INNOECOFOOD - Eco-innovative technologies for improved nutrition, sustainable production and marketing of agroecological food products in Africa

Grant agreement no: 101136739

Deliverable 3.1 - Innovative, sustainable fish feed from fish waste, spirulina and insects

Due date of deliverable: 30/06/2025
Actual submission date: 27/10/2025

Start date of the project: 01/01/2024 **Duration: 36 months**

Organisation name of lead contractor: CIIMAR - Centro Interdisciplinar de Investigação Marinha e Ambiental
Revision: V1

Project co-funded by the European Commission within the Horizon Europe Programme	
Dissemination Level	
PU Public	X
SEN Sensitive, only for members of the consortium (including the Commission Services)	

Table of Contents

Grant agreement: 101136739

Summary

Aquaculture is a rapidly growing industry that provides sustainable source of protein while supporting job creation, economic stability, and equal opportunities across communities. The expansion of aquaculture intends to meet the ever growing fish demands, while wild fish stocks nearing their ecological limits. Meanwhile, aquaculture must reduce reliance in fishmeal and fish oil for production of the aquaculture feed. Therefore, development of alternative protein sources in fish feed is crucial to ensure the sustainable development of the sector. The INNOECOFOOD project through deliverable 3.1 aims to strengthen fish farms by evaluating protein sources produced locally within ECOHUBS, including spirulina, black soldier fly (*Hermetia illucens*) larvae meal (BSFLM) and fish wastes as alternatives to fishmeal. To begin with, and due to delay in the establishment of the ECOHUBS, parallel aquafeed optimization trials have been conducted at CIIMAR (for tilapia) and one at KMFRI (for catfish). The optimized feeds will be validated at the ECOHUBS when the constructions are completed.

Objective

The objective is to ensure the production of sustainable feeds for catfish and tilapia using fish wastes, spirulina, and insects with less reliance on fishmeal as a protein source.

Methodology

The methodology involved the following steps:
(i) Ingredient screening: Commonly used ingredients including fish by-products, insects, and spirulina, as well as locally available plants, including agricultural waste, cereal bran, cassava, millet, and maize produced in the ECOHUBS and living labs was assessed using literature review (Annex I, Tables 1 - 3).

(ii) Fish feed formulation: Balanced diets with suitable protein, lipid, and mineral contents, carbohydrate were formulated with the selected ingredients based on ingredient screening above (Annex I).
Nile tilapia: Five practical diets were formulated by CIIMAR, TAFIRI and CLAR (35% crude protein and 8% crude lipids). A control diet contained 5% fishmeal (FM), while for the other experimental diets, FM-protein was partially (25%) replaced by either BSFLM, or spirulina, or totally (100%) by a combination of 75% BSF and 25% spirulina, or 100% BSF (Annex II, Table 4).

Catfish: KMFRI in Kenya, in collaboration with CIIMAR, has developed diet formulation used for catfish feed trial using four practical diets (32% crude protein and 8% crude lipids). A control diet contained 10% fishmeal (FM), while for the other experimental diets, FM-protein was partially (50%) replaced by BSFLM+corn meal+DCP (dicalcium phosphate), or spirulina+corn meal+DCP, or totally (100%) by a combination of 41% BSF + 30% spirulina + 19% corn meal + 10% DCP (Annex III, Table 5).

Grant agreement: 101136739

(iii) Fish feeds tests: One set of trials was undertaken for Nile tilapia (at CIIMAR) and one for catfish (at KMFRI) with the following conditions:

Nile tilapia: Fish feed (Annex IV, Figure 1) trials in triplicates were undertaken using juveniles (initial body weight: 8.3 g) as test fish based on the formulations above against the commercial feed control. Fish were fed during 80 days using a RAS system. Fish were stocked at 20 fish/tank maintained at 26-27°C and photoperiod 12L:12D. Different parameters were screened, namely feed conversion factor, feed crude protein ratio, growth, weight, survival, body composition and fish welfare.

Catfish: A 126 days fish feed (Annex IV, Figure 2) trials involves three replicates is being conducted using juvenile catfish as test fish. Fish was stocked at 100 fish per tank (average 10 g)/TK (250 L). Tanks and system (Annex V, Figure 3) are being cleaned daily, and the water level fixed throughout the whole experimental period. Different parameters are being assessed (Annex V, Figures 4 – 5) including feed conversion factor, feed crude protein ratio, growth, weight, survival and body composition.

