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REPORT

To: Joan La Rosa
Health Officer AID/Peru

From: Duckweed Group (I.I.N.)

Date: January 31, 1985.

Subject: Half-yearly report: "The Safety and Efficacy of Sewage Grown Duckweed as feed for Chicks, Layers and Broilers."

PREAMBLE

The first six months of this project have been unusually productive considering the need to develop an appropriate institutional infrastructure, train personnel, purchase (or build) and ship major pieces of equipment and establish reliable supply lines. We have in these 6 months been able to demonstrate that short term feeding of two species of duckweed to chickens provides a nutritious, highly acceptable and safe foodstuff which has characteristics similar to toasted soy bean. In addition we have found that duckweed has considerable pigment, a quality avidly looked for in a feed because of its high commercial value.

In the absence of any prescribed format for a report of this nature, we have chosen to organize this report under three major headings: (1) Institutions, (2) Equipment and Supplies, and (3) Scientific Research.

INSTITUTIONS

INSTITUTO DE INVESTIGACION NUTRICIONAL (IIN)

Facilities & Services: The IIN has provided most of the office and laboratory space required by the project. Room for one or more investigators, a secretary and two computers have been provided in the main office block of the IIN. The project shares space in the Microbiology and Biochemestry laboratories with on-going IIN research. Administrative and accounting services for the project are provided by regular IIN administrative staff. The IIN has also made available to project staff such services as transportation, administrative advice (negotiating contracts and agreements), expert advice, training (biochemistry and microbiology) and photocopying.

Rec'd in SCI: MAY 6 1985

Personnel: A bilingual secretary (Marissa Vega), already working at the IIN was transfered to the project, a laboratory technician (Gladys Ventura) already working in the IIN microbiology laboratory was transfered to the project, and a laboratory technician (Anna Theresa Haustein) was hired for the food biochemistry laboratory. The microbiology technician, having already worked for the IIN, was proficient in most procedures required by the project. Additional training in two assays was provided by Drs. Gilman and Spira. Training in assays to measure crude protein, fat, crude fiber, carbohydrates, total energy and ash was provided by IIN staff. Drs. A. Gastañaduy, R. Gilman and P. Skillicorn supervised and coordinated the various activities conducted at the IIN. Ms. J. Gilman played a pivotal role, on a voluntary basis, of trouble shooting, managing project staff, and coordinating the activities of the various institutions involved in the project.

UNIVERSIDAD NACIONAL AGRARIA (UNA)

Facilities & Services: Using the existing concrete floor, woven bamboo matting, burlap and a minimal superstructure of bamboo and wooden poles two simple structures were built to provide, for the experimental poultry, shelter from the sun and wind and isolation from free flying birds. As rainfall is a rare occurance in Lima waterproof roofing was not provided.

The UNA has made available to the project, the use of its large forced-air drier (for drying green Duckweed), and its biochemistry laboratory (for the TME assays). Office space for Mr. Victor Vergara and Dr. Victor Guevara are provided by the Department of Nutrition.

Personnel: Dr. Victor Guevara was detained in Australia through the middle of September, and was thus unable to facilitate the early search for permanent housing for the poultry.

Mr. Victor Vergara, a masters graduate in poultry nutrition, was hired fulltime by the project to supervise site management (growing, harvesting, drying and transporting Duckweed) at the San Juan lagoons and conduct the poultry experiments. He also supervised the design and fabrication of the poultry batteries and brooders, as well as the construction of the temporary housing for the poultry experiments.

In Dr. Guevara's absence, Drs. Lung and Talavera, both from the Department of Nutrition, provided valuable assistance in dealing with the University administration and suggesting alternative plans for developing housing for the poultry experiments. Subsequently, Dr. S. Rojas, Chairman of the Department of Nutrition, having taken an intense personal interest in the work undertaken by the project group, has provided valuable advice and assistance. Dr. S. Rojas has been involved in design of most of the recent experiments. In this regard we have decided to consider Dr. Rojas as a consultant for the duckweed group.

SEDAPAL

Facilities & Services: On November 20, after numerous revisions both parties (IIN and SEDAPAL) finally afixed their signatures to a *Convenio*. It specified the following key provisions: (1) the project would provide two "Duckweed workers" at the lagoon site; (2) the project would be permitted to grow duckweed on 3 ha. of lagoon space, but would not be allowed to completely cover a lagoon with Duckweed; (3) the contribution of SEDAPAL would be recognized in all project documents; (4) the project could install baffling of specified dimensions on 3 lagoons; and (5) the project could harvest and remove duckweed from the lagoon premises.

