ALMA MATER STUDIORUM
UNIVERSITÀ DI BOLOGNA

Effetti di mangimi circolari sulla modulazione del microbiota intestinale nell'orata e nella spigola

L. Parma[1]*, D. Scicchitano[2], M. Candela[2], G. Palladino[2], E. Nanetti[2], N. Cinti[2], S. Busti[1], A. Marchi[1], P. P. Gatta[1], A. Bonaldo[1]

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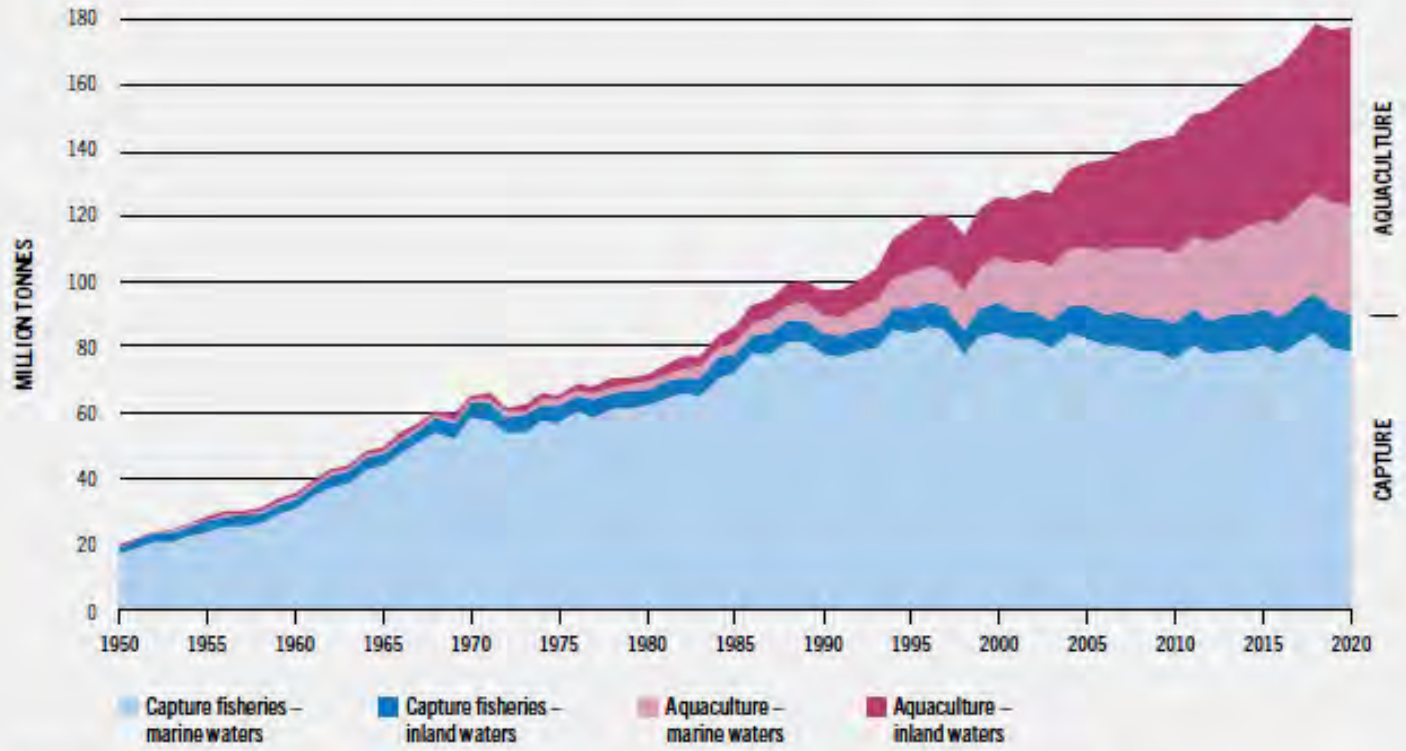
Background –Aquaculture Production



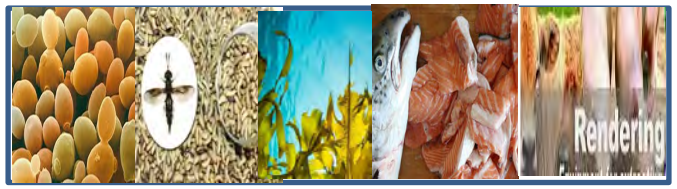
NOTES: Excluding aquatic mammals, crocodiles, alligators, caimans and algae. Data expressed in live weight equivalent.
SOURCE: FAO.

Background - Circular aquafeed ingredients

Aquafeed 1.0

Aquafeed 2.0

Aquafeed 3.0



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Aquaculture 230 (2004) 391–404

Aquaculture

www.elsevier.com/locate/aqua-online

Almost total replacement of fish meal by plant protein sources in the diet of a marine teleost, the European seabass, *Dicentrarchus labrax*

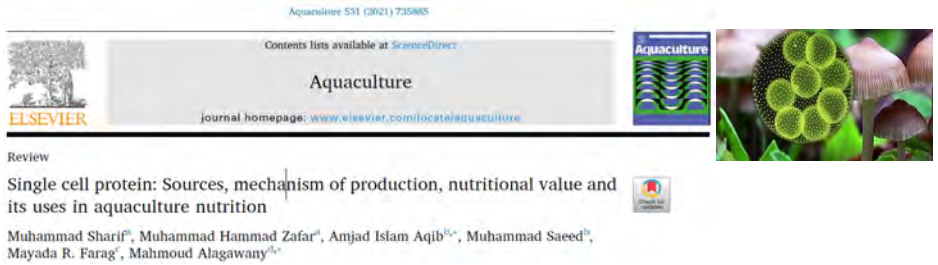
S.J. Kaushik[a,*], D. Covès[b], G. Dutto[b], D. Blanc[a]

Towards achieving circularity and sustainability in feeds for farmed blue foods

Stefanie M. Colombo[1] | Koushik Roy[2] | Jan Mraz[2] | Alex H. L. Wan[3]
Simon J. Davies[3] | Sean M. Tibbetts[4] | Margareth Øverland[5] |
David S. Francis[6] | Melissa M. Rocker[6] | Laura Gasco[7] | Emma Spencer[8] |
Marc Metian[9] | Jesse T. Trushenski[10,11] | Giovanni M. Turchini[12]

