Aquaculture 283 (2008) 116–122

Contents lists available at ScienceDirect

Aquaculture

journal homepage: www.elsevier.com/locate/aqua-online

Enhancing the nutritional value of pea seed meals (*Pisum sativum*) by thermal treatment or specific isogenic selection with comparison to soybean meal for African catfish, *Clarias gariepinus*

S.J. Davies [a], A. Gouveia [b,*]

[a] *Fish Nutrition and Aquaculture Health Unit, School of Biological Sciences, Faculty of Science, University of Plymouth, Portland Square, PL4 8AA, Plymouth, UK*
[b] *Departamento de Zoologia e Antropologia, Faculdade de Ciências do Porto, Universidade do Porto, 4169-007 Porto and CIMAR/CIIMAR – Centro Interdisciplinar de Investigação Marinha e Ambiental, 4050-123, Porto, Portugal*

ARTICLE INFO

Article history:
Received 26 March 2008
Received in revised form 18 June 2008
Accepted 19 June 2008

Keywords:
African catfish
Clarias gariepinus
Pea seed meals
Pisum sativum
Isogenic selection
Thermal treatment
Growth and feed utilization

ABSTRACT

A 42 day preliminary feeding trial was conducted using African catfish (initial mean weight 52 g), to assess the effects of thermal treatment on a defined (rr RbRb) pea seed meal (*Pisum sativum*) by dry and moist heat treatments to reduce inherent trypsin inhibitor activity (TI): dry heated at 180 °C: 30 min (D180/20 diet), dry heated at 180 °C: 30 min (D180/30 diet), dry heated at 210 °C: 10 min (D210/10 diet) and moist heated by autoclaving at 150 °C, 15 psi: 15 min (M150/15 diet). Dry thermal treatment yielded the lowest TI activity levels within the pea seed meals (from 12.92 in untreated to 1.25 TIU mg^{-1} DM) whilst the moist autoclaved method only reduced it to 8.20 TIU mg^{-1} DM.

African catfish fed diets with dry thermally treated pea seed meal had significantly increased weight gain (2.05 g $fish^{-1}$ day^{-1}) when compared to those fed raw or untreated pea seed meal (1.69 g $fish^{-1}$ day^{-1}). However, although moist heat treatment produced superior weight gain than catfish fed raw pea seed meal (1.78 g $fish^{-1}$ day^{-1}), this was less effective than dry thermal treatments. These trends were also supported by feed efficiency ratio (FER) values that ranged from 0.85 to 0.74, respectively, for the reference diet (untreated pea) and the pea source subjected to dry heat treatment (150 °C: 15 min). Protein efficiency ratio (PER) was also significantly improved for catfish fed pea treatments processed at higher temperature and duration.

In a separate 56 day trial with sub-adult African catfish (initial mean weight 55 g) a pea seed meal derived from a specific isogenic pea line (RR RbRb) developed for low TI (0.47 TIU mg^{-1} DM) content together with a commercial pea protein concentrate (Lysamine®) of relatively low TI activity level (1.90 TIU mg^{-1} DM) was tested at the expense of soybean meal. Also, graded inclusions of soybean meal were also evaluated (with and without) lysine and methionine supplementation at the highest dietary inclusion level. This trial confirmed that 30% dietary inclusion of both products was feasible in promoting effective feed utilization performance for catfish with pea seed meals outperforming catfish fed diets with higher soybean meal levels. Daily weight gain for catfish were 2.74 and 2.9 g $fish^{-1}$ day^{-1} for the pea protein concentrate and isogenic pea seed meal, respectively, compared to 2.41 g $fish^{-1}day^{-1}$ for the reference (high fish meal diet) and 2.3 g $fish^{-1}$ day^{-1} for all higher soybean meal fed catfish.

1. Introduction

It is increasingly accepted that fish meal constitutes 30–60% of the total cost of feeds for aquaculture and more sustainable protein sources are essential to meet the growing demand and escalating costs of fish feeds (Southgate, 2003). This is of particular concern to tropical countries which are dependent on fish meal imports such as the African continent and Southeast Asia with fish farming operations based on carp, tilapia and catfish species which contribute high value protein for human consumption in these regions (Robinson and Li, 2002; Adewumi, 2006; Goda et al., 2007; Nyina-wamwiza et al., 2007).

