Use of extruded products from the exotic species *Plecostomus punctatus*. Part 2: Extrusion conditions

Utilización de productos extrudidos de la especie exótica Plecostomus punctatus. Parte 2: Condiciones de extrusión

Leonardo Tenorio-Fernández*[a,3], Ruth Pedroza-Islas[b], Josefina Morales-de-León[c], María del Carmen Durán-Domínguez-de-Bazúa[a]

[a]Universidad Nacional Autónoma de México, Facultad de Química, Departamento de Ingeniería Química, Laboratorios 301, 302, 303 de Ingeniería Química Ambiental y de Química Ambiental. Edificio E-3 Alimentos y Química Ambiental, Conjunto E, Circuito de la Investigación Científica S/N, Ciudad Universitaria, Alcaldía Coyoacán, 04510 Ciudad de México, México
[b]Universidad Iberamericana, Ciudad de México, México
[c]Instituto Nacional de Ciencias Médicas y Nutrición "Salvador Zubirán", Ciudad de México, México
Emails (*Correos-e*): leonardo_tenorio@hotmail.com, mcduran@quimica.unam.mx

*Corresponding author / *Autor a quien debe dirigirse la correspondencia*

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Abstract

In Part 1, it was established that to solve the problem that is presently going on at the Lic. Adolfo López-Mateos dam, known as "El Infiernillo", in Michoacán, Mexico, created by the involuntary introduction of *Plecostomus punctatus*, known in the area as "Devil Fish", that has proliferated and dominated on the existing fisheries, this research suggests its use as protein source for extruded feedstuffs for other species with a higher commercial value. The objective was to obtain an extruded balanced feedstuff based on *Plecostomus punctatus* meal. A laboratory scale extruder made in Mexico and an imported laboratory scale extruder were started and put into operation to obtain the desired feedstuffs considering as protein source a commercial fish meal to evaluate its performance. Once the best processing conditions were established, *Plecostomus punctatus* meal was introduced in the selected diet suggested in the literature for tilapia (Oreochromis spp.) cultivated in aquaculture. A modification of the diet composition was done introducing other carbohydrates (wheat flour instead of wheat "afrechillo" and wheat bran instead of rice bran). Extruders operating conditions were modified to obtain low density *pellets,* that could float in a water column, just as the tilapia commercial feedstuffs used as controls. A 2^3 experimental design for the three main variables (temperature, initial moisture content, and extruder screw velocity) was set, considering the best results obtained in preliminary experiments, assigning a negative value (-) to the lowest range and a positive one (+) to the highest range. For these experiments, feeding screw velocity was considered constant. Conclusions derived from this research were: An acceptable fillet contents was found in big specimens. However, the effort to extract it from the animals is important, and might affect the economic and technical feasibility of the process. If the fish is dried, its manual separation is much easier. There is an initiative to exploit "Devil Fish" fillets for human consumption, and considering that this project were viable, its residues would be adequate as part of the proposed diet, since its protein contents is 27%. The nutritional quality of the dehydrated fillet as well as the meal given by the methodology used make its unit operations satisfactory, since very low mass losses were found. Dried fillets might be used as "surimi" or to be directly sold as dried fish, such as Norway cod but to a much lower price. Particle size of the different meals, fish and carbohydrates, play a very important role in the feedstuffs physical characteristics before and after extrusion. Thus, all raw materials were previously pretreated to comply with particle size specifications. Grinding and sieving unit operations are very important for industrial scaling-up and should be optimized to reduce costs. Based on bromatological analyses, "Devil Fish" integral meal has the necessary protein contents for tilapia extruded feedstuffs. Carbohydrates needed in the feed mixture to obtain good physical characteristics (hydrostability, matter losses, and expansion percentage) were 28%, according to this research. It also complies with its protein requirements. Low cost Mexican extruder as it was originally built, even with some modifications carried out during these experiments has not the desirable characteristics for low density feedstuffs production required for tilapia. If used for shrimp or langoustine feedstuffs that should be denser is ideal. Results obtained in the Wenger X-5 extruder for the experimental design 2^3 indicate that temperature and initial moisture

3 Current affiliation / *Adscripción actual*: Cátedra CONACyT-Instituto Politécnico Nacional-Centro Interdisciplinario de Ciencias Marinas, La Paz, México

contents should be around 130°C and 40%, respectively. To obtain this temperature, steam at 600 kPa (6.0 kg_f/cm^2) is required. The best operating conditions found with *Plecostomus punctatus* meal to get the same hydrostability as the commercial feedstuff and a very good value for pellets expansion index, 2.1, at the extruder die exit, with the minimum matter losses, were 130°C at the extrusion zone, 40% initial moisture contents, 850 rpm for the extruder screw, and 25 rpm for the feeding screw. For future tests, as well as for scaling-up, the following recommendations were drawn: Steam should have a minimum pressure of 588.40 kPa (6 kg_f/cm^2) entering the Wenger extruder. If lower pressure steam, for example, below 539.37 kPa (5.5 kg_f/cm^2) enters, temperature drops and hydrostability characteristics (floatation) are also altered. As the two Wenger X-5 extruders used in these experiments were located in different institutions this variable could not be controlled (increase of boiler pressure). As "Devil Fish" meal is the main raw material in the process, it is necessary to optimize the drying or dehydration unit operation. It is recommended to open a research line to design and operate equipments for this process, taking advantage of climatic conditions of the zone (dry-tropic), with solar-gas aerothermal driers, for example. As wheat and soy are not products grown in the "El Infiernillo" dam agricultural area, new substitutes for the tilapia diet should be tested considering byproducts of the region, such as sorghum. Once the desired *pellets*[4] are obtained new economical analysis should be carried out to minimize costs. Finally, grinding and sieving operations should be optimized with the new materials available in the zone, such as sorghum, to minimize production costs.

Keywords: Extruded feedstuff, fish meal, *Plecostomus punctatus*, "Devil fish"