Background - Circular aquafeed ingredients

Single cell proteins (SCP)


Aquaculture 531 (2021) 735865

Contents lists available at ScienceDirect

Aquaculture

journal homepage: www.elsevier.com/locate/aquaculture

Review

Single cell protein: Sources, mechanism of production, nutritional value and its uses in aquaculture nutrition

Muhammad Sharif, Muhammad Hammad Zafar, Amjad Islam Aqib, Muhammad Saeed, Mayada R. Farag, Mahmoud Alagawany

- Composition (high protein content 50-80%, dm)
- Limited need for arable land and water
- Year-round production at a fast rate, unaffected by seasonal or climatic variations

β-glucan, nucleic acids, nucleotides, Vit B

Insects meal

REVIEWS IN Aquaculture

REVIEW

Systematic review and meta-analysis of production performance of aquaculture species fed dietary insect meals

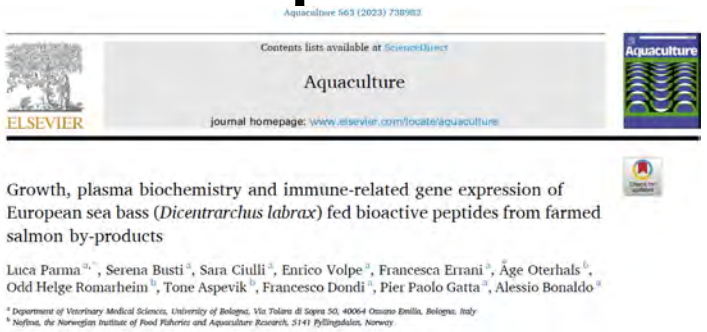
Hung Quang Tran, Tram Thi Nguyen, Markéta Prokešová, Tatyana Gebauer, Hien Van Doan, Vlastimil Stejskal

First published: 18 February 2022 | https://doi.org/10.1111/raq.12666 | Citations: 19

- Composition (high protein 38-68 % dm)
- Balanced aa profile
- Organic agricultural waste valorisation

Chitin, lauric acid

By-product from fisheries and aquaculture


Aquaculture 563 (2023) 739982

Contents lists available at ScienceDirect

Aquaculture

journal homepage: www.elsevier.com/locate/aquaculture

Growth, plasma biochemistry and immune-related gene expression of European sea bass (*Dicentrarchus labrax*) fed bioactive peptides from farmed salmon by-products

Luca Parma, Serena Busti, Sara Ciulli, Enrico Volpe, Francesca Errani, Åge Oterhals, Odd Helge Romarheim, Tone Aspevik, Francesco Dondi, Pier Paolo Gatta, Alessio Bonaldo

- High protein content
- High availability
- Reduced C foot print

Bioactive peptides

Functional compounds

Background – Fish microbiome

frontiers | Frontiers in Microbiology

TYPE Review
PUBLISHED 23 June 2023
DOI 10.3389/fmicb.2023.1200997

Check for updates

OPEN ACCESS

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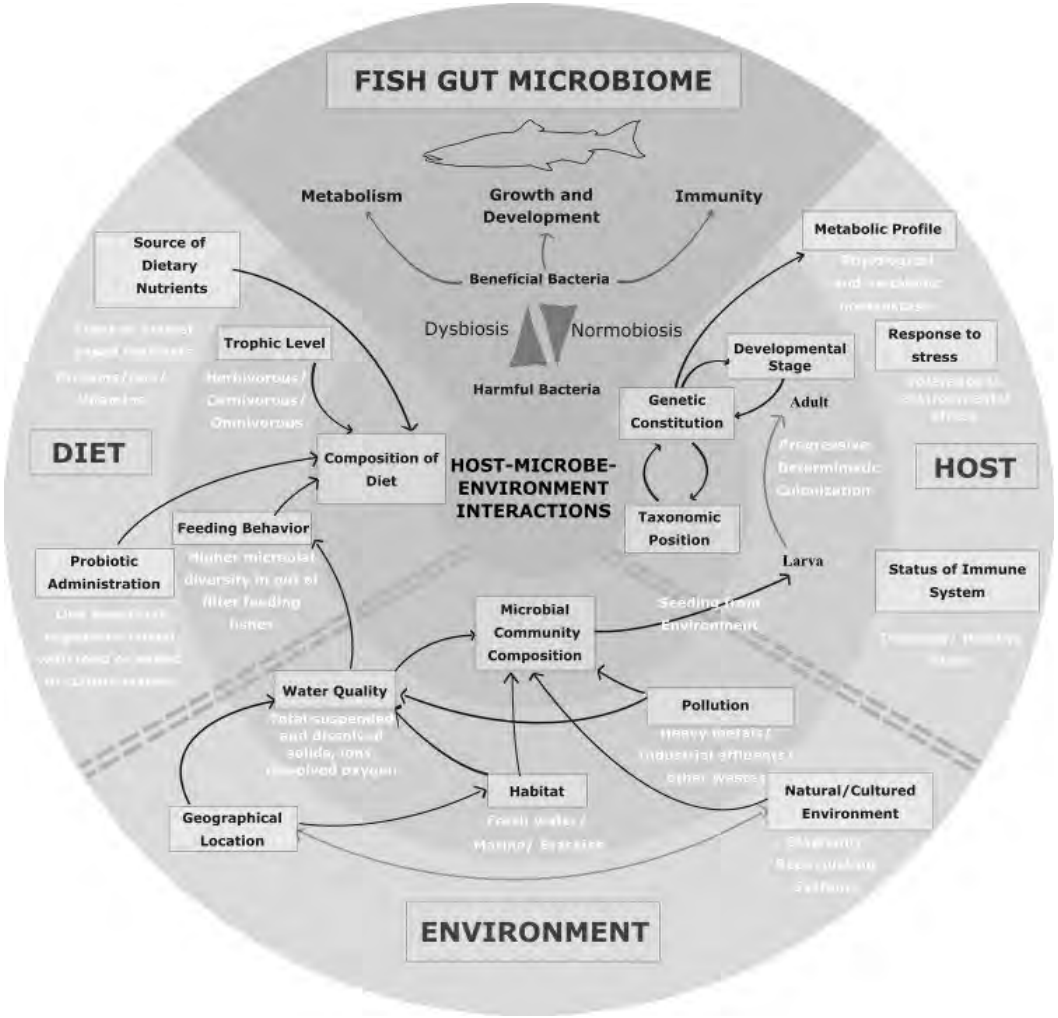
Lanzhou University, China