Results and Discussion

Tilapia: At the end of the trial, growth performance and feed utilization showed no differences among feeds (Annex VI, Figure 6). Final body weight averaged 88.7 g, daily growth index 3.43, and feed conversion ratio 1.14. Diet's nutrient digestibility, retention, and gain remained unaffected by the experimental diets. Moreover, the histomorphological evaluation revealed no significant effects on the intestinal integrity of fish fed these diets. Regarding the nutritional value of tilapia fillets, it remained similar among treatments (similar total lipids and EPA + DHA levels), but diets containing BSF showed potential to improve fillets' stability and shelf life, through reduced lipid peroxidation and greater antioxidant capacity (Annex VI, Figure 7).
Under normal growing conditions, spirulina-containing diets appeared to influence nutrient metabolism and enhance the immune status of fish. However, under acute cold stress (15 °C), the observed benefits under normal conditions seem to be lost, where BSF-based diets were more effective in supporting immune function (indicated by the increased total protein and globulin levels) (Annex VI, Figure 8). Finally, liver oxidative stress status was not affected by the inclusion of BSF and/or spirulina compared to the fishmeal-based diet.
Overall, the present results demonstrate the feasibility of replacing FM with spirulina and BSF as innovative feed sources in diets for Nile tilapia, without compromising growth, feed efficiency, or digestive health, and fish wellbeing. BSF inclusion may also improve the oxidative stability and extend the shelf life of tilapia fillets. Furthermore, while spirulina appears to enhance immune function under optimal conditions, BSF-based diets seem to offer greater benefits under stress conditions, such as cold-water temperatures. Nevertheless, given the lack of significant differences in zootechnical performance, an economic analysis could be useful in selecting the optimal diet. These data will help select the best feed (ideal density, floatability, water absorption and particle size, and economic analysis) that will be produced locally at the ECOHUBS in solar driers.

African Catfish: The catfish trial at KMFRI started in April 2025 and was finalized on 7th August, i.e., at day 126. The growth performance and feed utilization showed significant differences among feeds (final body weight was significantly higher in the fish fed on the spirulina at 50% (SPIR_50), followed by the BSF and spirulina at 100% (BSF_SPIR 100). The BSF_50 had the lowest weight but was not significantly different that the control. The daily growth rate was significantly higher in the SPIR_50 and BSF_SPIR 100, but without significant differences found between them. Feed conversion ratio was also statistically higher in fish fed BSF_50 (0.86 ± 0.05) followed by the control. The feed conversion ratio for all diets was below 1. Overall, the present results demonstrate the feasibility of replacing FM with spirulina meal as innovative feed sources in diets for African catfish. The African catfish utilized spirulina and spirulina mixed with BSF in a similar way of the control diet. Spirulina meal is therefore recommended for replacing the fish meal in African catfish diets (Annex VII, Table 6) when taking into account the zootechnical perspective.

The haematological analysis did not show significant differences among the treatments though the red blood cell counts, haemoglobin and lymphocyte counts were higher in the fish fed on spirulina (Annex VII, Table 7).

Grant agreement: 101136739

Annexes

Annex I – Fish feed screening from three ECOHUBS (Tanzania, Kenya and Egypt)

Table 1. Fish feed ingredient screening from Tanzania ECOHUB.

Type	Feeed ingredient	%CP	%CF	%EE	NFE	%Ash	Country
Animal by produc	Lake victoria sard	58.0-65.0	0-0.3	7.6-16.1	2.2-8.9	6.5-22.2	Tanzania
	Fresh water shrin	28.0-47.0	2.3-3.0	1.8-9.6	1.4	5.0-5.8	Tanzania
	Cock shrimp (mar	74.8	1.9	2.1	2.8	18.4	Tanzania
	Nile perch fish fra	35.8	0.7	15.3	1.1	47	Tanzania
	Prawn head wast	51.1	12.9	8	2.2	25.8	Tanzania
	Housefly maggot	48.1	1	24.9	19.2	6.9	Tanzania
	Earthworm	65.8	3.7	13.4	6.8	10.3	Tanzania
							Tanzania
Agricultural by-pr	Full fat soybean	43.0-47.0	2.0-6.4	17.0-17.9	27.3	5.6	Tanzania
	Rice bran	6.3-9.3	21.4	3.0-9.4	38.3	20.0-22.2	Tanzania
	Maize bran	13.1	5.8	17.5	57.6	6	Tanzania
	Soy bean	43.5	8.9	15.6	26.6	5.5	Tanzania
	Cotton seed cake	30.3-357	17.6-20.8	17.0-20.8	20.9-34.9	6.9-7.2	Tanzania
	Palm seed cake	15.2	13.6	14.9	52.8	3.5	Tanzania
	Coconut waste	14.8	21.3	48.7	9.2	6.1	Tanzania
	Sunflower seed c	26.8	24.4	11.9	15.8	21.1	Tanzania
							Tanzania
Plant leaves and v	Moringa leaf	29.8	5.2	5.2	47.8	11.9	Tanzania
	Taro leaf	29.2	17.4	3.2	30.1	17.4	Tanzania
	Lettuce vegetable	23.5	14.6	1.3	30.4	30.3	Tanzania
	Wandering Jew	18.6	17.4	0.5	38.8	24.7	Tanzania
	Sweet potato lea	22.7	13.3	4.7	45	14.4	Tanzania
							Tanzania
Aquatic plants	Azolla	23.5-23.8	11.0-12.8	1.6-2.0	39.8	11.6-22.3	Tanzania
	Duck weed	19.8	11.8	2.7	40	20	Tanzania
	Water lettuce	22.8	13.3	2.9	45.6	20.9	Tanzania
							Tanzania
Industrial by-prod	Spent brewer's gr	25.3	17.4	8.5	44.7	4.1	Tanzania
	Spent brewer's ye	35	0	0.5	61	3.5	Tanzania
	Local brewery wa	19.4	14.2	4.1	57.9	4.3	Tanzania
							Tanzania
Algae/ Single cell	Spirulina	50-65	9.8-11.4	6.6-6.8		9.5-9.9	Tanzania
	Chlorella	44-57		17-23	15-22		Tanzania
							Tanzania