Resumen

En la Parte 1 se estableció que para solucionar el problema que actualmente se presenta en la presa Lic. Adolfo López-Mateos, conocida como "El Infiernillo", en Michoacán, México, creado por la introducción involuntaria de Plecostomus punctatus, conocido en la zona como "Pez Diablo", que ha proliferado y dominado a las pesquerías existentes, se sugiere en esta investigación su uso como fuente de proteínas para alimentos balanceados extrudidos para otras especies de mayor valor comercial. El objetivo fue obtener un alimento extrudido balanceado a base de harina de Plecostomus punctatus. Se puso en funcionamiento un extrusor a escala de laboratorio fabricado en México y un extrusor a escala de laboratorio importado para obtener los alimentos deseados considerando como fuente de proteína una harina de pescado comercial para evaluar su desempeño. Una vez establecidas las mejores condiciones de procesamiento, se introdujo la harina de Plecostomus punctatus en la dieta seleccionada sugerida en la literatura para la tilapia (Oreochromis spp.) cultivada en acuicultura. Se realizó una modificación de la composición de la dieta introduciendo otros carbohidratos (harina de trigo en lugar de afrechillo de trigo y salvado de trigo en lugar de salvado de arroz). Las condiciones de funcionamiento de los extrusores se modificaron para obtener 'pellets' de baja densidad, que pudieran flotar en una columna de agua, al igual que los alimentos comerciales de tilapia utilizados como controles. Se estableció un diseño experimental de 2^3 para las tres variables principales (temperatura, contenido de humedad inicial y velocidad del husillo o tornillo del extrusor), considerando los mejores resultados obtenidos en los experimentos preliminares, asignando un valor negativo (-) al rango más bajo y uno positivo (+) al rango más alto. Para estos experimentos, la velocidad del tornillo de alimentación se consideró constante. Las conclusiones derivadas de esta investigación fueron: Se encontró un contenido aceptable de filete en los ejemplares grandes. Sin embargo, el esfuerzo por extraerlo de los animales es importante y podría afectar la viabilidad económica y técnica del proceso. Si el pescado está seco, su separación manual es mucho más fácil. Existe una iniciativa para la explotación de filetes de "Pescado Diablo" para consumo humano, y considerando que este proyecto era viable, sus residuos serían adecuados como parte de la dieta propuesta, ya que su contenido de proteína es del 27%. La calidad nutrimental del filete deshidratado así como la harina obtenida por la metodología empleada hacen que sus operaciones unitarias sean satisfactorias, ya que se encontraron pérdidas de masa muy bajas. Los filetes secos pueden utilizarse como "surimi" o venderse directamente como pescado seco, como el bacalao noruego, pero a un precio mucho más bajo. El tamaño de las partículas de las diferentes harinas, pescados y carbohidratos, juegan un papel muy importante en las características físicas de los alimentos antes y después de la extrusión. Por lo tanto, todas las materias primas fueron pretratadas previamente para cumplir con las especificaciones de tamaño de partícula. Las operaciones de la unidad de molienda y tamizado son muy importantes para la aplicación industrial y deben optimizarse para reducir los costos. Con base en los análisis bromatológicos, la harina integral de "pez diablo" tiene el contenido de proteína necesario para los alimentos extrudidos de tilapia. Los carbohidratos necesarios en la mezcla de alimento para obtener buenas características físicas (hidrostabilidad, pérdidas de materia y porcentaje de expansión) es del 28%, según esta investigación. También cumple con sus requerimientos proteicos. El extrusor mexicano de bajo costo tal como se construyó originalmente, incluso con algunas modificaciones realizadas durante estos experimentos, no presentó las características deseables para la producción de alimentos de baja densidad requeridos para la tilapia. Si se utiliza para piensos para camarones o langostinos sí resulta ideal ya que son más densos. Los resultados obtenidos con el extrusor Wenger X-5 para el diseño experimental 23 indicaron que la temperatura y el contenido de humedad inicial deben estar alrededor de 130°C y 40%, respectivamente. Para obtener esta temperatura, se requiere vapor a 600 kPa (6.0 kg_f/cm^2). Las mejores condiciones de operación encontradas con la harina de Plecostomus punctatus para obtener la misma hidrostabilidad que el alimento comercial y un muy buen valor para el índice de expansión de los pellets, de 2.1, a la salida de la boquilla o matriz del extrusor, con las pérdidas mínimas de materia, fueron 130°C en la zona de extrusión, 40% de contenido de humedad inicial, 850 rpm para el tornillo de extrusión y 25 rpm para el tornillo de alimentación. Para las pruebas futuras, así como para la ampliación, se elaboraron las siguientes recomendaciones: El vapor debe tener una presión mínima de 588.4 kPa (6 kg_f/cm^2) entrando al extrusor Wenger. Si entra vapor a presión más baja, por ejemplo, por debajo de 539.37 kPa (5.5 kg_f/cm^2), también se alteran las caídas de temperatura y las características de hidrostabilidad (flotación). Como los dos extrusores Wenger X-5 utilizados en estos experimentos estaban ubicados en diferentes instituciones, esta variable no se pudo controlar (aumento de la presión de la caldera). Dado que la harina de "Pez Diablo" es la principal materia prima del proceso, es necesario optimizar el funcionamiento de la unidad de secado o deshidratación. Se recomienda abrir una línea de investigación para diseñar y operar equipos aprovechando para estas operaciones unitarias las

4 Derived from Latin "*pila*", soft matter ball, such as snow, clay, etc., easily malleable

condiciones climáticas de la zona de la presa (trópico seco), empleando secadores aerotérmicos solar-gas, por ejemplo. Dado que el trigo y la soya no son productos cultivados en el área agrícola de la presa "El Infiernillo", se deben probar nuevos sustitutos para la dieta de la tilapia considerando subproductos de la región, como el sorgo. Una vez que se obtengan los gránulos ('pellets' [4]) deseados, se debe realizar un nuevo análisis económico para minimizar los costos. Finalmente, las operaciones de molienda y tamizado deben optimizarse con los nuevos materiales disponibles en la zona, como el sorgo, para minimizar los costos de producción.

***Palabras clave**: Alimentos balanceados extrudidos, harina de pescado, Plecostomus punctatus, "Pez Diablo"*

Introduction

The dam "*Lic. Adolfo López Mateos*", known as "*El Infiernillo*" ("The Little Hell", in Spanish) is an embankment dam on the Balsas river near La Unión, a city in the Mexican state of Guerrero. The dam supports a hydroelectric power station containing six turbine-generators for a total installed capacity of 1,120 MW. The dam is 149 m (489 ft) high, 344 m (1,129 ft) long and is owned by *Comisión Federal de Electricidad* (Mexico's Federal Electricity Commission), the state owned electricity company. Its first generator was operational on January 25, 1965 (CFE, 2010, 2012; Manzolillo and Egea, 2001). Placement of the dam embankment began in August 1962, and on December 7, 1963, the dam was topped off. The diversion tunnels were closed and the reservoir began to fill on June 15, 1964 (Wilson, 1973). A present problem in the embankment "El Infiernillo" is the accidental introduction of a fish species *Plecostomus punctatus* colloquially known in the area as "devil fish" ("Pez Diablo", in Spanish). This fish has proliferated dominating the existing population, particularly tilapia[5], that was introduced to support the communities living in the land that was covered by the embankment allowing them to change from agricultural activities to fishing ones. This environmental problem has created a complex socio-economical issue in the three communities living around the embankment, known in Mexico as "*municipios*".

The objective of this research was to give a short term solution, using the devil fish as a source of protein to produce feedstuffs, using the extrusion process, for tilapia cultivation in selected confined areas of the embankment to recover the tilapia population and to desirably make the devil fish disappear of the dam reverting the problem.

The present research goals were the following. The first one was to obtain a meal from whole *Plecostomus punctatus* catched with different fishing nets as tilapia, a part already carried out training the fishermen, as well as the characterization of dry meals from whole fish, filleted fish, and fish residues using bromatological analyses (Tenorio-Fernández, 2009; Tenorio-Fernández et al., 2009a,b).

With the meal, the objective of this part of the research was the second goal, that included: (a) Production of a balanced feedstuff based on *Plecostomus punctatus* whole dry meal as protein source, using the diet suggested in the literature for tilapia (Oreochromis spp.) grown in aquaculture installations; (b) starting-up and operating of a laboratory scale extruder constructed in Mexico to evaluate its performance preparing pellets with the *Plecostomus punctatus* whole dry meal based feedstuff; (c) starting-up and operating of a commercial laboratory scale extruder to evaluate its performance too; and (d) Comparison of the *pellets*[6], with a commercial counterpart evaluating its floatability in a water column, considering hydrostability and mass loss in water.

Methodology

The methodology used in this research is presented in Figure 1.

