

# EDUCATION AND PRODUCTION

## Duckweed, A Useful Strategy for Feeding Chickens: Performance of Layers Fed with Sewage-Grown Lemnacea Species

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**ABSTRACT** Layer performance and egg quality were assessed in hens fed sewage-grown *Lemna* species (duckweed) in order to examine the safety and efficacy of this plant as a feedstuff for poultry. Dried *Lemna gibba* was included in the diets of two commercial strains of laying hens at 0, 15, 25, and 40% inclusion. Egg production and egg weights were compared with those of hens fed a standard isocaloric and isonitrogenous control diet. At all levels of *Lemna*, hens maintained egg production and had mean egg weights similar to layers fed a control diet. Eggs from Leghorn hens fed 15 and 25% *Lemna* had higher protein content than control eggs. Also, the addition of *Lemna* to the diets significantly increased yolk pigmentation, an important commercial value for this plant. *Lemna* species may be a useful substitute for soybean and some fish meal in layer hen diets, especially in countries where some of these commodities are imported.

(Key words: layers, duckweed, egg quality, egg production, yolk pigmentation)

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### INTRODUCTION

The possibility of using sewage-grown *Lemna* species (duckweeds) in poultry feeding as a source of protein and energy in place of conventional ingredients appears to be of great potential importance. Duckweeds are the smallest, most simple of flowering plants. There are four common genera and more than 40 different species. Distribution is worldwide. Duckweed plants range in size from the giant

*Spirodella polyrrhiza*, reaching 1.5 cm, to the pin-size *Wolffia arrhiza*. These floating plants grow in dense clusters, forming blankets on the surface of nutrient-laden, open fresh water (Hillman, 1961).

Dried duckweed meal, which contains up to 40% protein, compares favorably with soybean as a source of plant protein (Porath *et al.*, 1979). The protein content of duckweed, however, responds quickly to the availability of nutrients in the aquatic environment. Consequently, duckweed grows slowly in clear, low nutrient waters and is high in fiber, ash, and carbohydrates, but contains relatively low protein (Muztar *et al.*, 1976). In contrast, duckweed grown on sewage lagoons grows rapidly and has a high protein content.

The nutritional value of duckweed as poultry feed has long been recognized (Lautner

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and Muller, 1954; Muzafonou, 1968; Abdulayef, 1969). More recently, Truax *et al.* (1972) have shown that dehydrated duckweed, when substituted for dried alfalfa meal at up to 5% of mixed poultry feeds, produces superior weight gain in chicks (up to 3 wk of age). This has been attributed to its well-balanced amino acid profile. In comparative feeding studies, chickens fed diets consisting of up to 10% duckweed consistently outperformed chickens fed diets containing similar percentages of alfalfa meal as well as chickens exclusively fed an "optimal" control diet (Muztar *et al.*, 1976).

Despite the variability of results reported, duckweed can successfully replace alfalfa in poultry diets. Although earlier studies have included duckweed in poultry feeds at relatively low concentrations (5% to 10%), their use of poor quality duckweed (high fiber and ash, and less than 20% protein) suggests that higher quality duckweed can be included in diets at higher levels. With higher quality duckweed (i.e., 30 to 40% protein, low ash, high carbohydrate) such as that obtained from regular harvesting of highly eutrophic waters, there is a good reason to think that duckweed may be substituted for not only alfalfa, but also soybean meal and fish meal. This study was designed to examine the safety and efficacy of duckweed meal as a source of protein and pigment for laying hens and to evaluate its impact on egg laying performance and egg quality.

#### MATERIALS AND METHODS

##### *Growth, Harvesting, and Drying of Duckweed*

*Lemna gibba*, a medium-sized duckweed, and *Wolffia arrhiza*, the smallest duckweed (.2 to .4 mm), were used. These two duckweed species were found growing naturally in tertiary effluent and lagoon runoff at the San Juan de Miraflores oxidation lagoons in the southern cone of Lima, Peru. Duckweed was manually harvested, using rakes, from the perimeter of the lagoons and transported wet to the Food Processing Plant at The Universidad Nacional Agraria (UNA) in Lima. There, duckweed was sun dried to approximately 40% moisture on concrete aprons. Drying was then completed to 10% total moisture using the Food Processing Plant's forced air oven for 15 to 30 min at 60 C. Complete sun drying was avoided to minimize

pigment loss due to ultraviolet exposure. This method ensured the retention of high xanthophyll levels (800 to 1,000 ppm as measured by the Purina Laboratories, Lima, Peru) in the finished duckweed meal. The dried *Lemna* was stored at room temperature in 200-L black plastic feed sacks to minimize protein and pigment loss. During the 4 mo of storage, no spoilage occurred.