REVIEWED BY
Prabhugouda Siriyappagouder,
Nord University, Norway
Mehdi Raissy,
Azad University, Iran

Microbiomes in the context of developing sustainable intensified aquaculture

Marlene Lorgen-Ritchie[1], Tamsyn Uren Webster[2], Jamie McMurtrie[3], David Bass[3,4], Charles R. Tyler[3], Andrew Rowley[5] and Samuel A. M. Martin[1*]

Emerging evidence indicates that microbial communities both within and surrounding an animal in aquaculture can contribute directly to productivity in terms of growth, disease resistance and animal welfare.



Aquaculture 519 (2019) 283–292
Contents lists available at ScienceDirect
Aquaculture
journal homepage: www.elsevier.com/locate/aquaculture

Effects of calcium carbonate inclusion in low fishmeal diets on growth, gastrointestinal pH, digestive enzyme activity and gut bacterial community of European sea bass (*Dicentrarchus labrax* L.) juveniles

Luca Parma[a,*], Manuel Yúfera[b], Carmen Navarro-Guillén[b,1], Francisco J. Moyano[c], Matteo Soverini[d], Federica D'Amico[d], Marco Candela[d], Ramon Fontanillas[e], Pier Paolo Gatta[a], Alessio Bonaldo[a]

Aquaculture 518 (2020) 734854
Contents lists available at ScienceDirect
Aquaculture
journal homepage: www.elsevier.com/locate/aquaculture

Effects of rearing density on growth, digestive conditions, welfare indicators and gut bacterial community of gilthead sea bream (*Sparus aurata*, L. 1758) fed different fishmeal and fish oil dietary levels

Luca Parma[a,*], Nicole Francesca Pelusio[a], Enric Gisbert[b], Maria Angeles Esteban[c], Federica D'Amico[d], Matteo Soverini[d], Marco Candela[d], Francesco Dondi[a], Pier Paolo Gatta[a], Alessio Bonaldo[a]

Aquaculture 565 (2023) 739132
Contents lists available at ScienceDirect
Aquaculture
journal homepage: www.elsevier.com/locate/aquaculture

Feeding gilthead sea bream with increasing dietary bacterial single cell protein level: Implication on growth, plasma biochemistry, gut histology, and gut microbiota

Arianna Marchi [a], Alessio Bonaldo [a], Daniel Scicchitano [b], Marco Candela [b], Antonina De Marco [a], Salvatore Falciglia [c], Maurizio Mazzoni [a], Giulia Lattanzio [a], Paolo Clavenzani [a], Francesco Dondi [a], Pier Paolo Gatta [a], Luca Parma [a,*]

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scientific reports

OPEN **Effects of dietary organic acids and nature identical compounds on growth, immune parameters and gut microbiota of European sea bass**

Serena Busti[1], Barbara Rossi[2], Enrico Volpe[3], Sara Ciulli[1], Andrea Piva[1,2], Federica D'Amico[3], Matteo Soverini[3], Marco Candela[3], Pier Paolo Gatta[3], Alessio Bonaldo[1,3], Ester Grilli[1] & Luca Parma[1]

Animal Feed Science and Technology 222 (2016) 204–216
Contents lists available at ScienceDirect
Animal Feed Science and Technology
journal homepage: www.elsevier.com/locate/anifeedsci

Next-generation sequencing characterization of the gut bacterial community of gilthead sea bream (*Sparus aurata*, L.) fed low fishmeal based diets with increasing soybean meal levels

Luca Parma [a,*], Marco Candela [b], Matteo Soverini [b], Silvia Turroni [b], Clarissa Consolandi [c], Patrizia Brigidi [b], Luciana Mandrioli [a], Rubina Sirri [a], Ramon Fontanillas [d], Pier Paolo Gatta [a], Alessio Bonaldo [a]

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Fish and Shellfish Immunology 107 (2020) 324–335
Contents lists available at ScienceDirect
Fish and Shellfish Immunology
journal homepage: www.elsevier.com/locate/fsi

Full length article

Effects of increasing dietary level of organic acids and nature-identical compounds on growth, intestinal cytokine gene expression and gut microbiota of rainbow trout (*Oncorhynchus mykiss*) reared at normal and high temperature

Nicole Francesca Pelusio [a], Barbara Rossi [b], Luca Parma [a,*], Enrico Volpe [a], Sara Ciulli [a], Andrea Piva [a,b], Federica D'Amico [c], Daniel Scicchitano [c], Marco Candela [c], Pier Paolo Gatta [a], Alessio Bonaldo [a], Ester Grilli [a]

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frontiers in Marine Science
ORIGINAL RESEARCH
published: 31 May 2021
doi: 10.3389/fmars.2021.684701

Interaction Between Dietary Lipid Level and Seasonal Temperature Changes in Gilthead Sea Bream *Sparus aurata*: Effects on Growth, Fat Deposition, Plasma Biochemistry, Digestive Enzyme Activity, and Gut Bacterial Community

OPEN ACCESS

Edited by: Fotis Kokou, Wageningen University and Research, Netherlands
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Nicole Francesca Pelusio[1†], Daniel Scicchitano[2†], Luca Parma[1*], Francesco Dondi[1], Elisa Brini[1], Federica D'Amico[2], Marco Candela[2], Manuel Yúfera[3], Neda Gilannejad[3], Francisco Javier Moyano[4], Pier Paolo Gatta[1] and Alessio Bonaldo[1]

Aquaculture 578 (2024) 740093
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journal homepage: www.elsevier.com/locate/aquaculture

Hermetia illucens larvae meal as an alternative protein source in practical diets for gilthead sea bream (*Sparus aurata*): A study on growth, plasma biochemistry and gut microbiota