5 ***Oreochromis*** is a large genus of tilapiine cichlids, fishes endemic to Africa and the Middle East. Several species from this genus have been introduced far outside their native range, and are important in aquaculture

6 Word that does not exist in the Spanish language (*Diccionario de la Academia de la Lengua Española*), derived from Latin "*pila*", soft matter ball, such as snow, clay, etc., easily malleable / *Palabra que no existe en español aunque es derivada del Latín "pila", pelota de material suave como la nieve, la arcilla, etc.*

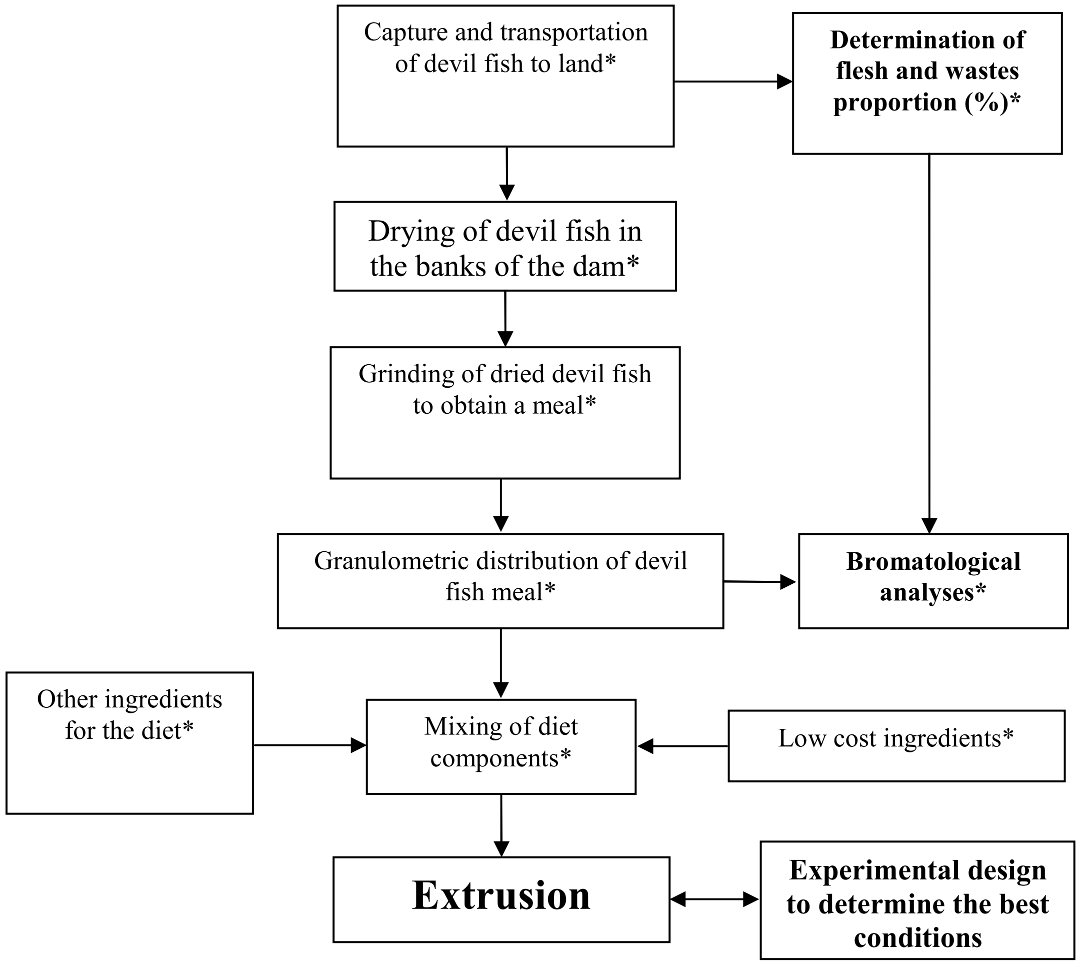


Figure 1. Experimental conditions (*these operations were the core of Part 1)

Lab scale production of devil fish meal

Freshly captured devil fish were refrigerated and frozen to -5ºC, and transported to Mexico City (Photograph 1). As mentioned in Part 1, once in the laboratory they were directly vacuum dried in a J.P. Devine Co., model 3SPC tray drier. Operating conditions were 41 kPa (12 in Hg) at a fluctuating temperature (60 to 80°C) during roughly 18 hours, as average drying time. Heat was provided by saturated steam. The batch was of 25.9±0.3 kg of frozen fish. Once dried, it was very easy to separate the plates (external part or exoskeleton of the fish) and spines (inner skeleton of the fish) from the dehydrated muscle part. Samples of dried fillets of 1 kg, as well as of whole dried fish, and of the wastes (plates and spines) were used to determine its bromatological characteristics (AOAC, 1990). Grinding was carried out in an Imperial teeth mill. The resulting meals, of whole devil fish, of fillets, and of wastes, were stored in ambar glass jars at -5°C in a freezer. Granulometry of meals was determined using a Montinox sieve with nine different mesh sizes trays: 10, 20, 30, 40, 50, 60, 70, 80, and 100. Samples of 250 g from each meal were set in the equipment during 20 minutes, weighing the amounts retained in each mesh tray and calculating the equivalent percentage from the total sample weight. Graphs were drawn to corroborate average particle size. For those cases where a higher retention was obtained intermediate mesh sizes were used.

Photograph 1. Devil fish (*Plecostomus punctatus*)

Bromatological analyses
Moisture content, crude protein, crude fat, crude fiber, and ash contents were determined for whole devil fish meal, fillet meal, and wastes meal, using the AOAC methods (1990) and, by difference to 100%, it was assumed that the rest were carbohydrates (Aragón and Novoa, 1994). Non proteic nitrogen for wastes meal was also evaluated following the methodology established by Tejada-de-Hernández (1992), to assess the real protein content of the feedstuffs (25 to 35% protein).

For preliminary studies in the extruders a commercial fish meal previously characterized by bromatological analyses was employed.

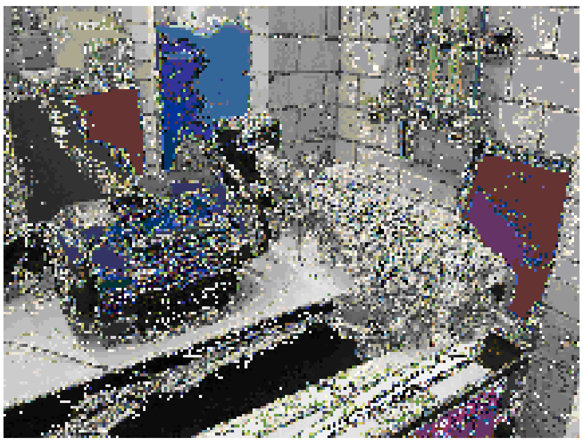
Extrusion tests
A commercial laboratory scale extruder Wenger X-5 and one constructed in Mexico for research purposes (Durán-Domínguez, 1978; Sánchez-Tovar y Durán-Domínguez-de-Bazúa, 2014) were used. Both extruders were of single screw. Experiments were performed in two phases: The first phase consisted of preliminary extrusion tests to determine the effect of the operating variables in the characteristics of the extruded products. For these experiments commercial fish meals were used. The second phase was performed using the devil fish whole meal in combination with the other ingredients of the feedstuff.

Low cost extruder (E-Laboratorio 301**)**
Several tests were performed in this extruder to evaluate the technical characteristics of the feedstuffs prepared following the recommended proportions for tilapia (Gaxiola, 2007). Modifications were carried out to reach higher output pressures and to obtain higher expansion in the pellets coming out of the extruder die. Isolation of the extruder barrel was also added to have a better control of extrusion temperature. The screw rotational speed was also changed to increase the residence time within the extruder. Table 1 shows the process conditions and the process variables for the low cost laboratory scale extruder constructed in Mexico (Photograph 2).