Less expensive plant protein feedstuffs such as soybean meal, lupins, pulses and grain by-products have been widely explored and many offer promising applications in practical diet formulations to effect the partial or even substantial replacement of fish meal in several fish species (El-Said and Gaber, 2003; Peres et al., 2003; Glencross et al., 2004, 2005; Evans et al., 2005; Sitjà-Bodadilla et al., 2005; Borgeson et al., 2006; Tibbetts et al., 2006; Gatlin et al., 2007). The main constraints to their use is principally due to problems encountered with their amino acid profile, digestibility and reduced palatability but more importantly the presence of various anti-nutritional factors (ANF's) such as protease inhibitors, alkaloids,

* Corresponding author. Tel./fax: +351 22 0402807.
E-mail address: agouveia@fc.up.pt (A. Gouveia).

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doi:10.1016/j.aquaculture.2008.06.022

Table 1
Trypsin inhibitor activity of each pea seed meal, isogenic pea, pea protein concentrate and solvent extracted soybean meal

Meals	Trypsin inhibitor activity (TIU mg^{-1} DM)
Untreated pea seed mealik[a]	12.92
Pea seed meal (180 °C 10 min^{-1}) [a]	5.99
Pea seed meal (180 °C 20 min^{-1}) [a]	6.02
Pea seed meal (180 °C 30 min^{-1}) [a]	1.25
Pea seed meal (210 °C 10 min^{-1}) [a]	1.48
Pea seed meal autoclave (150 °C 15 min^{-1}) [a]	8.20
Isogenic pea[a]	0.47
Pea protein concentrate (Lysamine® GP Pea Protein)[b]	1.90
Solvent extracted soybean meal[c]	2.75

[a] Union Nationale Interfrofisssionelle des Plantes Riches en Proteines (UNIP), Paris, France.
[b] Roquette Frères, Lestrem, France.
[c] Cherwell Valley Silos Ltd., Banbury, Oxfordshire, UK.

tannins, lectins and adverse oligosaccharides as well as dietary fibre (Vilhelmsson et al., 2004; Drew et al., 2007; Gatlin et al., 2007; Schulz et al., 2007). This is especially acute in unprocessed soybean meals where trypsin inhibitors are often present and interfere with the digestive physiology of the animal causing a reduction in growth and feed utilization efficiency (Alexis and Nengas, 2001; Drew et al., 2007).

Field pea (*Pisum sativum*) is the type of legume most widely used for human consumption. Furthermore it is widely cultivated around the world and in fact, it is fourth in world production of grain legumes (Ratnayake et al., 2002; Drew et al., 2007). The interest in its use as a feed ingredient in aquaculture has increased in recent years (Tibaldi et al., 2005; Drew et al., 2007; Gatlin et al., 2007; Schulz et al., 2007). Pea seed meal has been shown to be an acceptable component in practical diets for European sea bass by Gouveia and Davies (1998, 2000) with dietary inclusions of up to 30–40%.

The trypsin inhibitor activity (TI) level in field peas is generally low, typically one tenth of the level found in soybean seeds (Burel et al., 2000; Thiessen et al., 2003b; Allan and Booth, 2004; Schulz et al., 2007). For these reasons, studies to evaluate the nutritional value of peas for aquaculture are gaining interest.

Pea seed meal and pea protein concentrates have been tested for a number of freshwater species such as Nile tilapia (Fontaínhas-Fernandes et al., 1999; Borgeson et al., 2006; Schulz et al., 2007) and channel catfish (Peres et al., 2003) with varying success. More recently, advances in genetic breeding techniques have resulted in new isogenic lines of peas selected for different traits including low TI activity content. This has been developed in Europe for use in poultry and swine feeds.

The objectives of this study were to investigate the effect of different thermal treatment procedures of defined pea seed meal sources and to evaluate a genetically selected pea gene-line of reduced trypsin inhibition and a commercial pea protein concentrate in practical type diets for African catfish. Feeding trials were therefore undertaken to assess the performance of juvenile African catfish in terms of feed performance in a preliminary trial followed by a longer term production diet assessments.