Serena Busti [a], Alessio Bonaldo [a], Marco Candela [b], Daniel Scicchitano [b], Giulia Trapella [b], Fabio Brambilla [c], Côme Guidou [d], Christophe Trespeuch [d], Federico Sirri [e], Francesco Dondi [a], Pier Paolo Gatta [a], Luca Parma [a,*]

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ALMA MATER STUDIORUM
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Background - Microbiome in Fish

The aim of this study is to explore the gut microbiome modulation induced by circular aquafeed ingredients in gilthead sea bream and European sea bass by analysing several dose-response feeding trials

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https://nextgenproteins.eu/

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M&M - Diets

Gilthead sea bream

Trial 1 – SCP *Corynebacterium glutamicum*

Ingredients %	CTRL	SCP 10	SCP 15	SCP 20
Fishmeal	18.0	18.0	18.0	18.0
SCP (C. glutamicum)	**0.0**	**10.0**	**15.0**	**20.0**
Soy protein concentrate	17.0	8.00	4.00	0.00
Corn gluten	16.8	11.0	8.00	5.50
Sunflower meal	8.61	14.28	16.33	17.13
Soybean meal	14.2	12.8	12.4	12.4
Wheat meal	7.60	8.10	8.40	9.00
Fish oil	4.00	4.00	4.00	4.00
Salmon oil	12.0	12.0	12.0	12.0

Trial 2- SCP Torula yeast

Ingredients (%)	CTRL	SCP 5	SCP 7.5	SCP 10
Fish meal	22.00	17.94	15.92	13.89
SCP (Yeast)	**0.00**	**4.99**	**7.47**	**9.94**
Soy protein concentrate	13.20	13.17	13.16	13.14
Wheat gluten meal	3.07	3.06	3.06	3.06
Maize gluten meal	26.40	26.35	26.32	26.29
Soybean meal	11.44	11.42	11.40	11.39
Wheat meal	9.82	8.62	8.08	7.56
Fish oil	3.22	3.82	4.08	4.34
Rapeseed oil	7.52	7.09	6.93	6.77

Trial 3 – *Hermetia illucens* larvae meal

Ingredients (%)	Control	HI5	HI10	HI15
Fish meal	22.0	18.1	14.1	10.1
Inset meal (Mutatec)	**0.00**	**5.01**	**10.0**	**15.0**
Soy protein Concentrate	13.2	13.2	13.2	13.3
Wheat gluten	3.07	3.08	3.08	3.09
Maize gluten	26.4	26.4	26.5	26.5
Soybean meal	11.4	11.5	11.5	11.5
Wheat meal	9.82	8.47	7.12	5.79
Fish oil	3.22	3.71	4.09	4.56
Rapeseed oil	7.52	6.89	6.37	5.76

Trial 4 - By-product from fisheries and aquaculture

Ingredients (%)	Ctrl	0FM100FO	0FM0FO	0FM0FO-50SP	0FM0FO-0SP
Fish meal	25.0	-	-	-	-
FM Trimming	-	25.0	25.1	25.2	25.3
Fish Oil Extra	14.6	14.25	-	-	-
FO Trimming	-	-	14.25	12.5	12.5
Krill meal	2.5	-	-	-	-
Shrimp meal Trimming	-	2.5	2.6	8.5	8.6
Soy protein concentrate	8,0	8,0	8,0	4,0	-
Corn Gluten	4,0	4,0	4,0	10,0	15.5
Wheat Gluten	-	2.6	2.5	2.75	9.5
Soybean meal	19.2	19.3	19.4	9.6	-
Wheat meal	14.5	14.3	14.4	14.0	18.2

M&M – Fish and experimental set up

Trial 1- SCP *C. glutamicum*

- 45 fish per tank
- Initial weight 75 g
- 12, 500 L tanks
- Temperature 23 ± 1° C;
- Triplicate treatments
- Satiation twice a day
- The trial lasted 108 days

Trial 2- SCP T. yeast

- 40 fish per tank
- Initial weight 25 g
- 12, 500 L tanks
- Temperature 24 ± 1° C;
- Triplicate treatments
- Satiation twice a day
- The trial lasted 76 days

Trial 3 – *H. illucens*

- 50 fish per tank
- Initial weight 98 g
- 12, 500 L tanks
- Temperature 24 ± 1° C;
- Triplicate treatments
- Satiation twice a day
- The trial lasted 113 days

Trial 4 – By-product

- 50 fish per tank
- Initial weight 75 g
- 15, 800 L tanks
- Temperature 24 ± 1° C;
- Triplicate treatments
- Satiation twice a day
- The trial lasted 119 days