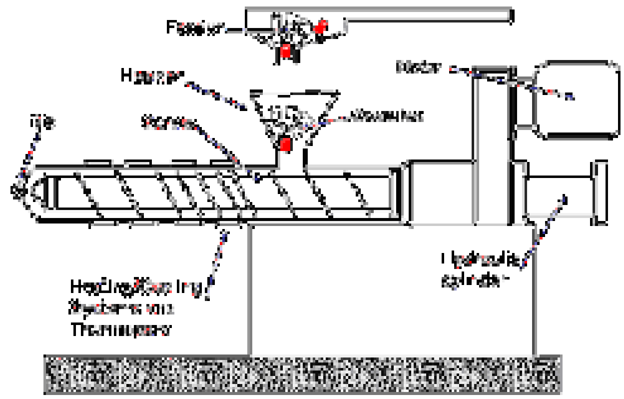
Table 1. Process specifications and process variables for the low cost extruder constructed in Mexico

Equipment specifications	Parameter
Barrel length	40 cm
Die diameter	4 orifices, 5.5 mm each
Screw type	Rapid transition metering type

Equipment specifications	Parameter
Thermopars position	No thermopars
Screw diameter	3.5 cm
Motor power	1.5 hp
Barrel diameter	6.5 cm
Process variables	**Parameter**
Screw speed range	30 rpm
External temperatures range	50 to 350°C
Transition temperature Extrusion temperature	(In this equipment temperatures are measured outside the barrel with two pyrometers located in the transition and extrusion zones)



Photograph 2a. Low cost laboratory scale extruder constructed in Mexico



Photograph 2b. Principle of operation of an extruder

The operation of this low cost extruder is much simpler than that of a Wenger X-5 extruder but, essentially, its performance is similar. The increase in temperature of the extruder barrel is done through external electric resistances, one in the extrusion zone and the other one in the transition zone. These are controlled by two thermostats. Feeding of the diet mixture containing the fish meal is manual and direct to the extruder feeding zone. Screw speed is constant. To modify it the transmission pulley is changed. It has an exchangeable die with different orifices. The one used had four orifices of 5.5 mm diameter each.

Wenger X-5 extruder

In Table 2 specifications of the extruder Wenger X-5 (Sabetha, Kansas, U.S.) are presented. Operating conditions range are the result of an important number of preliminary tests performed both in this study and in previous research tests (Gaitán-Hinojosa, 1996). Its operation is relatively simple: Steam is directly fed from a boiler at 5.5 kg_f/cm^2 (depending upon the heat needed in the extrusion process) to the barrel rings[7] through 5 valves, manually closed or opened according to the needs of the experiment to obtained the desired expansion of the extruded product (Photographs 3a,b). Cooling water can be introduced into the rings for controlling temperature of the extruding mass. If necessary valves are opened and closed to reduce or to increase temperature. The temperature in the rings 2, 5, 7, and 8 is measured using a pyrometer of four channels connected to 4 thermopars West-TCA 0039 of *constantane iron* type J. For this study temperatures were varied between 110 and 130°C (see Table 2), depending upon the different tests carried out. Once the temperature was controlled the

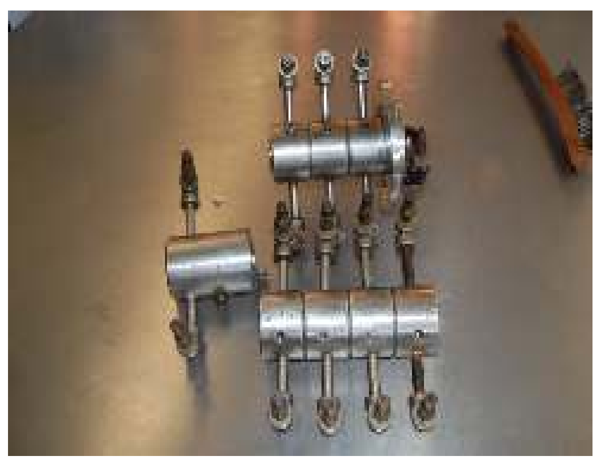
7 The barrel is divided into 8 doughnut-like rings. The internal wall of the rings is directly in contact with the extruding material and the inner part of the doughnut-like volume holds the steam heating that internal wall (Photograph 3)

motor of the extruder was started and with an optical tachometer screw velocity was measured in revolutions per minute (rpm). If measurements were out of the expected range motor pulleys were opened or closed, turning the mechanical handle (crank) below the extruder clockwise to increase the rotational speed in rpm or vice versa to decrease them. Once the screw velocity was set, the motor to move the feeding screw was started and adjusted using the optical tachometer. Similar to the extruder screw, if it is out of range, the little handle below the feeding hopper was adjusted.

Table 2. Process specifications and process variables for the Wenger X-5 extruder

Equipment specifications	Parameter
Barrel length	40 cm
Number of rings	8
Die diameter	4 mm
Screw type	Variable channel and depth
Feeding screw velocity	35 rpm
Thermopars location	Rings 2, 5, 7, 8
Screw diameter	2.5 mm
Motor power	5 hp
Extruder screw velocity variation range	352 – 1 000 rpm
Compression ratio	2:1
Steam pressure	From 3.5 to 6.0 kg_f/cm^2
Process variables	**Parameter**
Temperature range in the last ring (8)	From 110 to 130°C
Temperature of extrusion (T_1)	
Transition temperatures range for T_2 and feeding T_3	$T_{ambient}$-130°C
Moisture content of extruding mix range	From 36 to 42%
Screw velocity range	From 750 to 900 rpm

Photograph 3a. Wenger X-5



Photograph 3b. Wenger X-5 extruder rings

Feeding velocity is a very important variable of the process, since material flow is proportional to residence time within the extruder, and with it, specific conditions for the products (for example, more or less dense feedlots). Once the feeding motor is started, the feeding hopper starts to become full, without exceeding 50% of its capacity to avoid any congestion in the extruder. Another very important variable is the temperature since the extruder loses heat in contact with the material in the feeding and transition zones. If necessary, the steam valve should be opened to re-establish the desired temperature. For this action it is important to have high pressure steam availability. If this situation is not possible, another form to increase the temperature in the extrusion zone is reducing the exit diameter of the products, since diameter reduction increases friction and retropressure. The changing of the exit dies is relatively easy for either smaller or bigger diameters. In these experiments it was found that the heaters to produce steam located in two cooperating institutions[8] reached steam pressures of 6 kg_f/cm^2 (588.4 kPa) allowing maximum extrusion temperatures of 130°C. Thus, a new die with orifice of 2 mm was installed to substitute the existing one of 4 mm diameter. The extruded materials were received in trays previously identified with the corresponding run, since changes in the runs particularly process variables were carried out with the extruder operating continuously, separating and rejecting the product obtained in the interphases or transitions between each test (Photograph 4a). If temperature was to be changed then the extruder should be stopped and cleaned, particularly the screw (Photograph 4b).

Photograph 4a. Preliminary experiments with the Wenger X-5 extruder

Photograph 4b. Screw of the Wenger X-5 extruder

Extruded products were dried at room temperature since *"pellets"* obtained do not contain a high moisture content, resulting from the flash evaporation occurring in the extruder die. In Figure 2, the process flow diagram to produce the fish feedlots from grinding to extrusion is shown (ingredients of the feedlots such as soymeal, wheat flour, etc., were identified with their initials in Spanish: *HS* [*harina de soya*], *HT* [*harina de trigo*], etc.).

Experimental design

Figure 3 shows the entrance variables (independent variables), the controlling variables (constant ones), and the exit variables (dependent variables or response ones) (Montgomery, 2006). Among the first ones -at the beginning of the study- were temperature, initial moisture content, and screw velocity. As preliminary experiments were advancing, it was added a fourth variable, feeding velocity, to control residence time of the material. The second set of variables that were kept constant were extrusion pressure, type and length of the screw, diet composition, and residence time in the extruder. Hydrostability, loss of matter of the "*pellets*", and "*pellets*" expansion index were the dependent or response variables.