Grant agreement: 101136739

Table 2. Fish feed ingredient screening from Kenya ECOHUB.

Type	Feeed ingredient	CP	CF	EE	NFE	Ash	Moisture	Counntry
Animal by products	Lake victoria sardine	55.1	1.3	18.7	6.8	18.2		Kenya
	Fresh water shrimp	63.2	6.5	5.5	6.5	10.8	7.5	Kenya
	Cock shrimp (marine)	26.6	10.7	7.83	32.27	12.37	10.2	Kenya
	Nile perch fish frames							Kenya
	Prawn head waste							Kenya
	Housefly maggot							Kenya
	Earthworm							Kenya
	Prawn head waste							Kenya
Agricultural by-products	Full fat soybean	42.3	4.5	3.5		5.3		Kenya
	Rice bran	10.7	14.3	11.3	46.17	8.23	9.3	Kenya
	Maize bran	11.8	5.5	10.7	58.5	2.9	10.6	Kenya
	Soy bean	38.2	5.3	28.2	14.9	4.2	9.2	Kenya
	Cotton seed cake	38.8	24.9	10.7	19.2	6.3		Kenya
	Palm seed cake							Kenya
	Coconut waste							Kenya
	Sunflower seed cake	25.9	36.8	5.4	26.6	5.1		Kenya
	Wheat polland	16.3	18.6	5.1	36.64	13.62	9.7	Kenya
Plant leaves and weeds	Moringa leaf							Kenya
	Taro leaf							Kenya
	Lettuce vegetable							Kenya
	Wandering Jew							Kenya
	Sweet potato leaf							Kenya
Aquatic plants	Azolla							Kenya
	Duck weed	26.6		5.21		15.87		Kenya
	Water lettuce							Kenya
	Seaweed powder	5.1	5.22	1.78	38.9	27.1	21.88	Kenya
Industrial by-products	Spent brewer's grain							Kenya
	Spent brewer's yeast							Kenya
	Local brewery waste							Kenya
Algae/ Single cell protein	Spirulina	62.1		13.4		9	5.4	Kenya
	Chlorella							Kenya
Insects	Blacksoldier fly	50.8	9.1	19.4	4.7	6.9	8.4	Kenya
	Cricket							
Others								

Grant agreement: 101136739

Table 3. Fish feed ingredient screening from Egypt ECOHUB.

Type	Feeed ingredient	CP	CF	EE	NFE	Ash	Moisture	Country
Animal by products	Lake victoria sardine	60-65	0.26-1	8.07-9.2	1.6-4.34	20.2-22.5	0.03-8	Egypt
	Fresh water shrimp	39.9-46.8	1.25-8.5	3.9-6.1	2.97-19.4	22.5-44.7	0.03-9.7	Egypt
	Cock shrimp (marine)	60.5	2.09	7.09	7.92	22.4	0	Egypt
	Nile perch fish frames	35.2-39.5	0.73-1.5	3.5-15.1	0-3.03	45.5-46	0-10	Egypt
	Prawn head waste	52.3	9.2	4.6	0	29.3	4.6	Egypt
	Housefly maggot	50-50.4	5.7-13	10-18.9	3.7-7.3	10.1-13.3	7.6-10	Egypt
	Earthworm	57.9	2.4	9	6	14.9	9.8	Egypt
	Prawn head waste							Egypt
Agricultural by-products	Full fat soybean	35.1-39.9	5.18-8.81	17.7-23.8	14.8-26.2	4.63-5.8	6.3-13	Egypt
	Rice bran	12.7	11.6	13.7	41.4	11.6	9	Egypt
	Maize bran	10.2	7.8	4.3	64.5	4.1	9.1	Egypt
	Soy bean	44-48.6	3.2-7.3	1.2-1.5	27.7-29.5	5.5-6.5	12.0-13.0	Egypt
	Cotton seed cake	24.0-41.0	11.0-24.0	1.5-6	28.1-33.5	6.2-6-5	45572	Egypt
	Palm seed cake	15.9-18.7	16.93-20.1	1.4-11.8	40.8-49.6	3.9-5	5.3-9.1	Egypt
	Coconut waste	21-23.5	13-16.8	2.8-9.8	38.3-42.29	6.8-7	7.14-10.2	Egypt
	Sunflower seed cake	28.0-40.0	12.0-25.0	1.2-2	27.8-33	6-6.5	45603	Egypt
	Wheat polland	16.1-17.7	4.7-7	4-6.92	59.3-66.5	3.6-4.24	0-10	Egypt
Plant leaves and weeds	Moringa leaf	26.8	12.2	6.4	35	10.8	8.8	Egypt
	Taro leaf	22.3	17.6	4.5	31.6	13.7	10.3	Egypt
	Lettuce vegetable	-	-	-	-	-	-	Egypt
	Wandering Jew	-	-	-	-	-	-	Egypt
	Sweet potato leaf	13.2	19.9	2.8	40.8	11.8	11.5	Egypt