M&M – Fish and experimental set up

Growth Performance Parameters → **FBW, SGR, FCR, FI**

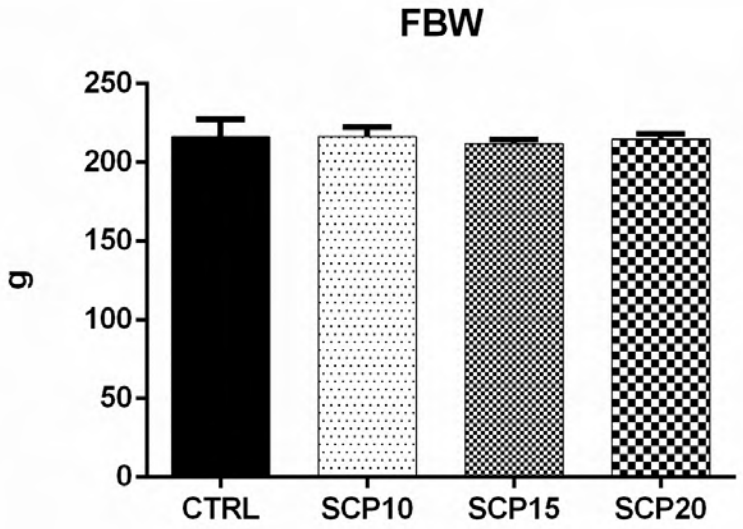
Gut microbiome

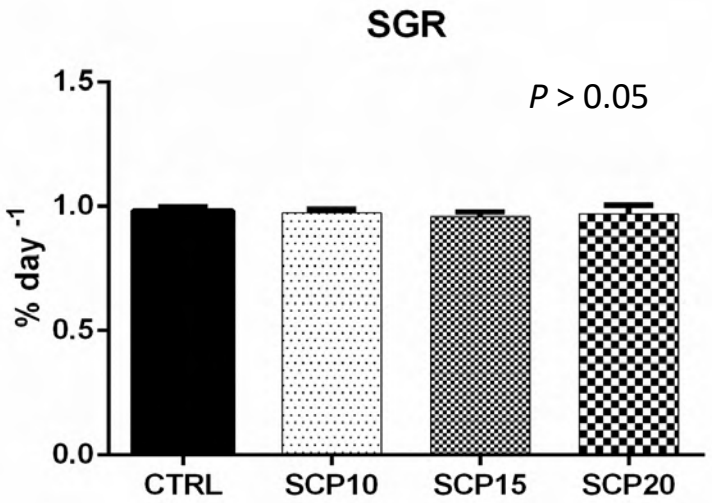
DNA extraction from distal intestine content (15 fish/treatment) → **Prediction of metagenome functions using Picrust2**

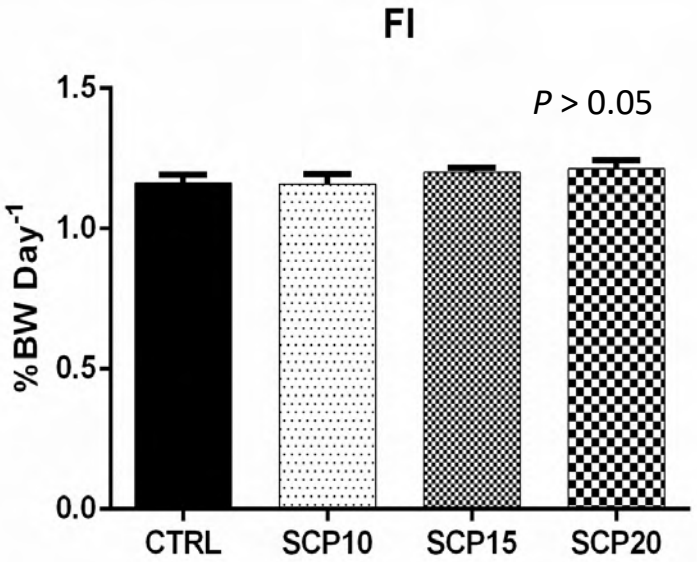
Douglas et al 2020 Nat. Biotech (2020)

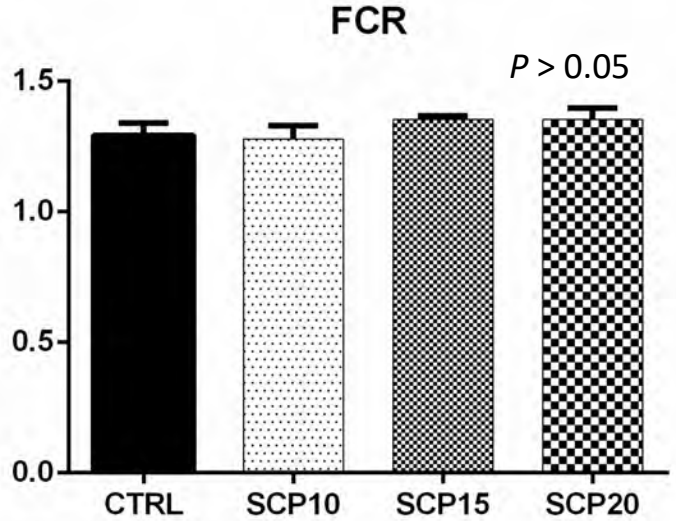
→ Illumina NGS of the V3 and V4 regions of the 16S rRNA gene (QIIME2; PANDAseq, VSEARCH)