8 In Mexico City there are at least two identical extruders Wenger X-5 model, one in the *Instituto Nacional de Ciencias Médicas y Nutrición "Salvador Zubirán"*, and the second one in the *Universidad Iberoamericana in Santa Fe*. Both were used in this research

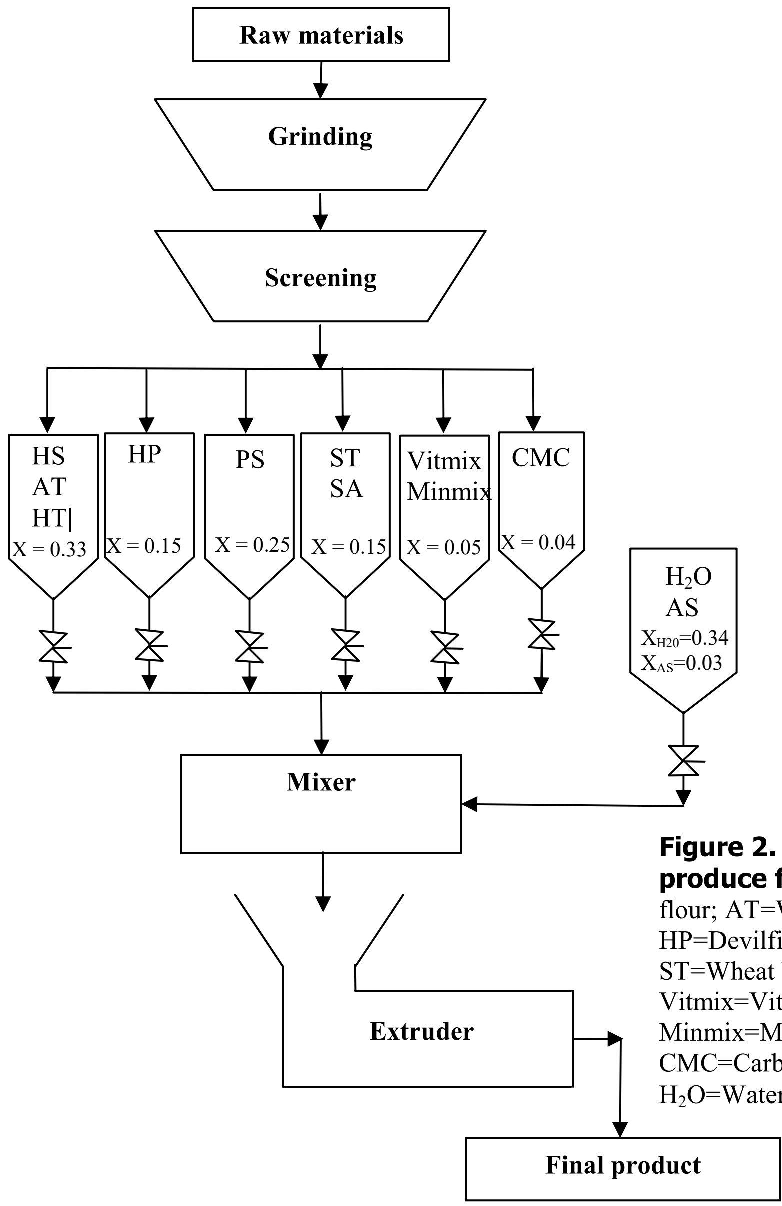


Figure 2. Process diagram to produce fish feedlots (HS=Sorghum flour; AT=Wheat grits; HT=Wheat flour; HP=Devilfish meal; PS=Soy paste; ST=Wheat bran; SA=Rice bran; Vitmix=Vitamins mixture; Minmix=Minerals mixture; CMC=Carboxymethylcellulose; H_2O=Water, AS=Soy oil)

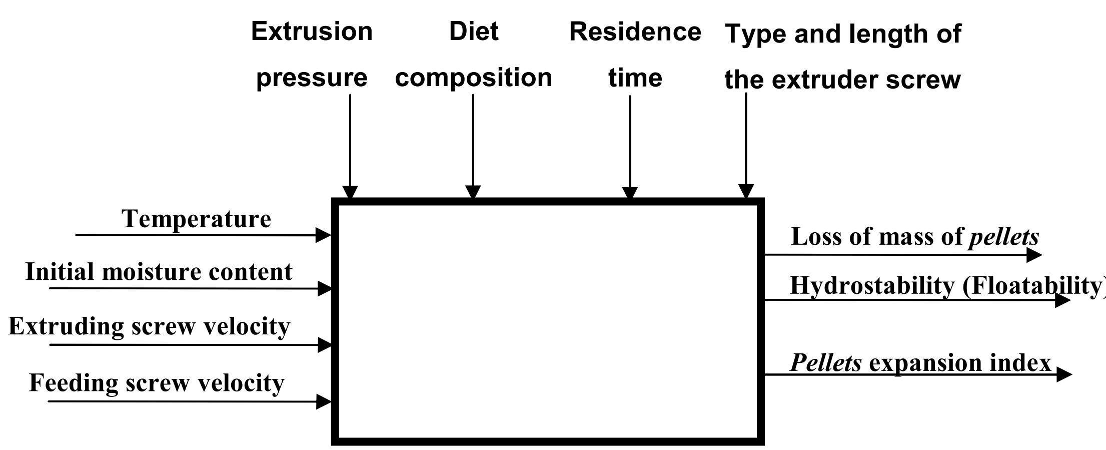


Figure 3. Experimental design

The interaction between the temperature within the extruder, the initial moisture content of the feedlot mixture, the extruder screw velocity, and the feeding screw velocity define the extruder pressure. These variables also define the expansion of the product at the die, and thus, its hydrostability (floatability) as well as the loss of matter in the aqueous medium of the product (Pedroza-Islas, 2000). Ranges for these four independent variables were specified using previous results obtained by Gaitán-Hinojosa (1996). To each variable a low and a high level were assigned (Table 3).

Table 3. Levels for the independent variables

Level	Temperature (°C)	Initial moisture content (%)	Extruder screw velocity (rpm)	Feeding screw velocity (rpm)
Low	110	36	750	25
High	130	40	850	35

These ranges were set considering preliminary tests selecting as independent variables to temperature and initial moisture content. The extruder screw velocity and the feeding screw velocity were kept constant at 800 and 35 rpm, respectively. Also, hydrostability (floatability) was considered as a qualitative dependent variable or response important variable, since this characteristic is vital for tilapia feeding (Figure 4).

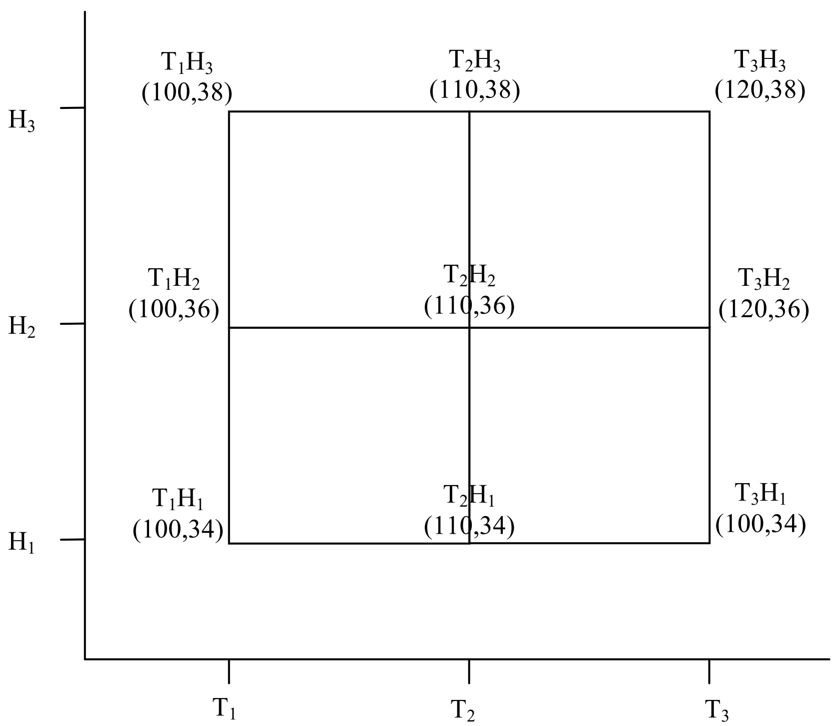


Figure 4. Matrix for operational variables (T=Temperature, H=Initial moisture content)