Proximate analyses were determined by methods of the Association of Official Analytical Chemists (1970). In addition, duckweed was assayed at the Occupational Safety and Health Administration laboratories for the presence of heavy metals. The concentrations found were well below those permitted for human consumption.

##### *Metabolizable Energy of Duckweed*

The ME of duckweed was determined to be 2,000 kcal/kg in mature roosters, using the method described by Sibbald (1976), subtracting endogenous energy. This ME value was used in the formulation of the experimental diets in Experiment 1.

The ME was later estimated using young broilers (14 to 28 days of age) (Hill *et al.*, 1960). The new ME value obtained for broilers was 1,200 kcal/kg. This value was used in formulating diets for Experiment 2.

##### *Diet Formulation*

All diets were formulated to meet the National Research Council requirements (NRC, 1984). Ingredients were purchased locally from the Ralston Purina Company. Dried *Lemna gibba* and *Wolffia arrhiza* were finely milled before the preparation of the diet mixtures. All diets were formulated to be isonitrogenous and isocaloric except when differences in energy between diets was the variable being tested (Trial 2, Experiment 1). Mixing of diets was performed in the feed mill at UNA.

##### *Housing of Birds*

A temporary structure made of bamboo matting and fence posts was built to house the layers. Individual pens measuring 2.5 m  $\times$  3.0 m were constructed within the "hen house", using wooden frames and heavy commercial grade fishing nets. Each housing unit was provided with a feeder, an automatic waterer, five nests, and wood shavings for bedding.

TABLE 1. Percentage composition of diets fed to TOPAZ layers

Ingredients	Control (1)	Lemma, 15% (2)	Wolffia, 15% (3)
Yellow corn, ground	52.0	51.0	51.0
Wheat middlings	18.9	16.3	16.3
Fish meal, 65% CP	7.5	7.5	7.5
Soybean meal, 46% CP	10.9	..	..
Lemma, 33% CP	..	15.0	..
Wolffia	..	..	15.0
Hydrogenated fish oil	2.5	2.7	2.7
Limestone	7.5	6.9	6.9
Dicalcium phosphate	.38	.28	.28
Iodized salt	.08	..	..
Premix <sup>1</sup>	.30	.3	.30
Total	100.00	100.00	100.00
Calculated analysis			
ME, kcal/kg	2,800	2,800	2,800
CP, %	17.1	16.86	17.5
Lysine, %	.94	.96	.94
Methionine, %	.35	.36	.35
Methionine and cysteine, %	.62	.65	.62
Calcium, %	3.3	3.3	3.3
Available phosphorus, %	.38	.38	.39
Ash, %	11.26	13	10.4
Fiber, %	3.76	4.87	3.75
Fat, %	6.01	7	6.02
Sodium, %	.18	.38	.38

<sup>1</sup>Supplied per kilogram of diet: vitamin A, 10,000 IU; vitamin D<sub>3</sub>, 1,000 IU; vitamin E, 10 IU; vitamin K, 3 mg; choline, 225 mg; riboflavin, 4.5 mg; pantothenic acid, 6 mg; vitamin B<sub>12</sub>, .012 mg; niacin, 22.5 mg; vitamin C, 19.5; antioxidant (butylated hydroxytoluene), 120 mg; Mn, 60 mg; Zn, 30 mg; Cu, 1.5 mg; I, 1.5 mg; Co, .15 mg.

### Laying Hens

Two different commercial strains of laying hens were utilized in the experiments. TOPAZ layers (41 wk of age), a heavy breed of hens producing brown eggs were used<sup>3</sup> for the first experiment. In the second study, HyLine White Leghorn hens were used.<sup>4</sup> These lighter, more delicate hens, which produce smaller eggs at lower production, were obtained for the study at the age of 39 wk.