Results/Growth -Trial 1 - SCP *Corynebacterium glutamicum*

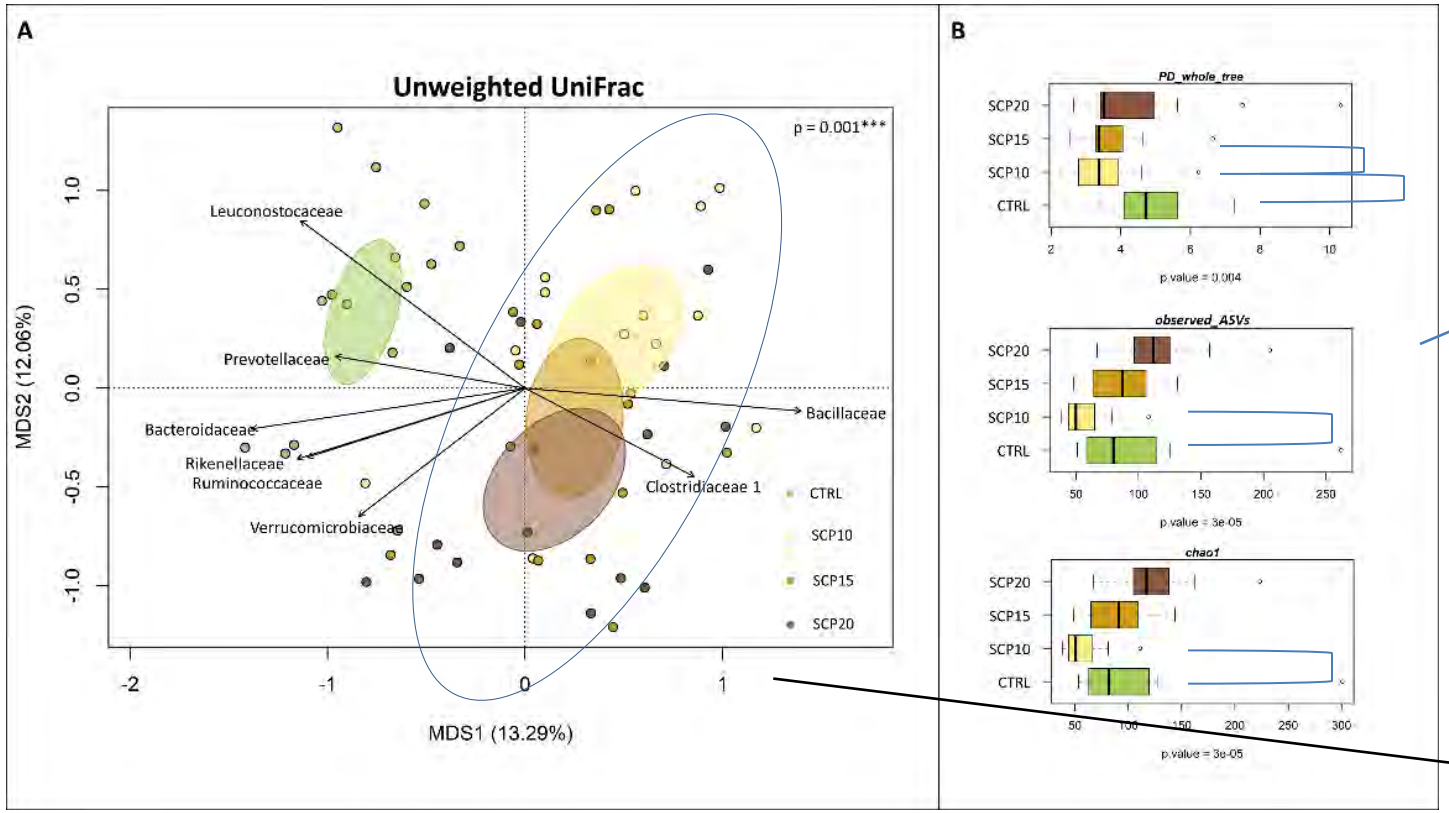








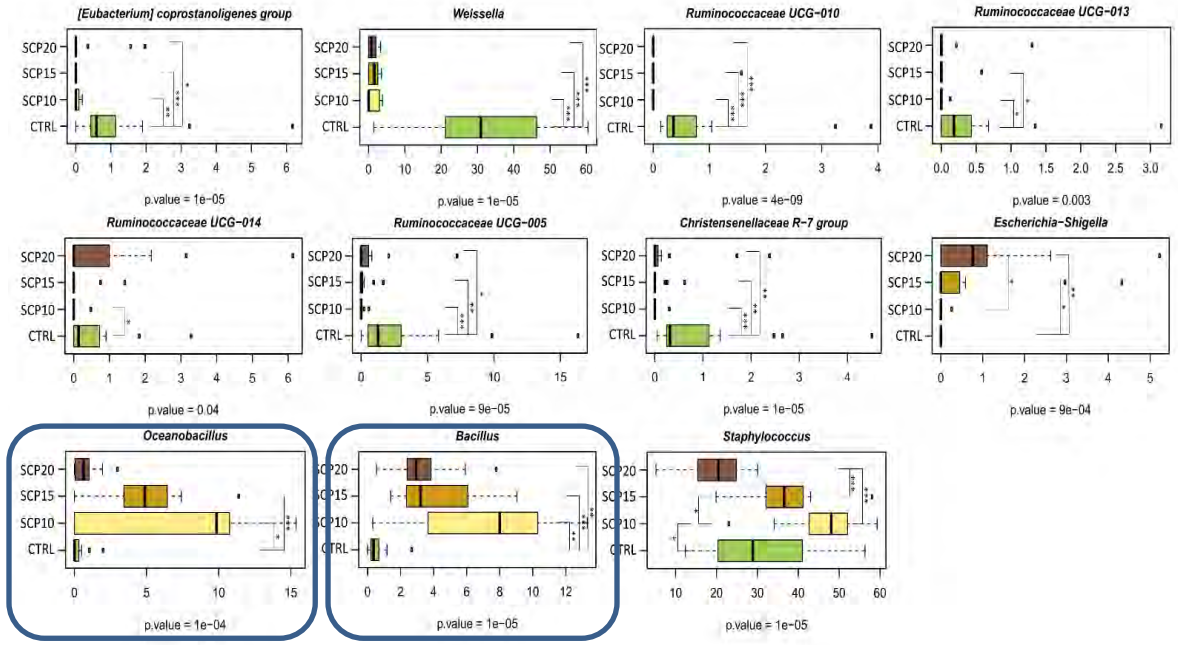
Results microbiome - Trial 1 SCP-*Corynebacterium glutamicum*



Inclusion at 10 and 15% showed a reduced alpha diversity indexes

SCP groups showed a significant variation compared to the control group, in terms of overall microbiome composition