Personnel: Two "Duckweed workers" were hired by the project in November. Their function is to maintain and harvest the Duckweed populations on the three lagoons specified in the *Convenio*. Occasional assistance has also been received from site operations engineer and lagoon laborers.

THE RALSTON PURINA COMPANY

Facilities & Services: Eng. Ricardo Fossa, Director of Diets and Nutrition at Ralson Purina (Lima), first at the direction of the parent company (St. Louis) and later through his own growing interest in the project, has been instrumental in the provision, through Ralston Purina, of the following services, facilities and supplies: (1) Computer generated diets, (2) Training (project technician) in the Purina food biochemistry laboratory, (3) Biochemical analysis of Duckweed samples, and (4) donations of premixed poultry feeds and feed ingredients.

THE UNIVERSIDAD PERUANA CAYETANO HEREDIA (UPCH)

Facilities and Services: Media, reagents, and computer supplies urgently required by the project, but not available locally were provided at short notice by the Department of Microbiology at UPCH.

AVICOLA HANNAN (AH)

Facilities and Services: All the chickens used in the experiments conducted thus far have been donated by the Avicola Hannan poultry company.

INTERNATIONAL POTATO INSTITUTE (CIP)

Services: The plant Pathology Laboratory at CIP has assisted in the identification of a fungal infection and an insect parasite affecting the Duckweed population at the San Juan lagoons.

PAN-AMERICAN HEALTH ASSOCIATION (PAHO - CEPIS)

Services: Drs. Carl Bartone and Henry Salas of CEPIS have provided invaluable technical assistance as well as helping in the difficult negotiations with SERPAR and SEDAPAL. Baseline water quality data on the San Juan lagoons have also been made available through the CEPIS laboratory. Dra. Carmen de Mayo has provided considerable help with microbiological techniques available for water bacteriology at Cepsis.

JHU - DEPARTMENT OF BIOLOGY

Services: Dr. Dennis Powers and his laboratory assistant Joyce Lilly conducted the amino acid analysis of Duckweed samples taken from the San Juan lagoons.

JHU - DEPARTMENT OF INTERNATIONAL HEALTH

Facilities and Services: Purchasing and shipping of equipment and supplies were handled through the office of the Nutrition Division of the Department of International Health and the Division of Geographic Medicine of the Department of Medicine.

OCCUPATIONAL SAFETY AND HEALTH ADMINISTRATION (OSHA)

Facilities and Services: Analysis of Duckweed samples for the presence of toxins and heavy metals was conducted at OSHA's Denver laboratories.

UNIVERSITY OF MARYLAND SCHOOL OF MEDICINE (UOM)

Facilities and Services: Filters prepared from the fecal matter of chickens were tested for the presence of *E. coli* at the microbiology laboratory of the Division of Geographic Medicine.

EQUIPMENT AND SUPPLIES

CHANGES

Changing circumstances have dictated the need for several major revisions in the equipment and supplies specified in the letter of agreement between USAID and the IIN:

1. Access, by the project, to existing equipment which is suitable for harvesting and processing Duckweed (i.e., the forced air ovens at UNA) obviates the need to spend $3000 on Duckweed harvesting & drying and Grinding/mashing equipment. Funds saved in this manner should be diverted towards defraying the cost of an additional Duckweed worker.

2. Two factors - anticipated decreases in microbiological analyses (Campylobacter assays in particular) required by the project, and the availability of space in a Revco at the UPCH microbiology laboratory - have eliminated the need to purchase a Revco freezer. The $ 2,915 budgeted for this purpose would be better spent on a small bomb calorimeter, an item the need for which was not anticipated in the original project budget.

3. Several revisions have proven necessary with respect to the projected computer requirements of the project. As the Osborne computer company has ceased to exist, we opted for IBM computers. Experience has shown that conventional Winchester hard disk drives are too sensitive to dust, changes in temperature and electricity fluctuations to be practical in Lima. A decision was taken therefore, to substitute the Bernoulli Box twin 10 M byte system, a more flexible mass storage technology, for the winchester drive specified in the original budget. Communications needs and Peru's unreliable power supply also prompted the need to a modem and uninterruptable power supplies.

4. The original budget did not anticipate the need to construct housing for the poultry experiments. The cost of this housing will be defrayed partially through savings in the cost of building broiler batteries, and partially through savings in the cost of chickens and feed.