2. Materials and methods

2.1. Trial I

Experimental diets were manufactured using a field pea (*P. sativum*), cultivar C443, Frilene line, with a nutrient composition as follows: crude protein 26.57%, crude lipid 2.17%, total starch 47.26% (amylose: amylopectin ratio 70:30), neutral detergent fibre (NDF) 10.83% and acid detergent fibre (ADF) 6.57% supplied by Union Nationale Interprofessionnelle des Plantes Riches en Proteines (UNIP) Paris, France.

The experimental diets differed only in the type and degree of thermally treated pea type employed. Table 1 shows the TI activity remaining after processing of the original source and an isogenic (low TI activity) pea line, a commercial pea protein concentrate and a standard solvent extracted soybean meal. Five of these heat treated materials and the untreated pea seed meal formed the substitute for fish meal in balanced diets for African catfish as described by Ali and Jauncey (2005). Diets were formulated to be *iso*-nitrogenous (46% crude protein) and *iso*-lipidic (11% crude lipid). In the reference diet, 57% by weight of Chilean brown fish meal (27% of dietary protein) and 33% by weight (15% of dietary protein) were replaced by Chilean brown fish meal and untreated pea seed meal respectively. The same proportion was applied for the 4 remaining diets in which brown fish

Table 2
Formulation and proximate analysis of the experimental diets of trial I (dry weight)

Ingredients (g kg^{-1})	Diets					
	International feed number	Reference	D180/20	D180/30	D210/10	M150/15
Chilean brown fish meal[a]	5-02-	570	570	570	570	570
Untreated pea seed meal[b]	5-08-481	330	–	–	–	–
Pea seed meal[b]	5-08-481	–	330	–	–	–
Pea seed meal[b]	5-08-481	–	–	330	–	–
Pea seed meal[b]	5-08-481	–	–	–	330	–
Pea seed meal[b]	5-08-481	–	–	–	–	330
Blood meal[c]	5-00-381	20	20	20	20	20
Cod liver oil[d]	7-01-994	50	50	50	50	50
Vitamin and mineral premixes[e]		30	30	30	30	30
Proximate analysis (% DM)						
Moisture		8.00	8.60	8.00	7.80	8.50
Crude protein		46.38	45.90	45.47	46.64	45.86
Crude lipid		10.79	10.51	11.85	12.81	11.8030
Ash		9.43	9.59	9.78	9.90	10.16
Nitrogen free extract[f]		25.40	25.40	24.90	22.85	23.70

[a] Callumpton, Exeter, UK.
[b] Union Nationale Interfrofisssionelle des Plantes Riches en Proteines (UNIP), Paris, France.
[c] Carne & Sons, Callington, Cornwall, UK.
[d] Seven Seas, Hull, Yorkshire, UK.
[e] Vitamin and mineral premix, Sigma Aldrich Company, Poole, Dorset, UK., providing the following per kg of dry feed: vitamin A, 1600 IU; vitamin D, 2400 IU; vitamin E, 160.0 mg; vitamin K, 16.0 mg; thiamin, 50.0 mg; riboflavin, 52.0 mg; pyridoxine, 40.0 mg; niacin, 200.0 mg; pantothenic acid, 100.0 mg; folic acid, 16.7 mg; biotin, 300.0 mg; cyannocobalamin, 48.0 µg; ascorbic acid, 720.0 mg; choline chloride, 2000.0 mg; Ca pentothenate, 12000 mg; Mg sulphate, 48450 mg; NaCl, 2280 mg; K chloride, 1900 mg; Fe sulphate, 950 mg; Zn sulphate, 209 mg; Mn sulphate, 96.0 mg; Cu sulphate, 298 mg; Cb sulphate, 18.1 mg; Ca iodide, 11.2 mg; chromic chloride, 48 mg; Na selenite, 2.5 mg.
[f] Nitrogen free-extract. calculated as: 100−(moisture+crude protein+crude lipid+ash).