### Experiment 1

**Trial 1.** One hundred and fifty 43-wk-old TOPAZ layers were randomly distributed into groups of 10 according to weight and placed in 15 pens following a 2-wk preexperimental period. Three diets were used: a control (0% *Lemma*), a diet containing 15% *Lemma*, and the

third containing 15% *Wolffia*. The latter was used only for one observation period due to limited supplies caused by a change in the distribution of species of duckweed in the sewage lagoons. The diets were formulated to be isonitrogenous (17% CP) and isocaloric (2,800 ME kcal/kg). The experiment lasted 90 days, including an adaptation period of 14 days during which no experimental data were collected. The *Wolffia* treatment was shorter because of seasonal variation in its growth in the sewage lagoons.

During the 2-wk preexperimental period, egg production was carefully observed, and hens were reassigned to different pens to balance egg production among pens. During this period all hens received a control diet (Table 1). Feed and water were supplied for *ad libitum* access. Feed consumption was measured weekly by subtracting residual feed from the total feed provided. Hens began receiving experimental diets on Day 15. Two wk later, data collection began (Week 2, Table 3). Each diet group consisted of 50 hens (five units of 10 each). Feeding and watering, and feed consumption measurement protocols

<sup>3</sup>Donated by Avicola Hannan S. A., Pacific Breeders, Lima, Peru.

<sup>4</sup>Donated by Universidad Nacional Agraria, Lima, Peru.

TABLE 2. Composition of the diets fed to HyLine Leghorn layers

Ingredients and analyses	Control	Lemma	
		15%	25%
		(%)	
Lemma, 33% CP	0	15	25
Wheat middlings	3	2	..
Yellow corn, ground	62	54	48
Fishmeal, 65% CP	7	7	2
Soybean meal, 46% CP	9	..	..
Cotton paste	5	5	5
Brown sugar	5	5	5
Hydrogenated fish oil	0	4	6
Ca(HPO <sub>4</sub> ) <sub>2</sub>	..	..	1
Limestone	5	4	4
Oyster shell	4	4	4
Premix <sup>1</sup>	.30	.30	.3
Total	100	100	100
Calculated analyses			
ME, kcal/kg	2,836	2,840	2,840
CP, %	16	16	16
Lysine, %	.83	.85	.85
Methionine, %	.38	.38	.38
Methionine and cysteine, %	.62	.62	.62
Calcium, %	3.2	3.2	3.2
Available phosphorus, %	.35	.35	.35

<sup>1</sup>Supplied per kilogram of diet: vitamin A, 10,000 IU; vitamin D<sub>3</sub>, 1,000 IU; vitamin E, 10 IU; vitamin K, 3 mg; choline, 225 mg; riboflavin, 4.5 mg; pantothenic acid, 6 mg; vitamin B<sub>12</sub>, .012 mg; niacin, 22.5 mg; vitamin C, 19.5; antioxidant (butylated hydroxytoluene), 120 mg; Mn, 60 mg; Zn, 30 mg; Cu, 1.5 mg; I, 1.5 mg; Co, .15 mg.

were identical to those of the preexperimental period. Eggs were collected, weighed, and classified daily. During a monthly "sampling week" (the 4th wk of each 28 day experimental interval), all eggs were weighed and classified individually; randomly selected "standard size" (56 to 63 g) eggs were used for external and internal quality measurements. The parameters used to measure the quality were yolk pigmentation and individual weights. The pigmentation was measured using the Hoffmann-La Roche colorimetric fan.<sup>5</sup>

The units were cleaned daily; cleaning included change of water and turning of bedding. Layers were weighed individually at the end of every 28-day period.

**Trial 2.** One hundred TOPAZ layers from Trial 1 were used in this study. These layers were kept in their previous pen allocations (Trial 1) during introduction of new diets. The control group was maintained on the same control diet, and three new isonitrogenous diets were formulated.

The new diets contained: 25% Lemma (2,800 ME kcal/kg), 25% Lemma with a higher ME (2,900 kcal/kg), and 40% Lemma (2,800 ME kcal/kg). The study lasted 2 mo. No preexperimental or adaption periods were used, because these layers had already been on diets containing Lemma.

Four of the five control units from Trial 1 were chosen at random to remain as controls. In the same manner, six more units from the previous experiment were chosen and the new formulated diets were supplied. The study lasted 2 mo. Feed and water supply, feed consumption, egg handling, and pen procedures were the same as in Trial 1. Layer weights were recorded at the beginning and at the end of the study.