5. Local transportation has proven to be the most restrictive budget item. The number of participating institutions, and their wide geographic distribution within Lima has resulted in travel expenses far greater than that anticipated in the original project budget. In an attempt to bring these costs down, a decision was made to minimize reliance on public transportation. Ms. Gilman purchased a Volkswagen car and agreed to rent it to the project for $10 per day. Without this arrangement the entire travel budget of the project would not have been sufficient for more than 12 months.

Pens (batteries and brooders) to house the experimental chickens were built under a contract with a local sheetmetal fabricator suggested by the UNA. Uncertainty with respect to the design and projected costs (inflation played a major role) prolonged the contract negotiations for a month and a half. All batteries and brooders were delivered by December 25.

SCIENTIFIC RESEARCH

SAN JUAN SEWAGE LAGOONS

Through careful observation of the San Juan lagoons during the months of June through December several critical factors have become apparent to project investigators.

1. Wind and Water Level Fluctuations play a major role in depleting standing populations of Duckweed (leaving Duckweed stranded on the shore).

2. **Species Differences** - Two species of Duckweed, *Lemna gibba* and *Wolffia* were found growing in the Lagoons. Growth blooms are seldom coincident, resulting in dominance swings between the two species. *L. gibba* would appear to be the heartier of the two species.

4. Disease - An infectious agent (most probably a fungus) which kills Duckweed (they turn white, and eventually lose their boyancy) in a slowly spreading (outward) circle is a problem (particularly apparant during periods of slow growth) which can result in significant reductions in plant populations in certain lagoons. During high growth phases the population growth far exceeds losses from the infection. The nature of this infection is currently being investigated through the plant pathology department of CIP. A common fungus, (_Oomycetes) has been isolated from diseased Duckweed samples and is thought to be the most likely cause of infection.

6. **Parasites** - Crowded Duckweed populations (conditions for low growth) found in primary and secondary lagoons were at times found to be heavily infected with aphids. Parasites do not appear to be problems in fast-growing populations.

7. **Baffling** - In order to prevent crowding of Duckweed against the shore, as well as effect a more even distribution of Duckweed throughout a lagoon, efforts were made to devise efficient and inexpensive floating baffles. The most successful effort involved stringing long strands of green *carriso* (a thin hollow bamboo-like cane) together and floating them across a lagoon. The green *carriso* strands sprouted, sending up small leafy fronds which also helped buffer plants from the wind. This is the current method used to baffle duckweed and prevent harmful wind effects.

8. **Harvesting** - No elaborate harvesting mechanisms have thus far proven necessary. Harvesting is effected by lowering a tined leaf rake beneath the duckweed and manually removing it into a wheelbarrow, bucket or plastic bag. Because the San Juan site is seldom without a brisk breeze major duckweed populations are always located adjacent to lagoon shorelines.

9. **Other Factors** - There is a surprisingly low growth of Duckweed in the quaternary lagoon adjacent to the tertiary lagoon in which the best duckweed growth is found. Conditions in the quaternary lagoon are, if anything more favorable than those in the neighboring tertiary lagoon, and yet virtually no Duckweed is found in the lagoon. Even those Duckweed plants which flow through the tertiary outflow into the quaternary lagoon appear to quickly disappear. The reason for this difference is speculative with possibilities including ingestion by talapia, adverse algal populations and toxic water conditions.

DUCKWEED:

Toxicity: Two uptake mechanisms have been postulated for Duckweed. In a high growth phase (i.e., suitable temperatures and high concentrations of nutrients in the growth medium) Duckweed appears to use only those elements and compounds which contribute directly to its growth. Heavy metals, complex organics and inorganics are not absorbed. In conditions where nutrients are constrained, Duckweed appears to act like an organic vaccume cleaner; absorbing virtually anything in its effort to grow. It can directly employ complex organics as food, can fix nitrogen, and removes trace amounts of phosporus from the water. Having survived for some time in such a "hungry" condition (low nutrients) Duckweed plants have been shown to have surprisingly

high concentrations of toxic minerals and metals.