Annex II – Fish diet formulation used for Nile tilapia feed trial at CIIMAR

Table 4. Dietary formulations for Nile tilapia.

Ingredients, %	CTRL	BSF25	SPIR25	BSF75_SPIR25	BSF100
	%	%	%	%	%
Fishmeal 60 (by-products)	**5.0**	**3.8**	**3.8**		
Insect meal (BSF PROTIX)		**1.4**		**4.2**	**5.8**
Spirulina			**1.2**	**1.2**	
Shrimp shell meal (by-products)	2.0	2.0	2.0	2.0	2.0
Brewer's spent yeast	5.0	5.0	5.0	5.0	5.0
Corn gluten meal	5.4	5.4	5.4	5.4	5.4
Soybean meal 44	26.0	26.0	26.0	26.0	26.0
Soybean meal full fat	8.0	8.0	8.0	8.0	8.0
Sunflower meal 28	14.0	14.0	14.0	14.0	14.0
Wheat bran	11.0	11.0	11.0	11.0	11.0
Rice bran full fat	11.0	11.0	11.0	11.0	11.0
Corn meal	8.6	8.3	8.5	7.5	7.2
Vitamin and mineral premix	0.5	0.5	0.5	0.5	0.5
DCP (Dicalcium phosphate)	0.6	0.8	0.8	1.3	1.3
L-Lysine HCl 99%	0.2	0.2	0.2	0.3	0.3
DL-Methionine	0.2	0.2	0.2	0.2	0.2
Soybean oil - MIXTURE	1.1	1.1	1.1	1.1	1.1
Poultry fat - MIXTURE	1.4	1.4	1.5	1.4	1.3
Total	**100.0**	**100.0**	**100.0**	**100.0**	**100.0**

Composition	CTRL	BSF 25	SPIR 25	BSF75_SPIR25	BSF 100
Crude protein, % feed	33.2	33.2	33.2	33.2	33.2
Digestible protein, %	28.1	28.1	28.1	28.1	28.1
Crude fat, % feed	8.5	8.5	8.5	8.5	8.5
Fiber, % feed	8.5	8.6	8.5	8.9	9.1
Starch, % feed	16.5	16.3	16.4	15.8	15.6
Ash, % feed	6.8	6.7	6.7	6.6	6.7
Gross energy, MJ/kg feed	19.2	19.2	19.2	19.2	19.2
Digestible energy, MJ/kg feed	13.3	13.3	13.3	13.3	13.3
DP/DE	19.9	19.8	19.9	19.9	19.8

1) Commercial diets for Nile tilapia do not contain FM in Egypt but still have low levels in other countries, so we **propose 5% of fish meal from by-products in the CTRL diet**

2) **BSF25: 25% of the fish-meal-protein was replaced by black-soldier fly meal (BSF)**

3) **SPIR25: 25% of the fish-meal-protein was replaced by spirulina meal (BSF)**

4) **BSF75_SPIR25: total replacement of FM, but 75 % of the fish-meal-protein was replaced by black-soldier fly meal (BSF) and 25% was replaced by spirulina meal (BSF) - this is due to the high cost of spirulina**

5) **BSF100: total replacement of FM by black-soldier fly meal (BSF)**

Grant agreement: 101136739

Annex III – Fish diet formulation used for African catfish trial at KMFRI

Table 5. Dietary formulations for catfish.