Factorial design 2^3 and factorial design 2^2

With the results obtained in the preliminary tests an experimental design 2^3 set, for the three entrance variables (temperature, initial moisture content, and extruder screw velocity), the ranges with the best conditions were assigned a negative value (-) for the lower limit of the range, and a positive one (+) for the higher limit. In this case, the feeding screw velocity (feeding velocity) was maintained constant. Table 4 shows the values for the 8 runs generated with this experimental design. It is important to mention that a replica for each of these tests was carried out. Based on these results, the influence of the feeding screw velocity influence on the process was assessed. This time temperature and initial moisture content were kept constant with the best values found in the experimental design 2^3, considered the best ones for the feedlots, with the posible improvement of the residence time due to the feeding velocity. For the next phase of the preliminary experiments, a factorial experiment 2^2, varying the extruder and the feeding screws velocities were performed. Two leveles were also considered (- low and + high). Table 5 shows the range of variation of the four tests carried out. In this case also replicas for each one were performed. Once the best operational ranges were known from these preliminary experiments, extrusion experiments using whole meal of *Plecostomus punctatus* were carried out.

Table 4. Factorial design 2^3

Run	Temperature (°C)		Initial moisture content (%)		Extruder screw velocity (rpm)	
1	-	110	-	36	-	800
2	+	130	-	36	-	800
3	-	110	+	40[9]	-	800
4	+	130	+	40	-	800
5	-	110	-	36	+	850[10]
6	+	130	-	36	+	850
7	-	110	+	40	+	850
8	+	130	+	40	+	850

Table 5. Factorial design 2^2

Run	Feeding screw velocity (rpm)		Extruder screw velocity (rpm)	
1	-	25	-	750
2	+	35	-	750
3	-	25	+	850
4	+	35	+	850

Statistical analyses

Experimental results were statistically evaluated using a commercial computer program (Statgraphics, version 5.1). Statistical differences at 5% for all analysis were considered.

Results and discussion

Vacuum drying and grinding

At the end of the drying process 4.535 kg of dry fish were obtained. Moisture final content was 5% (dry basis), with a drying efficiency to obtain 21.7% dry matter. Before grinding the dehydrated fish it was observed that the dehydrated fillets were intact and were easily separated without any problem. To know the amount of muscle of each specimen, dehydrated fish "fillets" were separated and, from the total mass of each specimen these were in average of 17%. If a high quality ground meal with a high content of protein were to be the desirable final product these parts of the fish might be taken (for surimi, for example). In the grinding operation, efficiency was 95%, obtaining 4535 g meal ready to be mixed and processed.

9 Tests were done with initial moisture contents up to 42% d.b., but the resulting mixtures had a fluid consistency that produced extruded materials too plastic and inadequate. Thus, the maximum initial moisture content was 40% dry basis

10 Tests were done up to 900 rpm but the die was blocked with material and the mixture adhered to the wall of the inner surface and the screw blades of the extruder. Thus, the maximum rotational velocity was 850 rpm

Preliminary extrusion tests

The preliminary extrusion tests in the equipment fabricated in Mexico allowed the familiarization with the process, to design the experiments using as dependent variables hydrostability (floatability), loss of matter in the aqueous medium, and the evaluation of the *pellets* expansion index (Tenorio-Fernández et al., 2009a,b, 2011). Runs 1 and 2 rendered feedstuffs that did not float (tilapia feedstuffs must float, while crustacean should take its feedstuffs from the bottom). Table 6 shows the process conditions and the results obtained.

Table 6. Preliminary tests using the extruder fabricated in Mexico with the original specifications

Run	Mixture (g)	Initial moisture content (%)	Transition zone temperature (°C)	Extrusion zone temperature (°C)	Hydrostability
1	500	28	150	250	Submersible
2	500	28	150	250	Submersible

Since '*pellets'* were dense and only apt for crustacean, the following actions were conducted:

1. Increasing of the pulley diameter increasing the residence time of the feed mix
2. Increasing initial moisture content to 30%
3. Isolation of the extruder surface and the die zone to improve heat transfer and increasing extrusion temperature
4. Substitution of the die, from one with eight orifices of 5 mm to one with four orifices of 5 mm increasing the residence time

Table 7 and Photograph 5 show the results for these preliminary tests (runs 3 to 5).

Table 7. Preliminary tests using the extruder fabricated in Mexico with the original specifications considering the changes 1 to 4

Run	Mixture (g)	Initial moisture content (%)	Transition zone temperature (°C)	Extrusion zone temperature (°C)	Hydrostability
3	500	30	250	300	Submersible
4	500	30	250	300	Submersible
5	500	30	250	300	Submersible

Photograph 5. Samples of extruded products using the extruder fabricated in Mexico

These runs 3-5 corroborated that this extruder is useful to produce crustacean feedstuffs but not tilapia ones, since it is a low temperature-low pressure system (Luna-Rodríguez et al., 2008).

Thus, preliminary experiments were started in a laboratory scale Wenger X-5 extruder, a high pressure-high shear rate equipment. Four preliminary tests were run to recognize the equipment features. Table 8 presents the results. Once these preliminary recognition tests were done, the experimental matrix presented in the methodology was performed. Table 9 shows the process conditions and the results obtained. Extruder and feeding screw velocities were maintained constant at 800 rpm and at 35 rpm, respectively.

Table 8. Preliminary tests using the Wenger X-5 extruder

Run	Mixture (g)	Initial moisture content (%)	T_1 (°C)	T_2 (°C)	T_3 (°C)	Hydrostability
6	500	28	50-60	40-50	25-35	Submersible
7	500	30	80-90	60-70	40-50	Submersible
8	500	32	90-100	70-80	50-60	Submersible
9	500	34	100-110	80-90	60-70	Submersible

Table 9. Preliminary tests to define process conditions (original diet)

Run	Experiment	Initial moisture content (%)	T_1 (°C)	Hydrostability	Expansion index
1	T_1H_1	34	100	Submersible	1.39
2	T_1H_2	36	100	Submersible	1.41
3	T_1H_3	38	100	Submersible	1.24
4	T_2H_1	34	110	Submersible	1.24
5	T_2H_2	36	110	Submersible	1.36
6	T_2H_3	38	110	Submersible	1.35
7	T_3H_1	34	120	Submersible	1.19
8	T_3H_2	36	120	Submersible	1.26
9	T_3H_3	38	120	Submersible	1.32

Temperature reported in Table 9 corresponds to the extrusion zone near the die (T_1). The two other temperatures, from the transition zone (T_2 and T_3) were maintained between 80 and 100°C. *Pellets* expanded with fractures and although much less dense that the ones produced in the low cost extruder allowed water to penetrate reducing its floatability.

According to these results the diet composition was modified. Wheat meal was substituted by wheat flour to increase available starches and to reduce particle size, to warrant gelatinization of the feedstuff and a higher expansion, and thus, a better hydrostability. Table 10 shows the obtained results.

Experimental design 2^3

Results for the experimental matrix 2^3, described in Methodology included, as independent variables, temperature for the extrusion zone, initial moisture content, and extrusion screw velocity. Feeding screw velocity was maintained constant at 35 rpm. Table 11 presents process conditions and results obtained. For hydrostability, significant diferences ($p<0.05$) were obtained for the initial moisture content and the temperature, with better results for the higher level. For the interaction with the extruder screw velocity, the lower level was better.