The Duckweed growing in primary, secondary and tertiary lagoons at the San Juan site experiences no nutrient constraints. Dried samples were tested at the OSHA laboratory in Salt Lake City for the presence of the following substances:

Substance	Detection Limit
Arsenic & compounds (as AS)	.0001%
Beryllium and compounds	.0005%
Cadmium fume (as Cd)	.001%
Cobalt, metal, fume & dust	.001%
Chromium, metal & insoluble Salts	.001%
Copper fume (as Cu)	.001%
Iron	.001%
Manganese & compounds	.001%
Mercury, inorganic (as Hg)	.00001%
Molybdenum (as Mo) (insolubles)	.002%
Nickel, metal & soluble compounds	.002%
Lead, inorganic fumes & dusts	.007%
Antimony & compounds (as Sb)	.005%
Vanadium (as V)	.001%
Zinc	.0001%

The duckweed taken from the primary lagoon tested positive for Zinc (.0063%), Antimony (.0126%) and Copper (.0066%); the Duckweed taken from the secondary lagoon tested positive for Zinc (.0031%); and the Duckweed taken from the tertiary lagoon tested positive for Zinc (.0022%). All other results were negative. We will continue to test for the presence of these (above) substances during the next 18 months, but do not anticipate obtaining substantially different results.

Based on these results we offer the preliminary conclusion that duckweed grown in the San Juan lagoons does not contain sufficient quantities of toxic metals or compounds to constitute a threat to humans if consumed as a feedstuff by poultry.

Human Pathogens - In numerous replications, duckweed taken from all three San Juan lagoons (primary, secondary and tertiary) were washed in fresh water, and then comprehensively tested for the presence of enteric bacteria. The only pathogen which recorded a positive identification was *Aeromonas* and *Vibrio* species both are common environmental contaminants found in fresh water lagoons and on water plants under natural conditions.

Biochemistry - Biochemical analyses of dried duckweed samples were conducted at the IIN and Purina laboratories. Results, in general, confirmed expectations. (See table # I) Protein (N x 6.25) content, which ranged from 25% to 39% was well within the expected range. With the onset of vigorous harvesting protein contents may even increase to above that of 40%. Low harvesting also contributed, in two ways (floating debris mixed with the plants and an ageing plant population), to ash content (11% to 24%) well above expected levels.

The value of duckweed as a feedstock is highly dependant on its amino acid profile. It was highly encouraging to find that duckweed had a high content of lysine. Like soybean it was however low in methionine. Initial results indicate considerable variation between samples (See table II). It is premature to speculate on the reason for this variation, but laboratory error has not been ruled out. Repeated analyses for determination of amino acids will be conducted at the JHU Biology department laboratories during the next 12 months in efforts to either confirm or refute these prelimary results.

The xanthophyll content, - from a commercial perspective a very important - component of Duckweed, deserves separate mention. The xanthophyll content of duckweed harvested from the San Juan lagoons was found to be extremely high (up to 1938 ppm). Exposure to direct UV radiation resulted in rapid degradation, however, with xanthophyll levels as low as 282 ppm recorded for sun-dried Duckweed.

TESTING MARKET CHICKENS FOR HUMAN PATHOGENS

An objective of this project is to demonstrate that Duckweed based feeds do not contribute to higher prevalence rates of human enteropathogens in chickens. In order to demonstrate this, 100 chickens were selected at random from wholesale and retail distribution centers throughout Lima. Stool samples taken from these chickens for tested for a complete battery of enteric pathogens. Particular emphasis was placed on testing for the presence of Campylobacter and Salmonella species - both reported in the literature to have high prevalence rates in chickens worldwide.

FEEDING TRIALS

The lengthy delay in reaching an accord with SEDAPAL prevented all but nominal harvesting of Duckweed from the San Juan lagoons. It was impossible, therefore, to conduct the major chicken feeding trials. But, as small quantities of Duckweed were available a decision was taken to conduct two preliminary trials (one using dried duckweed and one using fresh duckweed) rather than wait for the conclusion of negotiations to begin the poultry experiments.

FIRST PILOT EXPERIMENT WITH DRIED DUCKWEED

OBJECTIVES OF THE FIRST PILOT STUDY

1. To determine the performance of chicks when dried *Lemna gibba* replaces either soy cake or soy flour in the diet.

2. To measure the effects of *Lemna* on pigmentation of the skin of the chickens.

3. To determine the safety of sewage grown *Lemna* in chickens grown for fattening (for meat).

EQUIPMENT.-

1. Two batteries with heat control, containing 5 floors each with metal trays. Chickens were kept in the batteries for the length of the experiment (1 - 21 days).