Ingredients, %	CTRL	BSF 50	SPIR 50	BSF_SPIR_100
Fishmeal 60	10.000	5.000	5.000	
BSF Meal (defatted)		3.000		4.100
Spirulina			2.100	3.000
Shrimp shell meal	4.000	4.000	4.000	4.000
Baker's yeast	6.000	6.000	6.000	6.000
Soybean cake	30.000	30.000	30.000	30.000
Soybean meal (full fat) meal	20.000	20.000	20.000	20.000
Sunflower seed cake	12.000	12.000	12.000	12.000
Wheat bran	6.300	6.300	6.300	6.300
Rice bran	4.000	4.000	4.000	4.000
Corn meal (whole maize)	3.000	4.900	5.400	4.900
Vit & Min Premix PV01	0.500	0.500	0.500	0.500
DCP (Dicalcium phosphate)	1.200	1.500	1.500	2.200
L-Lysine HCl 99%	0.000	0.000	0.000	0.000
DL-Methionine	0.200	0.200	0.200	0.300
Soybean oil - MIXTURE	2.800	2.600	3.000	2.700
Total	**100.000**	**100.000**	**100.000**	**100.000**

Composition	CTRL	BSF 50	SPIR 50	BSF_SPIR_100
Moisture, % feed	6.0	6.0	6.0	6.0
Crude protein, % feed	33.2	32.2	32.2	32.2
Crude fat, % feed	8.0	8.0	8.0	8.0
Fiber, % feed	8.1	8.7	8.2	8.9
Starch, % feed	6.8	8.0	8.3	8.0
Ash, % feed	9.8	8.8	8.7	8.2
Gross energy, MJ/kg feed	18.8	18.9	18.9	19.0
Arg, % feed	2.1	2.0	2.0	2.0
His, % feed	0.7	0.7	0.7	0.7
Ile, % feed	1.3	1.2	1.2	1.3
Leu, % feed	2.2	2.2	2.2	2.2
Lys, % feed	1.7	1.6	1.6	1.5
Thr, % feed	1.2	1.1	1.1	1.1
Trp, % feed	0.4	0.4	0.4	0.4
Val, % feed	1.4	1.4	1.4	1.4
Met, % feed	0.7	0.7	0.7	0.7
Cys, % feed	0.4	0.4	0.4	0.4
Phe, % feed	1.4	1.4	1.4	1.4
Tyr, % feed	1.4	1.4	1.4	1.5
Asx, % feed	3.1	3.0	3.0	3.0
Glx, % feed	5.2	5.0	5.1	5.0
Ala, % feed	1.5	1.5	1.5	1.5
Gly, % feed	1.6	1.4	1.4	1.4
Pro, % feed	1.5	1.5	1.5	1.5
Ser, % feed	1.4	1.4	1.4	1.4
Tau, % feed	0.0	0.0	0.0	0.0
Met+Cys, % feed	1.1	1.1	1.1	1.2
Phe+Tyr, % feed	2.8	2.8	2.8	2.8
Total P, % feed	1.0	1.0	1.0	1.0
Ca, % feed	0.5	0.6	0.6	0.7
Na, % feed	0.1	0.1	0.1	0.1
Mg, % feed	0.0	0.0	0.0	0.0
K, % feed	0.1	0.1	0.1	0.2

Cu, mg/kg feed	4.7	4.7	4.7	4.7
Fe, mg/kg feed	13.6	13.6	26.1	31.4
I, mg/kg feed	0.3	0.3	0.3	0.3
Mn, mg/kg feed	10.1	10.1	10.9	11.2
Se, mg/kg feed	0.0	0.0	0.0	0.0
Zn, mg/kg feed	10.4	10.4	10.8	10.9
Vit A, IU/kg feed	9494.9	9494.9	9494.9	9494.9
Vit D3, IU/kg feed	949.5	949.5	949.5	949.5
Vit K3, mg/kg feed	11.9	11.9	11.9	11.9
Vit E, mg/kg feed	47.5	47.5	47.5	47.5
Vit B1, mg/kg feed	14.9	14.9	14.9	14.9
Vit B2, mg/kg feed	14.3	14.3	14.3	14.3
Vit B3, mg/kg feed	48.8	48.8	48.8	48.8
Vit B5, mg/kg feed	96.4	96.4	96.4	96.4
Vit B6, mg/kg feed	9.9	9.9	9.9	9.9
Vit B9, mg/kg feed	7.1	7.1	7.1	7.1
Vit B12, mg/kg feed	0.0	0.0	0.0	0.0
Vit C, mg/kg feed	474.7	474.7	474.7	474.7
Biotin, mg/kg feed	1.4	1.4	1.4	1.4
Choline, mg/kg feed	474.7	474.7	474.7	474.7
Inositol, mg/kg feed	237.4	237.4	237.4	237.4
Betaine, mg/kg feed	237.4	237.4	237.4	237.4
Astaxanthin,mg/kg feed	0.0	0.0	0.0	0.0
Canthaxanthin, mg/kg feed	0.0	0.0	0.0	0.0
Apo-ester, mg/kg feed	0.0	0.0	0.0	0.0
Beta-carotene, mg/kg feed	0.0	0.0	0.0	0.0
C14, % feed	0.0	0.0	0.0	0.0
C16, % feed	0.4	0.4	0.4	0.4
C18, % feed	0.1	0.1	0.1	0.1
C18:1n9, % feed	0.9	0.8	0.9	0.9
LNA (C18:2n6), % feed	1.7	1.6	1.8	1.6
ALA (C18:3n3), % feed	0.2	0.2	0.2	0.2
ARA, % feed	0.0	0.0	0.0	0.0
EPA, % feed	0.0	0.0	0.0	0.0
DHA, % feed	0.0	0.0	0.0	0.0
EPA+DHA, % feed	0.0	0.0	0.0	0.0

