucoxanthines from brown algae. In this regards Vieira & Ventura (2019) mention that brown algae have high concentrations of carotenoids.

Egg chemical quality. Cholesterol concentration in eggs of hens fed 6% *Sargassum* (226 mg/100 g)

was lower than in the rest of the treatments. This is consistent with the studies of Carrillo *et al.* (2008, 2012a, 5% a 26%), Al-Hathi & El-Deek (2012), Rizk (2017), Abu *et al.* (2019), and Kulshrestha *et al.* (2020) who point that the inclusion of algae in the diet of posture hens reduces egg cholesterol content. Some compounds such as sterols, polysaccharides, and fatty acids, interfere with egg cholesterol synthesis (Hassan *et al.*, 2005; Yuan, 2008). These results are of importance for producing eggs with lower cholesterol levels, particularly when there is a growing preference to consume low-cholesterol food.

Our results showed no significant differences in triglyceride concentration; however, lower triglyceride levels were recorded in eggs from hens fed 3% and

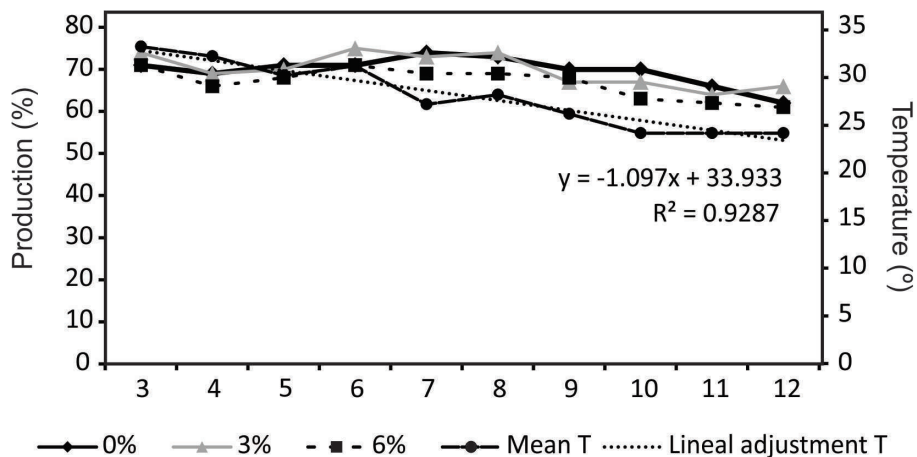


Figure 2. Correlation of egg production per week with mean temperature: Control (0%), *Sargassum* (3%), and *Sargassum* (6%).

6% *Sargassum* diets (257 mg/g) in relation to control (265 mg/g). Al-Hathi & El-Deek (2012) did find a significant reduction in triglyceride content when incorporating 3% and 6% of *S. dentifebium* in the diet.

Iodine concentration in eggs of hens fed 3% and 6% *Sargassum* diets was significantly higher than in the control group; iodine increased with higher seaweed concentration in feed (57.7 µg/100 g, 75.7 µg/100 g, and 96 µg/100 g for 0%, 3%, and 6% *Sargassum* feed, respectively). This is consistent with Rodríguez (2018), who supplemented hen feed with 2%, 4%, and 6% *Macrocystis pyrifera* and recorded high egg iodine contents of 146 µg/100 g, 171 µg/100 g, and 199 µg/100 g, respectively. The concentration of iodine in this brown alga is 830-1530 µg/100 g (Rodríguez, 2018), while Teas *et al.* (2004) reported concentrations of 39 µg/g for *Sargassum* spp. When diets were supplemented with mineral (calcium iodate) used 2, 4, 8 mg/kg to enrich the iodine content of the egg, the iodine content in the egg increased (0.73, 0.97, 1.56 mg/kg) as it increased in the diet (Sarlak *et al.*, 2020); however, the concentrations obtained were lower than with algae. Although the values recorded in this study were lower than the ones reported by Rodríguez (2018), iodine is present in significant concentrations in our research, which are higher than what it has been reported for fresh whole eggs that are 9.5 – 57.6 µg/100g (Fordyce, 2021).

Iodine deficiency is a global issue and affects around 2 billion people worldwide, with pregnant women as a high-risk group (Krela-Kazmierczak *et al.*, 2021). A diet deficient in iodine can lead mental retardation, hypothyroidism, congenital anomalies, gout or low IQ (Santos *et al.*, 2019). Iodine is an essential element for the synthesis of thyroid hormones; its deficiency in humans causes adverse effects known as “Iodine Deficiency Disorders” (IDD) (Arrizabalaga *et al.*, 2013).

The intake of iodine-rich food and iodized salt is the only route of administration of iodine in humans (Krela-Kazmierczak *et al.*, 2021; Smyth 2021). For this reason, any measures to promote a reduced salt intake also led to a reduction in iodine intake and an increase in IDD. In addition, there is limited consumption of marine products — the other source of iodine — in areas far away from the coast (Krela-Kazmierczak *et al.*, 2021). An alternative to compensate for iodine deficiency could be the consumption of seaweed, particularly brown algae containing high iodine levels, but these algae are not generally consumed as food. Therefore, increasing iodine content in eggs is a convenient way of supplying it to the population, as egg consumption is widespread. Surai & Fisinin (2015) and Lesnierowski & Stangierski (2018) have stressed the positive functional aspects of egg enrichment with microelements.

Acceptance. The supplementation of *Sargassum* in the diet of laying hens did not convey a different taste relative to the control diet. Similar results were

Table 4. Cholesterol, triglyceride, and iodine concentrations in lyophilized eggs of Rhode Island laying hens fed with different levels of *Sargassum* meal for 12 weeks.

Treatment	Cholesterol (mg/100 g)	Triglycerides (mg/g)	Iodine (µg/100 g)
Control 0%	249.9 ± 8.5 ^a	264.9 ± 11.12	57.7 ± 1.53 ^a
3% <i>Sargassum</i>	248.8 ± 13.5 ^a	257.0 ± 34.46	75.7 ± 2.08 ^b
6% <i>Sargassum</i>	226.3 ± 21.2 ^b	257.4 ± 37.35	96.0 ± 2.65 ^c

a,b,c Different letters in each column indicate a significant differences (p < 0.05).

obtained by Rodríguez (2018) when using the brown algae *Macrocystis* as a supplement in hen feed. Furthermore, there are records that when ingredients such as fish oil are added to hen feed, the organoleptic quality of eggs is reduced from the unpleasant fish flavor and odor, attributed to lipids and amines (Honkatukia *et al.*, 2005; Castillo-Badillo *et al.*, 2005; Carrillo *et al.*, 2008).

Studies of this type are important because *Sargassum* is an abundant resource that is readily available in coastal areas, where the general population and poultry producers can use it in the proportions indicated to supplement the diet of posture hens.

CONCLUSIONS

The addition of up to 6% of *Sargassum* seaweed to the diet of rustic Rhode Island laying hens is a natural alternative to increasing the egg weight and yolk color, lowering cholesterol content, and rising iodine concentration of eggs without negatively affecting the production variables of hens, egg quality, or egg taste. Thus, it could be considered as a functional feed, giving an additional value as food for humans.

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