ANIMALS.-

1. 80 Top line Titan variety chickens one day old, were donated by "Avicola Hannan".

METHODS.-

1. Chickens were randomized into 4 treatment groups, ten per group. On the first day, they were weighed in groups (See table III).

2. For days 0 to 9, chickens were fed with Maiz-Soya commercial feed. On the 9th. day they were weighed individually (See table III). In each group, the 2 heaviest and the 2 lightest were removed.

3. Four diets were formulated, a) Maiz-Soya, b) Maiz-Lemna, c) Trigo-Soya, d) Trigo-Lemna (See table IV & V). Chickens were fed these diets from days 9 to 21. Free water with multivitamin drops.

4. Variables measured: Weight gain, pigmentation, formation of stool, food and water consumption and health appearance.

5. Pigmentation was measured by Roche color chart from yellow to orange on the 21st. day, by a person not involved in the experiment. (See table VI).

6. Excreta were collected on 3, 5, 7, 9 and 12 days and weighed for each group. Chemical analysis was performed on 7 and 12 days. Rectal swabs for microbiology were taken on days 9 and 21. (See tables VII & VIII).

RESULTS

A. **Weight Gain** - Using a 3-way randomized ANOVA it was determined that the major factor ($p < .01$) contributing to differences between groups was the carbohydrate feedstuff (corn/wheat). While the protein factor (soybean/duckweed) was also a significant ($p < .01$) contributor to differences between groups, it is important to note that the chickens fed the duckweed/corn diet performed significantly better (Student t: $p < .01$) than did the chickens fed the soy/wheat diet. It is also noteworthy that, while not statistically significant (Student t: $p > .05$), the duckweed/wheat chickens' weight gain was obvicusly better than that of the soy/wheat chickens. In a trial using larger numbers of chickens, the latter trend would be expected to achieve statistical significance.

B. **Pigmentation** - Both duckweed groups (duckweed/corn and duckweed/wheat) had significantly better pigmentation than the two non-duckweed groups (Student t: $p < .01$).

C. **Human Pathogens** - No human enteropathogens were cultured from the excreta of any of the experimental chickens.

D. **Excreta** - Both Duckweed groups produced significantly more (Student t: $p < .01$) excreta than the two non-duckweed groups.

DISCUSSION

While the size of this pilot study precludes definitive conclusions, several significant and noteworthy trends were observed:

1. Duckweed would appear to be an adequate replacement for soybean meal in poultry diets. While it clearly does not perform as well as soybean meal when mixed with corn, it appears to outperform soybean in combination with wheat. Acceptance of the duckweed based feed by chickens was not a problem.

2. Sewage grown Duckweed appears to be a safe additive to poultry diets. It does not transmit human enteropathogens to chickens, and appears (in the instance of the San Juan lagoons) not to contain significant concentrations of common toxins and heavy metals.

3. Duckweed is an excellent agent for providing pigmentation to poultry. The level of pigmentation of poultry is generally acknowledged in the U.S. to be the major factor employed by consumers to discriminate between chickens in the market place. Judging by the content of current advertizing, this would appear also to be the case in Peru.

SECOND PILOT EXPERIMENT USING FRESH DUCKWEED

The second experiment employed the same general methods as the first experiment, but used only two groups. One group (control) was fed the Soy/Corn diet while the experimental group was fed a diet consisting of 10% (on a dry weight basis) fresh duckweed and 90% standard Soy/Corn mix. Noting that fresh duckweed consists of approximately 95% water, and assuming a normal consumption for a given chicken of 100 gms of standard dry feed per day, that chicken would be required to consume approximately 290 grams (of which 190 grams would consist of water) of the duckweed mix to maintain the same dry volume intake.

RESULTS

Chickens clearly liked the duckweed mix, but were unable to consume the same dryweight volume of feed as the control chickens. This was reflected in a slightly lower weightgain over 21 days. The excreta of the experimental chickens was also significantly greater in volume and had a more fluid-like consistency. The experimental chickens were also observed to be eating more of the time.

The only enteropathogen isolated from either the feed or the chickens was Aeromonas hydrophiliae. Aeromonas is a common bacteria present in fresh water, and in several different animal species.We have isolated it in market chickens. Its role as a pathogen is uncertain at the present time. Interestingly chickens fed wet duck weed did not become infected with Aeromonas.No chicken fed wet duckweed had diarrrea and none died.

Autopsy was carried out on 8 duckweed fed and 4 control chickens and the only abnormal finding was that of one helminthic parasite (still to be identified) found in one of the duckweed fed chickens.