Table 10. Preliminary tests to define process conditions (diet with wheat flour)

Run	Experiment	Initial moisture content (%)	T_1 (°C)	Hydrostability	Expansion index
1	T_1H_1	34	100	Submersible	1.38
2	T_1H_2	36	100	Submersible	1.28
3	T_1H_3	38	100	Submersible	1.37
4	T_2H_1	34	110	Submersible	1.32
5	T_2H_2	36	110	Submersible	1.79
6	T_2H_3	38	110	Submersible	1.44
7	T_3H_1	34	120	Submersible	1.50
8	T_3H_2	36	120	Floating	1.24
9	T_3H_3	38	120	Floating	1.55

Table 11. Results for the experimental design 2^3. Definitive tests

Run	T_1 (°C)		H (%)		Extruder screw velocity (rpm)		Hydrostability (%)	Loss of matter (%)	Expansion index
1	-	110	-	36	-	800	14	2.28	1.2250
2	+	130	-	36	-	800	11	2.25	1.1603
3	-	110	+	40	-	800	33	2.01	1.3407
4	+	130	+	40	-	800	62	1.78	1.1976
5	-	110	-	36	+	850	33	2.56	1.3525
6	+	130	-	36	+	850	4	1.06	1.3353
7	-	110	+	40	+	850	49	1.07	1.2290
8	+	130	+	40	+	850	12	0.32	1.1870

Experimental design 2^2

Once the results for the experimental design 2^3 were obtained, the effect of the fourth variable, feeding screw velocity, was assessed. Previous results showed that extrusion temperature and initial moisture content as independent variables give the best results in its higher levels (130°C and 40%, respectively). Thus, these two parameters were maintained constant and the feeding screw velocity variable was introduced. Table 12 presents the results of the four experiments corresponding to the experimental design 2^2.

Table 12. Results of the experimental design 2^2

Run	Feeding screw velocity (rpm)		Extruder screw velocity (rpm)		Hydrostability (%)	Loss of matter (%)	Expansion index
1	-	25	-	750	99	2.5292	1.1300
2	+	35	-	750	85	1.0666	1.2100
3	-	25	+	850	85	1.2546	1.4153
4	+	35	+	850	81	1.1617	1.4606

From these results for hydrostability, it is considered important to emphasize that in the first 60 minutes the values were suitable, considering that tilapia ingests the feedstuff in this lapse. For this case, it was in this period where the values were higher. Also, the values corresponding to the analysis of matter loss in the aqueous medium is reported to 240 minutes, that was the higher limit to which the feedstuffs were exposed in the water column. These two tests and the previous ones were performed at 35 rpm for the feeding screw velocity. The modification in this experiment to the feeding screw velocity of 25 rpm with an extruder screw velocity of 750 rpm also gave good results. Run 3 is the one with the best behavior since the product is the most stable, with values higher than those for commercial feedstuffs, labeled "CB" (Tenorio-Fernández et al., 2011).

According to these results, the four experiments of this design had an acceptable performance, indicating that the results obtained in the previous experiment (2^3) gave the idoneous extrusion temperature and initial moisture content. The C3 experiment showed that the products obtained had similar hydrostability compared with the commercial feedstuff, lost less matter than the commercial product, and had an excellent expansion. Table 13 resumes these process conditions and the specifications to produce the extruded feedstuff from commercial fishmeal.

Table 13. Final process conditions and specifications for the Wenger X-5 extruder

Equipment specifications	**Parameter**
Barrel length	40 cm
Number of rings	8
Die diameter	4 mm
Screw type	Variable channel and depth
Thermopars location	Rings 2, 5, 7, 8
Screw diameter	2.5 mm
Motor power	5 hp
Compression ratio	2:1
Steam pressure	From 3.5 to 6.0 kg_f/cm^2
Process variables	**Parameter**
Temperature in the last ring (8)	**130°C**
Initial moisture content of the extruding mixture	**40%**
Extruder screw velocity	**850 rpm**
Feeding screw velocity	35 rpm

Production of the feedstuff for tilapia using Plecostomus punctatus meal

Up to this point of the research, commercial fish meal was the main ingredient of the diet to study the performance of the Wenger X-5 extruder with the different operational variables (Table 13). Once they were established the diets were prepared using the *Plecostomus punctatus* meals. For the first tests in the Wenger X-5 extruder, the new die with 2 mm diameter of the orifice was used. This condition allowed the equipment to maintain the temperatute at 130°C. Another interesting feature was that the product had a final diameter of 4 a 6 mm, perfectly suitable for the socalled tilapia fattening stage.

Results for these tests are shown in Table 14. The 2 mm die promotes the increase of the temperature in the last section of the extruder by the friction provoked and the retro or backpressure within the inner part of the extruder. Better results for all the response variables were obtained when compared with the die with the orifice of 4 mm in the die. Photograph 6 shows one of the tests for hydrostability for the *pellets* using the die with the orifice of 2mm. The *pellets* have an excellent behavior since almost all of them are on the surface. This behavior, in combination with the good apparent gelatinization, is what it is giving this acceptable performance for the feedstuff produced with the *Plecostomus punctatus* meal, especially with the 2 mm diameter die (Figures 5a,b).

Table 14. Results for the production of extruded feedstuffs for tilapia based on *Plecostomus punctatus* meals using 4 and 2mm diameter dies in a Wenger X-5 extruder

Time	Hydrostability		Loss of matter		Expansion index	
	4mm	2mm	4mm	2mm	4mm	2mm
1	72.59	98.96	0.2387	0.4812	1.4595	2.1695
15	60.26	87.96	0.4330	0.6189		
30	51.46	94.54	0.5486	0.9210		
60	64.41	93.18	0.6100	0.7928		
120	58.72	92.45	0.7516	0.7898		
240	59.10	90.22	1.0193	0.9520		

Photograph 6. Test for hydrostability of tilapia's feedstuffs using *Plecostomus punctatus* meal with other nutrients, example of use of die with orifice diameter of 2mm

The final bromatological analyses of the ***Plecostomus punctatus*** extruded pellets (Table 15) was carried out to corroborate the nutritional quality, particularly protein content, compared with a commercial product made for tilapia feeding using undetermined fish meal. Results are quite satisfactory, particularly for the growth stage or fattening stage of tilapia (Oreochromis spp.), when the requirement is 25% crude protein (Morales, 2003).

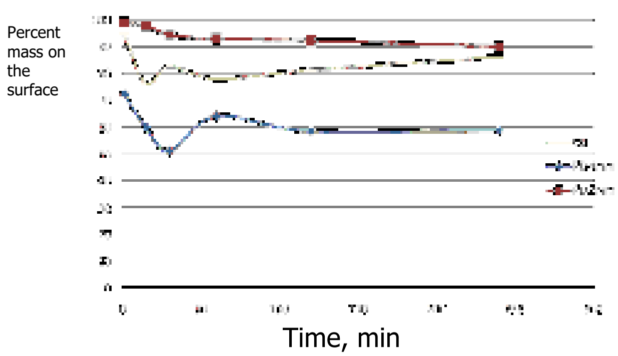


Figura 5a. Results for hydrostability of the extruded product containing *Plecostomus punctatus* dry meal using dies with 2 and 4 mm orifice diameter (PD2mm y PD4mm) compared with a commercial tilapia feedstuff (CB)

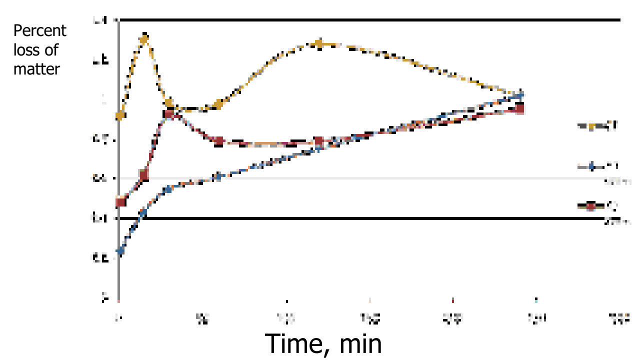


Figura 5b. Results for the loss of matter for the extruded product containing *Plecostomus punctatus* dry meal using dies with 2 and 4 mm orifice diameter (PD2mm y PD4mm) compared with a commercial tilapia feedstuff (CB)