Annex IV – Developed fish diets used for Nile tilapia and African catfish feed trial

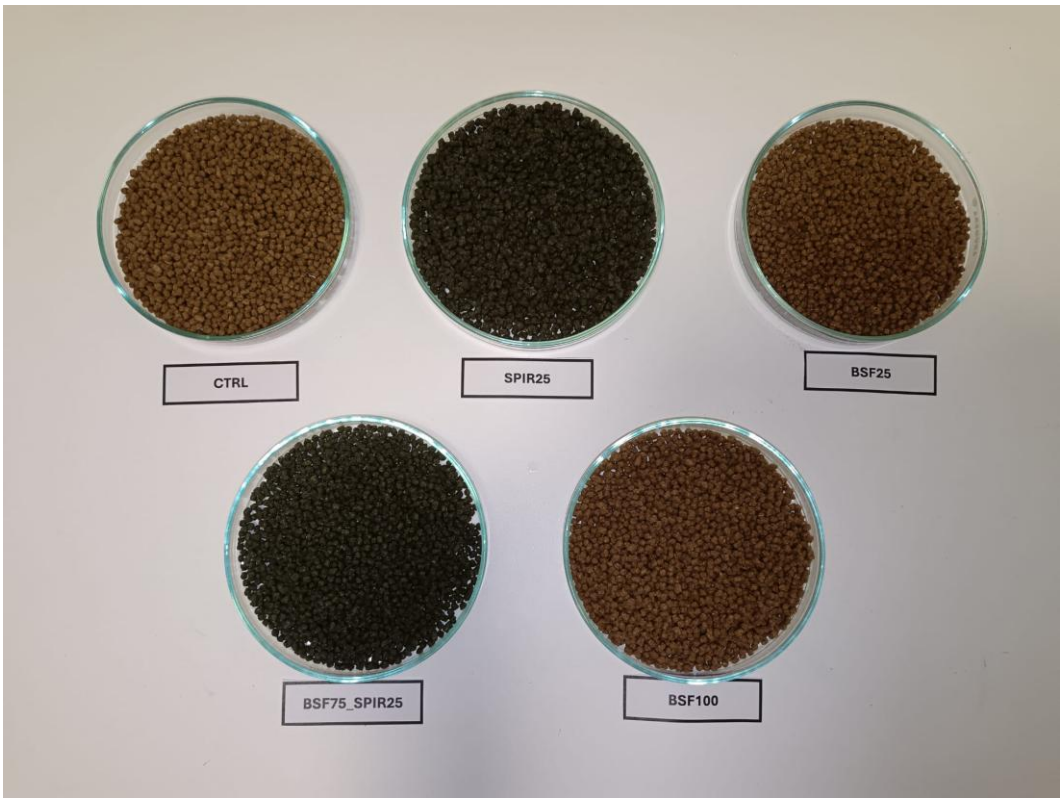


Figure 1. Photo evidence of the diets for tilapia feed trial at CIIMAR.

Figure 2. Photo evidence of catfish diets feed trial at KMFRI.

Annex V – Experimental system set up for African catfish feed trial at KMFRI

Figure 3. Photo evidence of the experimental system for African catfish trial at KMFRI.

Figure 4. Sampling of African Catfish at KMFRI sangoro.

Figure 5. Blood collection for hematological analysis from African catfish.

Figure 6. Growth performance and feed efficiency of Nile tilapia after 71 days of feeding with the experimental diets. Data are presented as means (n = 3) ± standard deviation. Absence of different superscript letters indicates no significant differences between treatments (p > 0.05). DGI: Daily growth index (100 x [final body weight1/3 – initial body weight1/3]/days of the experiment; FCR: Feed conversion ratio (dry feed intake/weight gain).