DISCUSSION

Ten percent (10%) fresh duckweed (on a dry weight basis) would appear near the upper limit on the amount of fresh duckweed it is possible for chickens to consume. It is doubtful they could tolerate consuming more than twice the weight of their dry food intake in liquid. The experiment did, however, demonstrate that fresh, unprocessed (and therefore very inexpensive) duckweed can be reasonably and safely included in poultry diets at up to 10%.

TRUE METABOLIZABLE ENERGY

We determined the total metabolizable energy of duckweed in 4 groups of animals. One group received corn, and the other 3 groups received duckweed at three different levels, 15 g, 20 g, and 25 g.

RESULTS.-

Duckweed when fed at 3 different levels showed little variation in its total metabolizable energy value. They were respectively for each of the three levels, (15 gs, 20 gs, and 25 gs.) 2794, 2853 and 2706 cal/g.

DISCUSSION.-

The TME values of duckweed are about the same as those obtained for toasted soybean. They are much higher than TME values obtained for other sewage products such as sludge and algae which have been fed to chickens.

FUTURE STUDIES

The next study which has been completed but is awaiting biochemical analysis is a determination of metabolizable energy (ME). This determination will help us design the best diet for layer experiments which will start in the first week of March. We will also start chicken and broiler diet experiments during the next half year.

Our main concerns(problems which are not currently funded) now is with determining growth rates in the lagoon and improving methods for drying duckweed. We can now predict with a high degree of confidence that duckweed will be nutritious and safe. In the future we will need to determine whether duckweed in scale up projects can be produced in sufficient quantity and at a competitive price. In addition its utility for the small integrated farmer needs to be explored. The two major constraints in duckweed production are drying and growth conditions.We need to determine the factors responsible for duckweed growth and also to find the most economical technology available for drying duckweed. It is hoped that further funding will be obtained to help explore these new avenues of research.

TABLE I

Sample	Date	Drying: Sun (Hrs)	Drying: Oven (T Hrs)	Humidity	Protein	Fiber	Ash	Calcium	Phosph.	Fat	Xanthophyls (parts/million)	Salt	Sand
L. gibba	29-Aug	-	Purina	95.08	33.71	14.72	12.15	00.90	00.50	03.69	469	-	-
L. minor (?)	29-Aug	-	Purina	93.67	38.80	07.91	15.00	01.00	00.58	04.02	866	-	-
Wolfia	10-Sep	-	96 Hrs UNA	-	-	-	-	-	-	-	1938	-	-
L. gibba	10-Sep	-	45C 24 Hrs UNA	-	-	-	-	-	-	-	958	-	
L. gibba + wolfia	10-Sep	72 Hrs	-	-	-	-	-	-	-	-	720	-	
L. gibba + wolfia	10-Sep	48 Hrs	12 Hrs UNA	-	-	-	-	-	-	-	282	-	-
Wolfia	13-Sep	?	-	12.01	31.36	11.68	23.82	02.48	00.82	03.62	974	3.57	6.59
L. gibba	13-Sep	48 Hrs	80C 5 Min UNA	10.00	33.97	08.88	19.13	01.60	00.85	03.83	658	5.09	1.78
L. gibba	13-Sep	12 Hrs	50C 2 Hrs UNA	09.42	33.53	09.01	22.01	01.70	00.88	04.52	1083	5.06	4.88
L. gibba	13-Sep	24 Hrs	50C 10 Min UNA	09.28	32.85	09.18	23.61	01.69	00.90	03.42	868	5.12	5.40
L. gibba	12-Oct	09 Hrs	80C 10 Min IIN	-	-	-	-	-	-	-	137	-	-
L. gibba	12-Oct	09 Hrs	80C 10 Min IIN	-	-	-	-	-	-	-	130	-	-
L. gibba	12-Oct	-	80C 7 Hrs IIN	-	-	-	-	-	-	-	642	-	-
L. gibba	12-Oct	-	80C 7 Hrs IIN	-	-	-	-	-	-	-	631	-	-
L. gibba	12-Oct	2:40 Hrs	80C 4:30 Hs IIN	-	-	-	-	-	-	-	731	-	-
L. gibba	12-Oct	2:40 Hrs	80C 4:30 Hs IIN	-	-	-	-	-	-	-	704	-	-