#### Experiment 2

Two hundred 41-wk-old Leghorn HyLine hens were distributed in the pens in groups of 10 according to weight. The diets used in this study were a control (0% Lemma), 15% Lemma, and 25% Lemma with an ME of 2,800 kcal/kg (Table 2). Each diet group consisted of 40 hens (four duplicates of 10 layers each). The study lasted 3

<sup>5</sup>Hoffmann-La Roche, Inc., Nutley, NJ 07110.

mo, including a 2-wk adaption period when no experimental data were collected.

Egg production balancing procedures were identical to those in Trial 1 of Experiment 1. Feed and water were supplied *ad libitum*, and feed consumption was recorded every 14 days. At the end of the preexperimental period, the hens received the experimental diets. Sampling weeks, pen procedures, and egg handling were also identical to those in Trial 1, Experiment 1. Hens were weighed at the beginning and at the end of the study.

### Egg Taste Panel

Sensory tests on eggs collected during the sampling periods in Experiments 1 and 2 were performed at the Organoleptic Laboratory at the UNA by well-trained panelists. Eggs were cooked in a variety of ways and were checked by the panelists for general appearance, odor, color, and taste.

### Egg Composition

Protein measurements were performed on randomly selected standard size eggs collected on the last sampling week in Experiment 2. The eggs were hard boiled to facilitate yolk and albumen separation (Bair and Marion, 1978), then stored at 4°C until assayed. Yolk and albumen protein was measured using standard micro-Kjeldahl techniques with bovine serum albumin as a standard. Shell calcium concentration was measured on standard size eggs collected from three sampling weeks in Trial 1, Experiment 1. The shells were digested with a HCl solution (68% vol/vol), and the grams of Ca/100g shell (%) were determined by atomic absorption spectroscopy (Tietz, 1980).

### Microbiological Testing of Eggs

Two hundred rectal swab samples (100 from control units and 100 from *Lemma* and *Wolffia* fed units) were analyzed for pathogens such as *Vibrio*, *Aeromonas* species, *Campylobacter*

*jejuni*, *Shigella*, and *Salmonella* species, using standard techniques (Annual Report, Diarrhea and Nutrition Project, 1984). Hens from all study groups over a 2.5-mo period (Trial 1, Experiment 1) were sampled for bacterial enteropathogens using rectal swabs. Swabs were streaked on plates of McConkey agar,<sup>6</sup> thiosulfate-citrate-bile-sucrose agar,<sup>6</sup> Butzler's agar,<sup>6</sup> and ampicillin blood agar<sup>6</sup> plates in order to isolate enteropathogens including *Vibrio* species, *Aeromonas* species, and *Campylobacter* species. Swabs were also incubated in Selenite F broth and streaked on *Salmonella-Shigella* agar in order to isolate *Salmonella* species. Suspicious isolates were picked for further identification by standard techniques (Annual Report, Diarrhea and Nutrition Project, 1984).

### Data Handling and Statistics

All data were collected in forms, which were then transferred to Lotus 1-2-3 worksheets<sup>7</sup> on a personal computer<sup>8</sup> and Bernoulli disks.<sup>9</sup> Data were calculated on a 28-day basis for each 10-hen unit, except for Period 1, where the first 2 wk (adaption period) were not included. Mean egg weights were calculated using the total number and weight of the eggs produced in each unit during each period (28 days). Conversions represent the total egg weight produced over the total food consumed over a 1-wk period (Experiment 1) or over a 2-wk period (Experiment 2). Statistical analysis of the data was performed using a one-way ANOVA based on STATPAC<sup>10</sup> and Student's *t* test.

## RESULTS

### Experiment 1

The inclusion of 15% *Lemma* and 15% *Wolffia* in the diets produced no significant differences in egg production, in feed conversion, or in mean egg weights when compared with those of the control group (Table 3). In the first experimental period (Week 2) there was a difference in consumption between the control and the *Lemma* 15% ( $P < .02$ ) and the *Wolffia* 15% group ( $P < .03$ ). When the initial and final periods (2 wk versus 10 wk) were compared, the control showed decreases in feed consumption ( $P < .04$ ) over time, but the *Lemma* 15% group showed only a slight decrease ( $P < .07$ ) in feed consumption. Egg number and egg weight parameters were not significantly different among the groups.

<sup>6</sup>Difco, Detroit, MI 48232.

<sup>7</sup>Lotus Development Corporation, Cambridge, MA 02138.