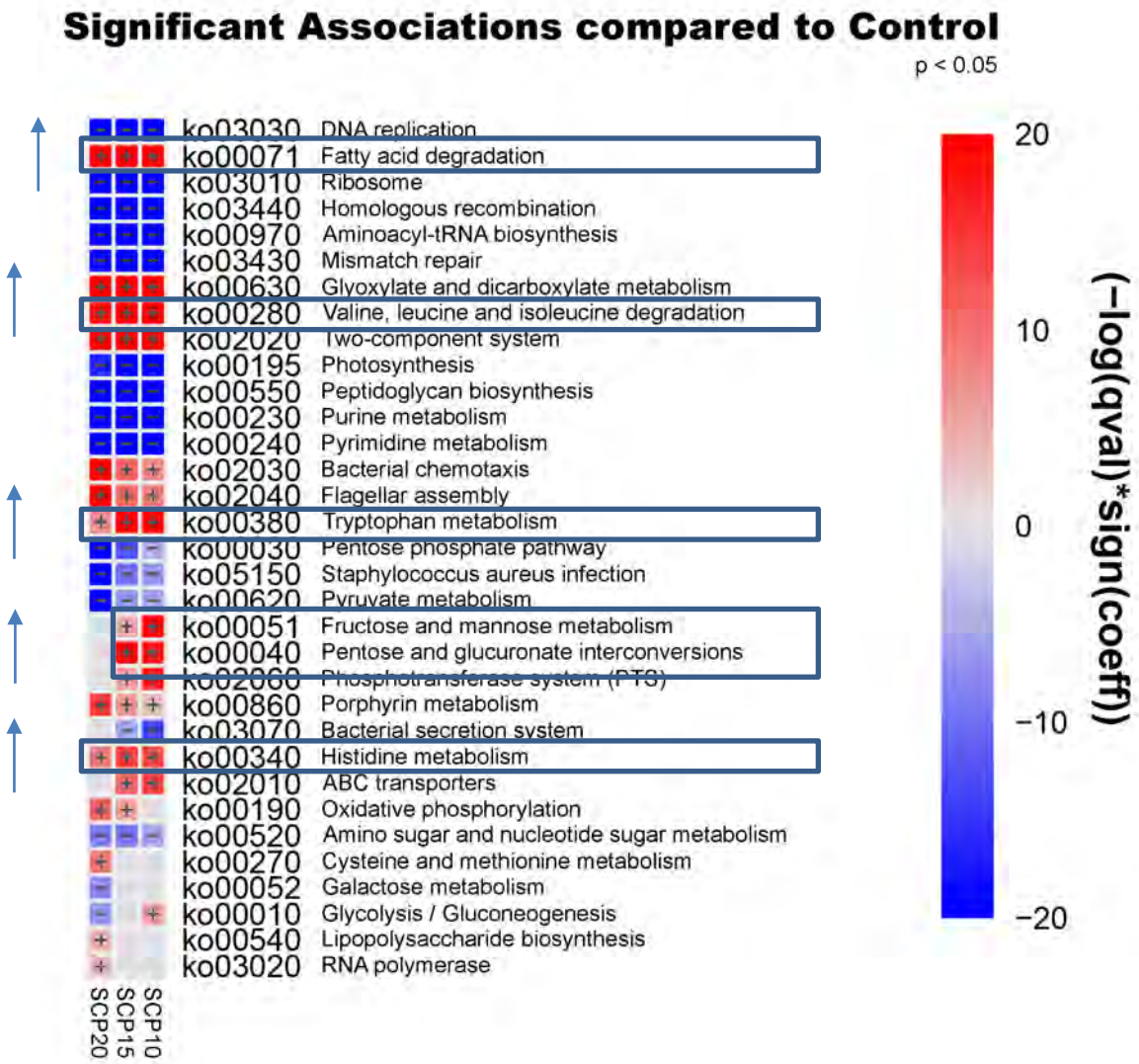
Results microbiome - Trial 1 SCP-*Corynebacterium glutamicum*