TABLE II

	Lemna Gibba 10-19-04 .5% pond #3	Lemna Gibba 10-19-04 .25% pond #3	Lemna Gibba 10-19-04 .5% pond #3
Aspartic	26.2	17.6	17
Threonine	7.2	4	4.4
Serine	7	4	4.3
Glutamic	31.1	18.7	17.1
Proline	(6.8)	5.3	(4.5)
Glycine	12.8	6.9	7.8
Alanine	17	9.2	9.8
Valine	13.7	8	9.9
Methionine	2.6	(1.5)	1.6
Isoleucine	8.2	(2.7)	4.9
Leucine	12	4.5	6.7
Tryptophane	2.2	1.2	1.9
Phenylalinine	4.8	2.7	(5.7)
Histidine	3	2.6	2.1
Lysine	10.1	6.5	6.6
Arginine	12.2	6.7	7

NOTES: The numbers in parenthesis are negative numbers

TABLE III

	WEIGHT RESULTS		
	1st. Day Alive (gm.)	9th. Day Initial (gm.)	21st. Day Final (gm.)
111	39.00	180.00	700.00
112	39.00	183.00	720.00
113	39.00	160.00	615.00
114	39.00	160.00	615.00
115	39.00	183.00	775.00
116	39.00	168.00	780.00
117	39.00	172.00	773.00
118	39.00	171.00	640.00
121	39.60	164.00	615.00
122	39.60	169.00	580.00
123	39.60	187.00	670.00
124	39.60	160.00	540.00
125	39.60	188.00	685.00
126	39.60	184.00	640.00
127	39.60	186.00	645.00
128	39.60	174.00	590.00
211	39.20	159.00	460.00
212	39.20	168.00	560.00
213	39.20	165.00	479.00
214	39.20	186.00	535.00
215	39.20	180.00	575.00
216	39.20	170.00	474.00
217	39.20	189.00	510.00
218	39.20	159.00	510.00
221	38.00	165.00	468.00
222	38.00	184.00	559.00
223	38.00	172.00	495.00
224	38.00	162.00	475.00
225	38.00	185.00	552.00
226	38.00	160.00	495.00
227	38.00	160.00	490.00
228	38.00	178.00	492.00

(Cont.)

WEIGHT RESULTS (Cont.)

	1st. Day Alive (gm.)	9th. Day Initial (gm.)	21st. Day Final (gm.)
311	40.00	180.00	480.00
312	40.00	160.00	430.00
313	40.00	180.00	512.00
314	40.00	174.00	478.00
315	40.00	180.00	555.00
316	40.00	165.00	400.00
317	40.00	175.00	425.00
318	40.00	170.00	445.00
321	39.50	159.00	436.00
322	39.50	165.00	450.00
323	39.50	186.00	442.00
324	39.50	168.00	508.00
325	39.50	186.00	442.00
326	39.50	176.00	475.00
327	39.50	178.00	465.00
328	39.50	188.00	535.00
411	38.00	184.00	506.00
412	38.00	174.00	460.00
413	38.00	164.00	508.00
414	38.00	184.00	555.00
415	38.00	174.00	460.00
416	38.00	182.00	488.00
417	38.00	160.00	400.00
418	38.00	185.00	585.00
421	41.00	174.00	520.00
422	41.00	174.00	480.00
423	41.00	159.00	400.00
424	41.00	184.00	500.00
425	41.00	188.00	540.00
426	41.00	190.00	548.00
427	41.00	170.00	470.00
428	41.00	164.00	485.00

XYZ: X = Treatments (Diets 1....4)
Y = Replicates
Z = Chicken number per experimental unit.

TABLE IV

DIET FORMULAS (%)

	Dieta 1 Maiz/Soya	Dieta 2 Maiz/Lemna	Dieta 3 Trigo/Soya	Dieta 4 Trigo/Lemna
Corn	65.00	66.00	-	-
Wheat	-	-	62.00	63.23
Fish Meal	5.00	5.00	4.00	4.00
Soybean Cake	27.00	-	27.00	-
Lemna	-	27.50	-	27.50
Soy oil	0.63	-	4.50	3.60
Bone fluor	1.70	1.12	1.88	1.27
Calcium carbonate	0.30	-	0.24	-
DL-Methionine	0.17	0.18	0.20	0.20
Premix (vit.and min)	0.20	0.20	0.20	0.20
Total	100.00	100.00	100.00	100.00

TABLE V

ANALISIS QUIMICO PROXIMAL DE LAS DIETAS*

HUMEDAD	PROTEINA	GRASA	FIBRA	CENIZA	NIFEX
12.92	20.34	4.35	5.23	4.85	52.31
11.48	18.52	4.53	7.00	8.10	50.37
10.88	20.02	6.02	5.35	4.50	53.23
9.18	16.70	6.46	5.98	8.85	52.83

* Analisis realizados en los laboratorios de Nutricion, U.N.A.