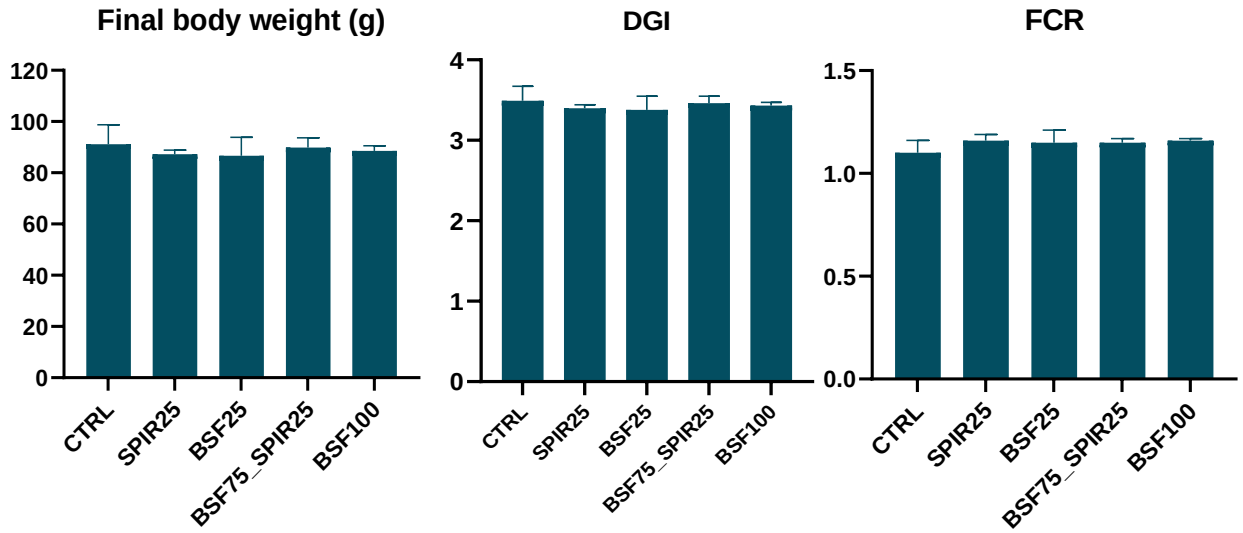


Figure 7. Muscle antioxidant capacity of Nile tilapia after 71 days of feeding with the experimental diets.
LPO: Lipid peroxidation (TBARS nnom/g); ORAC: Oxygen Radical Absorbance Capacity (µM of Trolox equivalent/g). Data are expressed as means (n = 15) ± standard deviation. Different superscript letters indicate statistically significant differences between treatments (p < 0.05).

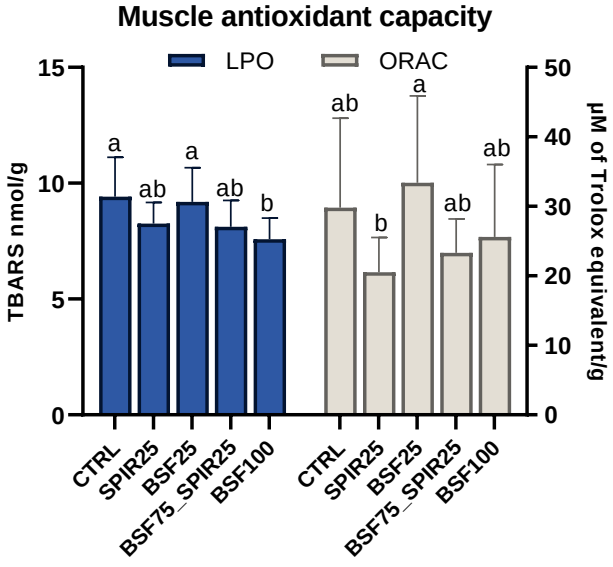
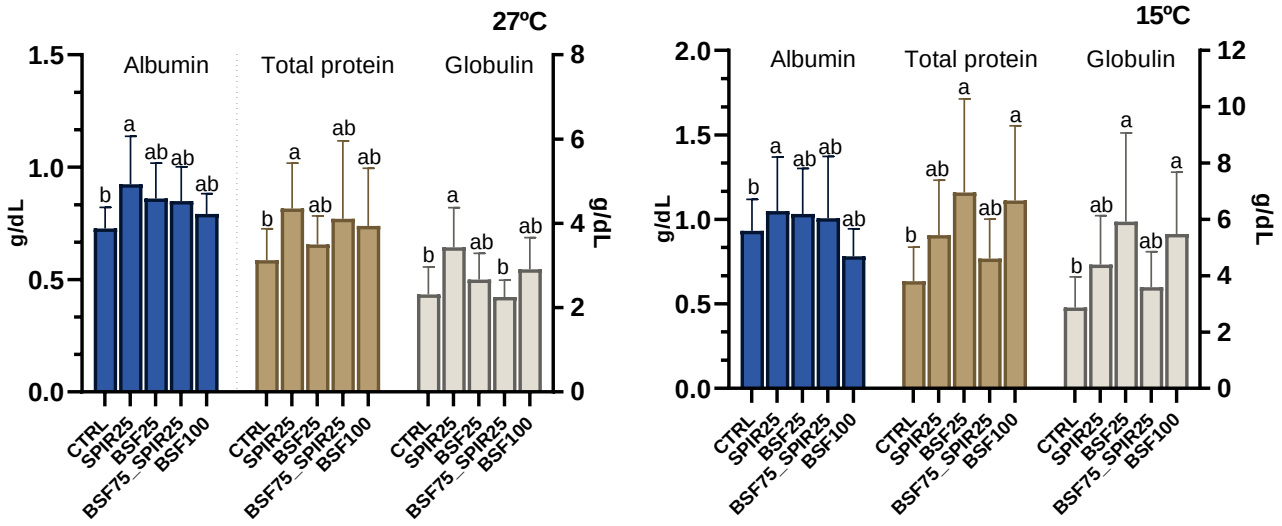