<sup>8</sup>IBM, Valhalla, NY 10595.

<sup>9</sup>IOMEGA Corporation, Ogden, UT 84403.

<sup>10</sup>Walnick Associates, Minneapolis, MN 55423.

TABLE 3. Performance of TOPAZ layers fed a control diet, a diet containing 15% *Lemna* species, or a diet containing 15% *Wolffia* species

Diet	Measurement	Week				
		2	6	10		
Control	Egg production, %	92.29 ± 4.44	91.50 ± 5.10	88.79 ± 5.71		
	Feed consumption, kg	.149 ± .003 <sup>a</sup>	.145 ± .004 <sup>a</sup>	.133 ± .003 <sup>b,x</sup>		
	Feed conversion, kg/kg	2.438 ± .090	2.404 ± .069	2.347 ± .094		
	Mean egg weight, g <sup>1</sup>	66.18 ± 1.71	65.96 ± 1.97	64.21 ± 1.99 <sup>y</sup>		
	Mean weight gain, g	163 ± 75 <sup>a,x</sup>	48 ± 68 <sup>b</sup>	-19 ± 86 <sup>c</sup>		
	Number of eggs/hen per week	6.460	6.405	6.215		
	Yolk pigmentation, 2 to 10 wk			9.43 ± 1.73 <sup>y,f</sup>		
<i>Lemna</i> , 15%	Egg production, %	93.71 ± .95 <sup>a</sup>	92.71 ± 1.67 <sup>ab</sup>	90.07 ± 2.12 <sup>b</sup>		
	Feed consumption, kg	.147 ± .004	.146 ± .005	.140 ± .006 <sup>y</sup>		
	Feed conversion, kg/kg	2.347 ± .044	2.376 ± .071	2.392 ± .087		
	Mean egg weight, g	66.76 ± 1.46 <sup>a</sup>	66.19 ± 1.62 <sup>ab</sup>	65.17 ± 1.03 <sup>b,x</sup>		
	Mean weight gain, g	117 ± 124 <sup>a,y</sup>	60 ± 106 <sup>b</sup>	31 ± 154 <sup>c</sup>		
	Number of eggs/hen per week	6.560	6.490	6.305		
	Yolk pigmentation, 2 to 10 wk			12.45 ± .80 <sup>x,e</sup>		
<i>Wolffia</i> , 15%	Egg production, %	89.71 ± 3.31	ND <sup>2</sup>	ND		
	Feed consumption, kg	.146 ± .004	ND	ND		
	Feed conversion, kg/kg	2.429 ± .118	ND	ND		
	Mean egg weight, g	67.10 ± 1.12	ND	ND		
	Mean weight gain, g	181 ± 115 <sup>x</sup>	ND	ND		
	Number of eggs/hen per week	6.280	ND	ND		
	Yolk pigmentation, 2 wk	12.79 ± .65 <sup>e</sup>				

<sup>a-c</sup>Means (± SD) within a row with no common superscripts differ significantly (P<.05).

<sup>e,f</sup>Means within two columns for the same trait with no common superscripts differ significantly (P<.05).

<sup>x,y</sup>Means (± SD) within a column for the same trait with no common superscripts differ significantly (P<.05).

<sup>1</sup>Mean egg weights represent the total kilograms produced per period divided by the total production number.

<sup>2</sup>ND = Not determined due to a change in the distribution of *Lemna* and *Wolffia* species in the lagoon.

In Trial 2, no significant differences were found between the control group and the hens fed 25% *Lemna* in any of the variables shown in Table 4. However, in the first period, feed consumption of hens fed the 40% *Lemna* diet decreased significantly from that of controls (P<.005). There was also a nonsignificant decrease in egg production in the first and second periods of the study between the control hens and those fed 40% *Lemna* species. Feed conversion values were not significantly different between the groups fed different diets. No differences in egg weight were seen throughout the study. Egg production and feed consumption declined over time.

The effects of dietary *Lemna* species levels on yolk pigmentation and egg quality are presented in Table 4. In Trial 1, pigmentation increased significantly (P<.001) when 15% *Lemna gibba* or 15% *Wolffia arrhiza* were included in the diets. Higher levels of *Lemna* species produced smaller, but still significant (P<.005), incremental changes in yolk pigmen-

tation when eggs from hens fed 25% *Lemna* species were compared with eggs from the 40% group. No differences were found in the calcium concentration of the shells among the groups fed with 0, 15, or 25% of *Lemna* in their diets; calcium concentrations were 43, 42.9, and 43%, respectively (25 eggs per group).