TABLE VI

PIGMENTATION

111	3	211	8	311	3	411	7
112	5	212	6	312	1	412	4
113	3	213	6	313	2	413	5
114	2	214	5	314	1	414	5
115	5	215	9	315	1	415	6
116	4	216	5	316	2	416	4
117	4	217	5	317	2	417	6
118	4	218	8	318	1	418	6
121	5	221	8	321	1	421	6
122	7	222	7	322	1	422	6
123	4	223	7	323	1	423	4
124	3	224	5	324	2	424	6
125	4	225	6	325	2	425	5
126	6	226	7	326	2	426	7
127	4	227	7	327	1	427	6
128	3	228	5	328	1	428	6

* Measured with the colorimetric fan "ROCHE"

CODING: XYZ
X = DIET
Y = DUPLICATE
Z = CHICKEN NUMBER

TABLE VII

PESO DE EXCRETA (GRS.)
[Base humeda]

	PERIODO DE COLECCION (Dias)					
	3	5	7*	9	12*	TOTAL
11	465.0	380.0	370.0	510.0	875.0	2600.0
12	450.0	395.0	410.0	540.0	975.0	2770.0
Prom.	457.5	387.5	390.0	525.0	925.0	2685.0
21	710.0	820.0	950.0	1310.0	2100.0	5890.0
22	525.0	525.0	630.0	762.0	1250.0	3692.0
Prom.	617.5	672.5	790.0	1036.0	1675.0	4791.0
31	400.0	306.0	348.0	465.0	650.0	2169.0
32	410.0	365.0	350.0	455.0	870.0	2450.0
Prom.	405.0	355.5	349.0	460.0	760.0	2329.0
41	580.0	580.0	680.0	950.0	1530.0	4320.0
42	655.0	685.0	780.0	880.0	1500.0	4500.0
Prom.	617.5	632.5	730.0	915.0	1515.0	4409.0

* Muestras enviadas al laboratorio para su analisis quimico proximal.

TABLE VIII

*Sample	Humidity	Protein	Fat	Ash	Calcium	Phosphor.	Fiber	Salt	Na (mg%)	K (mg%)
E7-11	10.30	29.33	2.52	13.05	2.35	1.46	14.29	0.47	0.088	0.196
E7-12	11.65	26.11	2.43	12.23	2.11	1.31	16.21	0.56	0.096	0.213
E7-21	11.60	25.50	5.37	21.06	2.19	1.11	10.92	4.15	0.814	0.368
E7-22	11.20	24.66	5.00	20.39	2.11	1.09	11.15	3.97	0.802	0.376
E7-31	10.70	25.00	5.31	12.71	2.03	1.21	14.91	0.35	0.083	0.201
E7-32	11.55	23.74	3.09	12.26	1.91	1.27	15.08	0.47	0.079	0.201
E7-41	10.90	21.48	8.91	18.30	2.08	1.03	10.05	3.21	0.705	0.304
E7-42	11.40	20.14	7.34	18.71	2.10	1.09	10.29	3.33	0.718	0.300
E12-11	10.00	29.43	3.18	14.00	2.66	1.61	14.75	0.53	0.087	0.194
E12-12	10.20	30.77	3.32	14.58	2.77	1.66	15.02	0.58	0.097	0.208
E12-21	10.30	26.00	5.32	23.02	2.40	1.38	11.25	4.33	0.764	0.392
E12-22	10.00	25.46	4.83	20.85	2.23	1.32	11.65	4.09	-	0.382
E12-31	10.00	26.07	2.64	13.79	2.31	1.46	13.31	0.29	0.073	0.205
E12-32	10.00	26.88	3.47	14.18	2.31	1.45	9.76	0.35	0.080	0.215
E12-41	9.55	21.92	7.57	19.35	2.24	1.22	9.28	3.33	0.712	0.330
E12-42	10.55	22.19	5.20	19.84	3.57	1.95	9.72	3.45	-	0.326

(*) FIRST EXPERIMENT NOV 84