Table 15. Bromatological analyses for the extruded pellets made with *Plecostomus punctatus* compared with a commercial tilapia feedstuff (CB)

Chemical analysis	**% Wet basis**		**% Dry basis**		**Control of errors**	
	Extruded *pellets*	Commercial product	Extruded *pellets*	Commercial product	Extruded *pellets*	Commercial product
Dry matter	99.26	94.42	-	-	-	-
Moisture	0.74	5.58	-	-	0.060%	0.047%
Crude protein (Nitrogen*6.25)	**24.27**	**22.76**	**24.45**	**24.105**	0.056%	**0.057%**
Ether extract	6.78	3.83	6.83	4.05	**0.030%**	0.081%
Ashes	12.61	14.79	12.70	15.66	0.086%	0.218%
Crude fiber	5.38	4.84	5.42	5.12	0.240%	0.378%
Nitrogen free extract	50.21	48.20	50.58	51.04	-	-

Conclusions and perspectives

Based on the information obtained in this research, according with the objective of obtaining a balanced extruded feedstuff for tilapia being its main ingredient a dried meal from *Plecostomus punctatus* to be suitable for feeding farm fish that require floating characteristics and a good hydrostability, the following conclusions can be drawn:

- The low cost lab extruder fabricated in Mexico, according to its original design and the modifications done in this research, is suitable for producing high density feedstuffs that can be fed to crustacean, either shrimp or prawns. For fish, such as tilapia (Oreochromis spp.), feedstuffs have not the desirable characteristics of floatability
- The results obtained in the Wenger X-5 extruder for the experimental design 2^3 indicated that an extrusion temperature of a 130°C and an initial moisture content of 40% are idoneous to obtain good extruded products. To reach this temperature with a 4 mm orifice die it is required to supply steam with a pressure equal or higher than 6.0 kg_f/cm^2 (ca. 590 kPa)
- The experiment defining the best operational conditions to produce a balanced feedstuff using *Plecostomus punctatus* dry meals is the one with an extrusion zone temperature of 130°C, an initial moisture content in the feeding mixture of 40%, an extruder screw rotational velocity of 850 rpm, a feeding screw rotational velocity of 25 rpm, and a die with an orifice diameter of 2 mm, being its hydrostability similar or even better than the one for commercial feedstuff (93% for the extruded *pellets* during the first hour *versus* 78% for the commercial product), with lower losses of matter in the water column (0.79% *versus* 0.97% for the commercial product), and with a higher post-extrusion expansion (expansion index of 2.1). The extruded product complies with the commercial feedstuffs used as a control: Protein requirements if the extruded *pellets* had a final protein content of 24.5% (*versus* 25% for the commercial product)
- The reduction of the die orifice to 2 mm demonstrates that the possibilities to diversify the use of the extruders just by changing the die orifices diameters opens the possibility to produce feedstuffs for different species specimens at different stages of growth
- The especifications for the extrusion process found in this research show the technical feasibility to produce a balanced feedstuff based on *Plecostomus punctatus* dry meals.

Considering the experimental design carried out in this investigation the following recommendations for future tests in the Wenger X-5 extruders, and its implementation at real scale are presented:

- It is necessary to maintain the heating system of the extruder with a steam pressure of at least 588.40 kPa (6 kg_f/cm^2). If the extruder starts to receive steam to a lower pressure, for example, lower than 540 kPa (5.5 kg_f/cm^2) temperature at the extrusion zone will decrease and the products will be more dense losing its hydrostability (floatation). As both Wenger X-5 extruders were located in institutions where there was no control on this variable (i.e., increasing the boiler pressure), mechanical changes are crucial to obtain the desired stable extruded products

- Results and experience gained indicate that, for producing low density products a new low cost extruder operating at high pressure and high temperature conditions may be constructed using the same basic dimensions of the present one but changing its external features to warrant these processing conditions:

  1. Constructing new dies with different orifice diameters (for example, 0.55 mm diameter orifices, two, three, or four of them). This feature will increase pressure within the extrusion zone of the extruder, favoring the expansion of the material and reducing its density
  2. To preserve the pressure within the extruder it is highly convenient to have a lateral feeding hopper that be kept half full with the feeding mixture during the extrusion process to avoid air entrance and the possible leakage of evaporated water through the feeding sector of the extruder
  3. To maintain the temperature control the installation of thermowells for two additional pyrometers will allow the follow-up of the temperature, both in the transition and the extrusion zones within the extruder, controlling them with a thermostat the electric resistances installed

- It is important to consider the technical and economical feasibility for filleting *Plecostomus punctatus*. and that the ratio of fillets is low, although due to its protein content it might substitute the imported Norway fillets for Christmas, a traditional dish. One of the findings of this research, in its first stage, was about the quality of the meals produced with the whole fish since it has a good protein content to produce extruded feedstuffs for higher commercial value fish species such as tilapia. Considering the economical analysis, the extruded product obtained will be excellent to feed tilapia (Oreochromis spp.) in a more economical way reducing the costs for balanced feedstuffs. The final product of an aquacultural farm in the dam "El Infiernillo" would be the cultivation of a fish with a much higher ratio of fillet to whole fish body, that has an established consumption market as it used to be before the *Plecostomus punctatus* invasion
- Since the meal of *Plecostomus punctatus* is the main raw material for this process, it is very important to scale up the process looking for the optimization of the drying stage. It is recommended to consider the climate of the area, a dry tropical area, that may allow the use of aerothermal dryers, considering the patent for a solar-LPG system (Espinosa-Aquino et al., 2011)
- The communities living near the dam basin of "El Infiernillo" are not producing wheat, nor soybeans, and as these two ingredients are the fundamental elements of the diet it is necessary to study an alternative formulation considering byproducts of the region. Once the new *pellets* with the chemical and physical desirable characteristics, both with the control diet as well as with *Plecostomus punctatus* whole meal, containing carbohydrate sources of the area such as sorghum (*Sorghum bicolor*) they can be produced and *in vivo* tested in a cooperating *aquafarm*. This new phase will allow the correct cost calculations as well as the real effects of the diet
- Other improvement points are the grinding and screening operations, particularly if sorghum is to be added to the feedstuffs, minimizing production costs
- Since Mexican technicians are quite creative, they might construct a prototype low cost extruder following the abovementioned suggestions for the use of dies with orifice diameters of about 2mm, obtaining a better expansion y quite good *pellets* physical characteristics.

The substitution of any fish meal by the *Plecostomus punctatus* whole meal will bring the following benefits for the study area in the state of Michoacán, Mexico (Anonymous, 1998; Juárez, 1989; Martínez, 2005):

- An economical benefit from the open capture of *Plecostomus punctatus*, a species considered as a pest

- The increase of tilapia cultivation changing the open systems for fish farms using contained systems within the dam avoiding the depredation of *Plecostomus punctatus*
- Reduction of pollution of the dam bordering land areas where dead *Plecostomus punctatus* are dumped and its concomitant water pollution when it rains and run off send these carcasses to the dam "El Infiernillo"
- Economical reactivation for the communities dependent on the fishing activities after their cultivation lands were flooded with the dam water introducing a new economical activity, the production of extruded feedstuffs, particularly for tilapia (Oreochromis spp.) but also to sell to other fish farms
- Improvement of the living conditions of the communities, not only from the economical point of view but also from the environmental one, since what presently is a waste it may become a raw material for extruded pellets used as high value fish feedstuffs.

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