There was no difference in the rate of enteropathogens isolated from fecal samples collected from 100 layers receiving either *Wolffia* or *Lemna* and 100 layers fed a standard diet. *Campylobacter jejuni* was isolated from 18% of the controls compared to 24% of the layers consuming *Lemna* species. *Aeromonas* species were isolated at low rates in both layer groups: 1% of the controls versus 4% of the *Lemna* species group. *Salmonella* species were not isolated in either group.

#### Experiment 2

There was a gradual, but not significant, increase in the rate of egg production of the

TABLE 4. Performance of TOPAZ layers fed diets containing varying percentages of *Lemna gibba* at different energy levels comparable to a control diet

Diet	Measurement	Week	
		14	18
Control	Egg production, %	86.88 ± 3.87 <sup>x</sup>	84.46 ± 4.60
	Feed consumption, kg	.141 ± .003 <sup>a,x</sup>	.131 ± .004 <sup>b</sup>
	Feed conversion, kg/kg	2.494 ± .096 <sup>x</sup>	2.413 ± .092 <sup>x</sup>
	Mean egg weight, g <sup>1</sup>	65.40 ± 1.93	64.25 ± 1.58
	Mean weight gain, g		46
	Number of eggs/hen per week	6.081	5.913
	Yolk pigmentation, 11 to 18 wk		8.88 ± .70 <sup>x</sup>
<i>Lemna</i> , 25% (ME = 2,900 kcal/kg)	Egg production, %	86.96 ± 1.96 <sup>x</sup>	84.10 ± 2.68
	Feed consumption, kg	.138 ± .009 <sup>x</sup>	.131 ± .004
	Feed conversion, kg/kg	2.504 ± .112 <sup>a,x</sup>	2.473 ± .020 <sup>b,x</sup>
	Mean egg weight, g	63.31 ± .18	63.14 ± .389
	Mean weight gain, g		114
	Number of eggs/hen per week	6.088	5.888
	Yolk pigmentation, 11 to 18 wk		12.98 ± .95 <sup>y</sup>
<i>Lemna</i> , 25% (ME = 2,800 kcal/kg)	Egg production, %	89.64 ± 1.79 <sup>x</sup>	87.32 ± .54
	Feed consumption, kg	.141 ± .007 <sup>x</sup>	.132 ± .003
	Feed conversion, kg/kg	2.466 ± .044 <sup>x</sup>	2.382 ± .013 <sup>y</sup>
	Mean egg weight, g	63.96 ± .76	63.61 ± .91
	Mean weight gain, g		134
	Number of eggs/hen per week	6.275	6.113
	Yolk pigmentation, 11 to 18 wk		13.13 ± .71 <sup>y</sup>
<i>Lemna</i> , 40%	Egg production, %	82.86 ± .36 <sup>a,y</sup>	79.46 ± .89 <sup>b</sup>
	Feed consumption, kg	.123 ± .002 <sup>y</sup>	.125 ± .001
	Feed conversion, kg/kg	2.328 ± .003 <sup>b,y</sup>	2.476 ± .009 <sup>a,x</sup>
	Mean egg weight, g	63.82 ± 1.05	63.57 ± .19
	Mean weight gain, g		-118
	Number of eggs/hen per week	5.800	5.563
	Yolk pigmentation, 11 to 18 wk		13.39 ± .52 <sup>x</sup>

<sup>a,b</sup>Means (± SD) within a row with no common superscripts differ significantly (P<.05).

<sup>x-y</sup>Means (± SD) within a column for the same trait with no common superscripts differ significantly (P<.05).

<sup>1</sup>Mean egg weights represent the total kilograms produced per period divided by the total production number.

Leghorn layers during the first two periods in the control group during the 2.5 mo studied (Table 5). The group fed 15% *Lemna* also exhibited a gradual increase in production during the first two periods with a subsequent, but not significant, decline during the last period. Inclusion of 25% *Lemna* in the diet significantly decreased production during the last period (10 wk), over that of the control (P<.05). There were no significant differences in feed consumption and feed conversion between the control and the *Lemna* groups.