Figure 8. Plasma metabolites profile of Nile tilapia after 71 days of feeding the experimental diets (27 ºC) and after cold stress challenge (15ºC). Data are expressed as means (n = 15) ± standard deviation. Different superscript letters indicate statistically significant differences between treatments ($p < 0.05$).



Grant agreement: 101136739

Table 6. Results of the feed trial on African catfish. Abbreviations: initial weight (IW), initial length (IL), final weight (FW), final length (FL), feed conversion rate (FCR), specific growth rate (SGR), weight gain (WG), dry weight gain (DWG).

Parameter	Diet				
	Control	**SPIR_50**	**BSF_50**	**BSF_SPIR 100**	**P value**
IW	2.83 ±0.11[a]	2.76 ± 0.09[a]	2.83 ± 0.11[a]	2.88 ± 0.11[a]	0.888
IL	7.10 ± 0.1a	7.13 ± 0.09[a]	7.06 ± 0.11[a]	7.05 ± 0.11[a]	0.943
FW	42.08 ± 2.29[a]	61.17 ± 6.95[b]	33.64 ± 4.35[a]	60.36 ± 0.51[b]	<0.001
FL	17.84 ± 0.34[b]	18.43 ± 0.49[b]	15.93 ± 0.49[a]	19.33 ± 0.51[b]	<0.001
FCR	0.61 ± 0.03[b]	0.44 ± 0.03b[a]	0.86 ± 0.05[c]	0.42 ± 0.03[a]	<0.001
SGR%	2.09 ± 0.04[b]	2.22 ± 0.07[b]	1.81 ± 0.06[a]	2.24 ± 0.06[b]	<0.001
WG	39.25 ± 2.28[a]	58.40 ± 6.96[b]	30.80 ± 4.35a	57.48 ± 6.45[b]	0.001
DWG	0.31 ± 0.02[a]	0.46 ± 0.05[b]	0.24 ± 0.03[a]	0.45 ± 0.05[b]	<0.001
WG%	1522.02 ± 97.43[a]	2381.82 ± 29.23[b]	1255.12 ± 85.36[a]	2142.75 ± 28.73[ab]	<0.001

Table 7. Hematological analysis.

Parameter	**Diet**					
	Control	**SPIR_50**	**BSF_50**	**BSF_SPIR 100**	**F-value**	**P-value**
White Blood cell count (10^3/uL)	30.72 ± 2.20	25.69± 3.63	21.59± 3.96	27.15± 3.42	1.29	0.28
Lymphocyte (%)	91.87± 0.85	90.17± 2.10	90.60± 1.19	90.04± 1.96	0.29	0.83
Mid-sized cell percentage (%)	4.77± 0.42	4.22± 0.34	4.21± 0.33	4.36± 0.25	0.57	0.63
Granulocyte (%)	3.37± 0.45	5.61± 1.88	5.19± 1.13	5.59± 1.82	0.62	0.61
Lymphocyte count (10^3/uL)	28.10± 1.95	23.84± 3.37	19.91± 3.71	25.13± 3.23	1.20	0.32
Granulocyte count (10^3/uL)	1.08± 0.20	0.72± 0.12	0.70± 0.12	0.80± 0.09	1.49	0.23
Red Blood Cell count (10^6/uL)	2.47± 1.83	2.28± 0.18	1.83± 0.26	2.01± 0.23	2.19	0.10
Hemoglobin (g/dL)	12.09± 0.21	10.41± 1.00	9.31± 1,26	9.56± 1.11	1.85	0.15
Hematocrit (%)	24.48± 0.92	22.43± 1.87	18.03± 2.60	19.40± 2.18	2.41	0.07
Mean Corpuscular Volume (fL)	99.03± 1,79	98.24± 1.37	99.41± 2.15	97.19± 1.50	0.33	0.81
Platelet count (103/uL)	54.57± 6.62	82.94± 17.41	70.46± 14.29	50.50± 5.87	1.72	0.17

Grant agreement: 101136739