Egg number and total egg weight were maintained by the *Lemna* groups during the 1st 6 wk when compared with the control group (Table 5). However, the *Lemna* 25% group showed a decrease from the controls in these variables during the last period (P<.05 and P<.04, respectively). No changes in the mean egg weights were seen with increasing amounts

of *Lemna* species. All groups showed a gradual significant increase in the mean egg weight over time.

The effects of dietary *Lemna* on pigmentation in this study were similar to the results obtained in Trial 1, Experiment 1 (Tables 3, 4, and 5). Yolk pigmentation was greater in eggs from hens fed 15% *Lemna* than in eggs from the control group (P<.001). Eggs from hens fed 25% *Lemna* were significantly more pigmented than those from hens fed 15% *Lemna*. However, the rate of increment in pigment was much less marked than that between the control and the 15% group.

The chemical values obtained for protein on eggs collected on the last period of this experiment are presented in Table 6. Protein content increased significantly (P<.001) both in the albumen and in the yolk when layers were fed 15% *Lemna* or 25% *Lemna* compared with

TABLE 5. Performance of HyLine Leghorn layers fed control diets or diets containing 15 or 25% of *Lemna gibba* in the diets

Diet	Measurement	Week		
		2	6	10
Control	Egg production, %	76.96 ± 9.43	79.46 ± 8.65 <sup>xy</sup>	82.23 ± 6.73 <sup>x</sup>
	Feed consumption, kg	.123 ± .005 <sup>x</sup>	.122 ± .005	.120 ± .001
	Feed conversion, kg/kg	2.682 ± .380	2.500 ± .251 <sup>xy</sup>	2.308 ± .154 <sup>y</sup>
	Mean egg weight, g <sup>1</sup>	60.60 ± 1.20 <sup>b</sup>	62.09 ± 1.76 <sup>ab</sup>	63.65 ± 1.52 <sup>a</sup>
	Mean weight gain, g			149 ± 195
	Number of eggs/hen per week	5.388	5.563	5.756
	Yolk pigmentation, 2 to 10 wk			7.67 ± .82 <sup>x</sup>
<i>Lemna</i> , 15%	Egg production, %	82.14 ± 4.79	83.75 ± 4.27 <sup>x</sup>	79.38 ± 5.73 <sup>x</sup>
	Feed consumption, kg	.113 ± .002 <sup>y</sup>	.118 ± .005	.118 ± .005
	Feed conversion, kg/kg	2.293 ± .079	2.311 ± .050 <sup>y</sup>	2.384 ± .225 <sup>xy</sup>
	Mean egg weight, g	60.22 ± 1.53 <sup>b</sup>	60.84 ± .75 <sup>b</sup>	62.85 ± .73 <sup>a</sup>
	Mean weight gain, g			122 ± 214
	Number of eggs/hen per week	5.750	5.863	5.556
	Yolk pigmentation, 2 to 10 wk			13.30 ± .60 <sup>y</sup>
<i>Lemna</i> , 25%	Egg production, %	76.07 ± 8.86	71.16 ± 6.46 <sup>y</sup>	70.36 ± 4.38 <sup>y</sup>
	Feed consumption, kg	.118 ± .004 <sup>x</sup>	.119 ± .004	.118 ± .004
	Feed conversion, kg/kg	2.624 ± .304	2.811 ± .266 <sup>x</sup>	2.654 ± .156 <sup>x</sup>
	Mean egg weight, g	60.05 ± 1.22 <sup>b</sup>	60.03 ± .25 <sup>b</sup>	63.18 ± 1.14 <sup>a</sup>
	Mean weight gain, g			74 ± 234
	Number of eggs/hen per week	5.325	4.981	4.925
	Yolk pigmentation, 2 to 10 wk			14.09 ± .79 <sup>z</sup>

<sup>a-c</sup>Means (± SD) within a row with no common superscripts differ significantly (P<.05).

<sup>x-z</sup>Means (± SD) within a column for the same trait with no common superscripts differ significantly (P<.05).

<sup>1</sup>Mean egg weights represent the total kilograms produced per period divided by the total production number.

the protein content of eggs from hens fed the control diet.

Formal double-blind taste tests of eggs from both hen lines were performed at the UNA by well-trained panelists. Pigmented yolks were preferred over the paler, control yolks; when rated, the overall quality (flavor, smell, color, and appearance) of eggs from the group fed 15% *Lemna* had the highest rating compared with eggs from the control, 25%, and 40% *Lemna* diet groups. No unusual tastes were detected in any of the groups tested.

## DISCUSSION

Human waste represents an abundant source of nutrients for duckweed. Naturally occurring populations of *Lemna gibba* and *Wolffia arrhiza* growing in Lima's urban sewage lagoons were examined for their nutrient value as a constituent of feed for commercial poultry.

Duckweed species grow in the Lima area throughout the whole year. Harvesting of

duckweed plants is an easy task, as they form a floating mat with no structural unity that would make cutting, chopping, or separation necessary. Simply skimming the fronds from the water surface is sufficient (Oron *et al.*, 1986). Duckweed was harvested from a tertiary oxidation lagoon and from a lagoon formed by seepage from a nearby tertiary lagoon. Duckweed harvested from these lagoons was a very useful addition to diets for layer hens.

Duckweed, when fresh, contains between 92 to 95% water. This high volume of water limits the amount able to be eaten by chickens and, therefore, significantly reduces the amount of effective nutrient intake (personal observation). Drying reduces volume, concentrates nutrients, and is the key to the successful use of this water plant in high levels as a feed source. The amino acid profile of duckweed is very similar to that of soybean, being high in lysine and other amino acids but somewhat low in methionine (Rusoff *et al.*, 1980).

Using two different lines of hens, the results indicate that duckweed can be used as a

TABLE 6. Protein content of eggs from HyLine Leghorn hens fed diets containing 15 or 25% *Lemna* species or an isonitrogenous, isocaloric control diet

Protein content	Treatments		
	Control	15% <i>Lemna</i>	25% <i>Lemna</i>
		(%)	
Albumin	84.302 ± .332 <sup>C</sup>	84.746 ± .168 <sup>B</sup>	86.095 ± .576 <sup>A</sup>
Yolk	15.642 ± .232 <sup>C</sup>	16.283 ± .125 <sup>B</sup>	17.238 ± .141 <sup>A</sup>

<sup>A-C</sup>Means within a row for the same trait with no common superscripts differ significantly (P<.001).

nutrient source for layers. Using 15% *Lemna* in the diets maintained egg production levels and mean egg weight. Also, augmenting the amount of duckweed in the diets of Leghorn hens increased the total protein of the eggs and desirability in the taste of the eggs.

*Lemna* may substitute for fish meal in the poultry diets. In places such as Peru, fish meal is often used in concentrations of up to 15%. Diets containing high concentrations of fish meal may be associated both with black vomit, a toxic effect in chickens, and poor tasting eggs (Rojas and Bernuy, 1976). In HyLine Leghorn hens fed diets containing 25% *Lemna*, soybean was eliminated and the percentage of fish meal decreased to 2% in comparison with 7% in the control diet.

In diets containing 40% *Lemna*, both soybean and fish meal were eliminated completely from the diet, leaving corn as the major constituent. Hens fed this diet were able to maintain rates of egg production similar to that of controls. Hens fed a diet of 40% *Lemna* produced feces that were bulky and somewhat wet, a factor undesirable in commercial operations. However, diets containing 40% *Lemna* may be useful in household farms in the third world, especially in places where fish meal or soybean are costly or not available.

Pigmentation is an important attribute that adds to the economic value of duckweeds as dietary ingredients. High yolk pigmentation is commercially desirable and correlates highly with dietary duckweed levels. The addition of 15% *Lemna* in the diets resulted in higher egg pigmentation than in eggs of controls. Higher levels of duckweed produced increased pigmentation but less efficiently.

Duckweed may successfully substitute for soybean and some fish meal without affecting the hens or the quality of the eggs. In countries where corn and soybean meal, the key

ingredients in poultry feeds, are often imported, substantial savings may be realized through the large-scale development of a sewage duckweed poultry industry.

It was concluded that sewage-grown duckweed can be successfully utilized as a protein source in diets for layer hens. The optimal level of *Lemna* in the diets of chickens was 15%, but even at 40%, egg quality was not affected, and egg production was significantly reduced in only one of two periods. In areas where fish meal, or soybeans are not available, duckweed represents a readily available source of high quality protein that can be produced indigenously, utilizing unexploited resources such as sewage for its growth medium. The large-scale production costs of duckweed are unknown. Further exploration of the use of *Lemna* as a cost-effective protein and pigment source for the diet of animals is a high priority for countries where protein sources are both expensive and scarce.

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