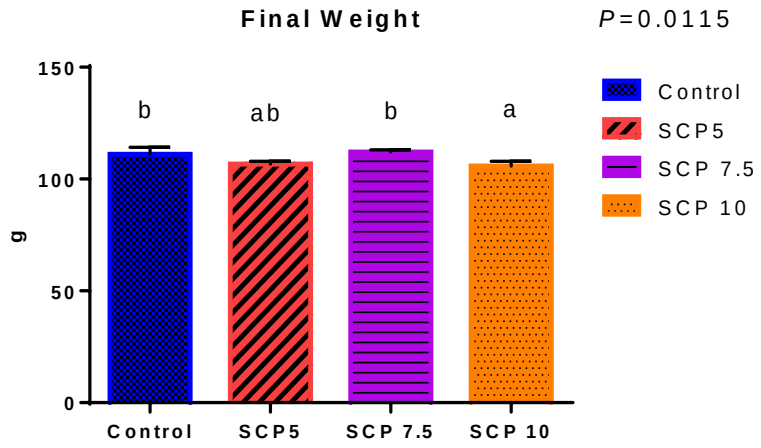
SCP led to increase

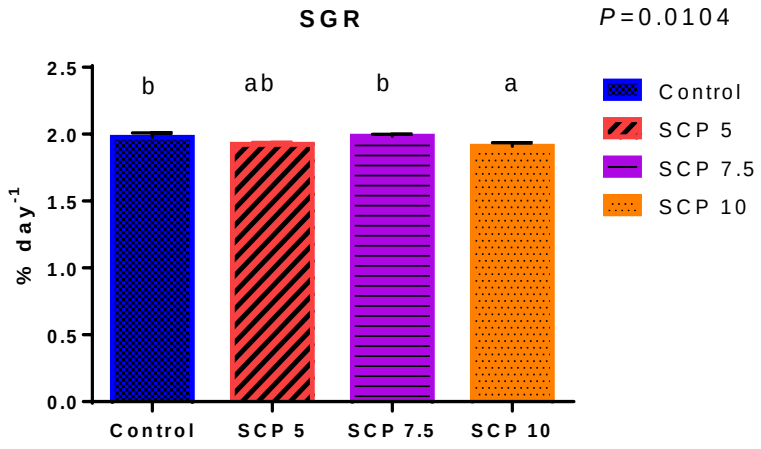
- *Bacillus*
- *Oceanobacillus*

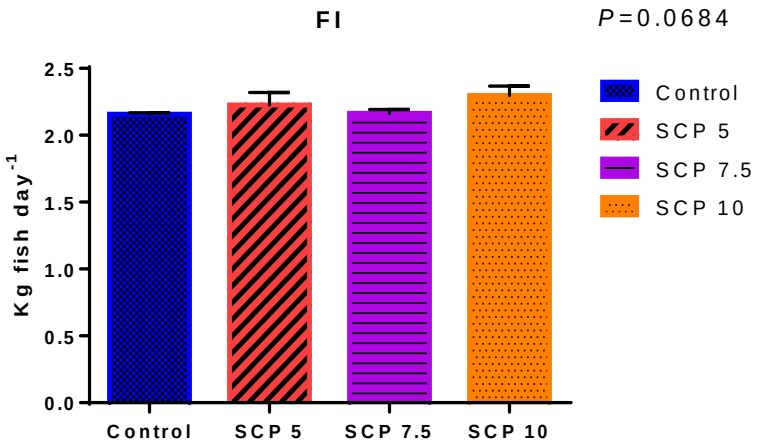
Prediction of metagenome functions

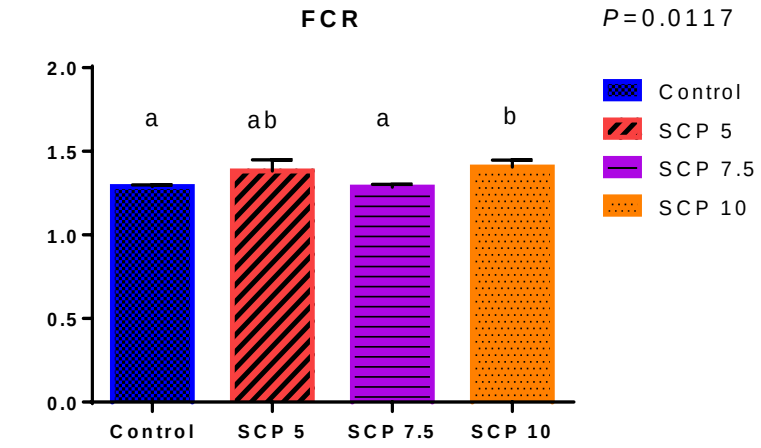


Results Growth - Trial 2 SCP Torula yeast

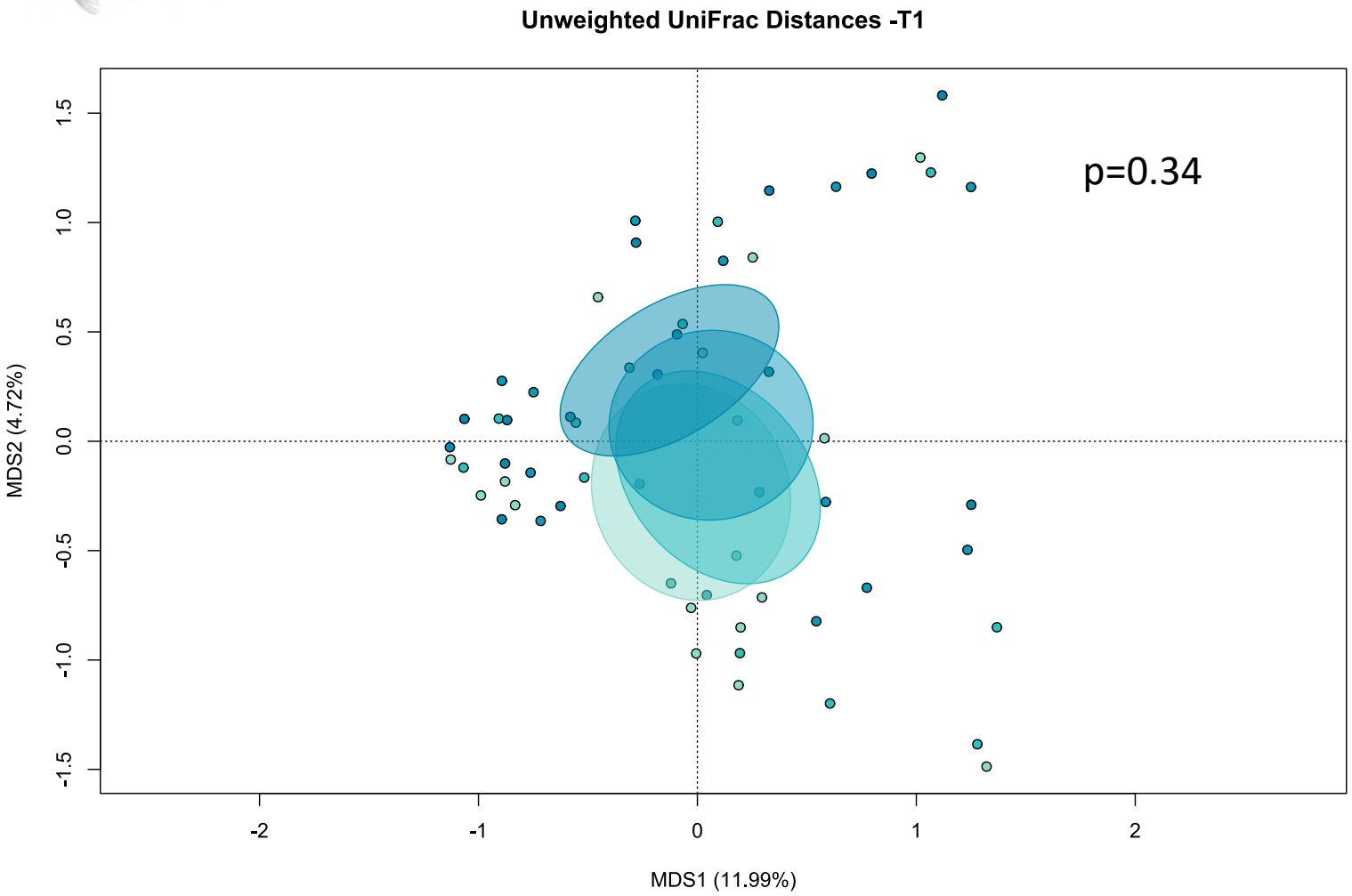


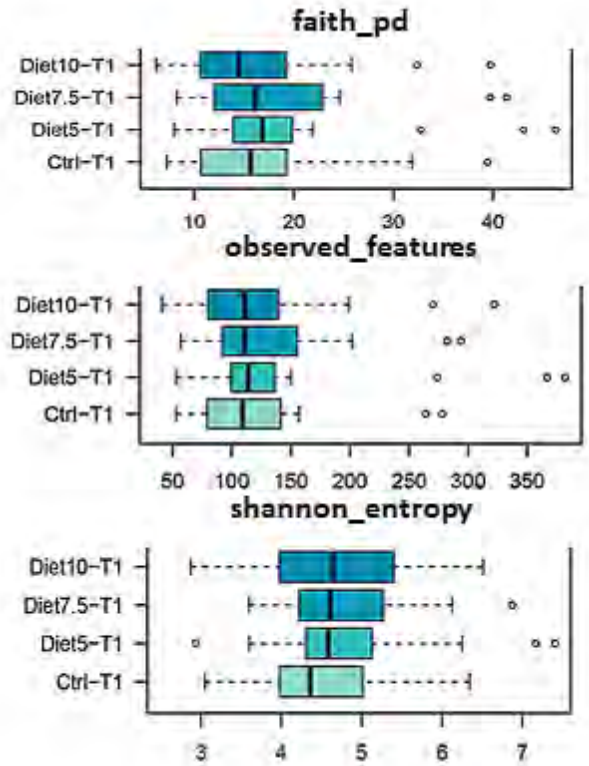






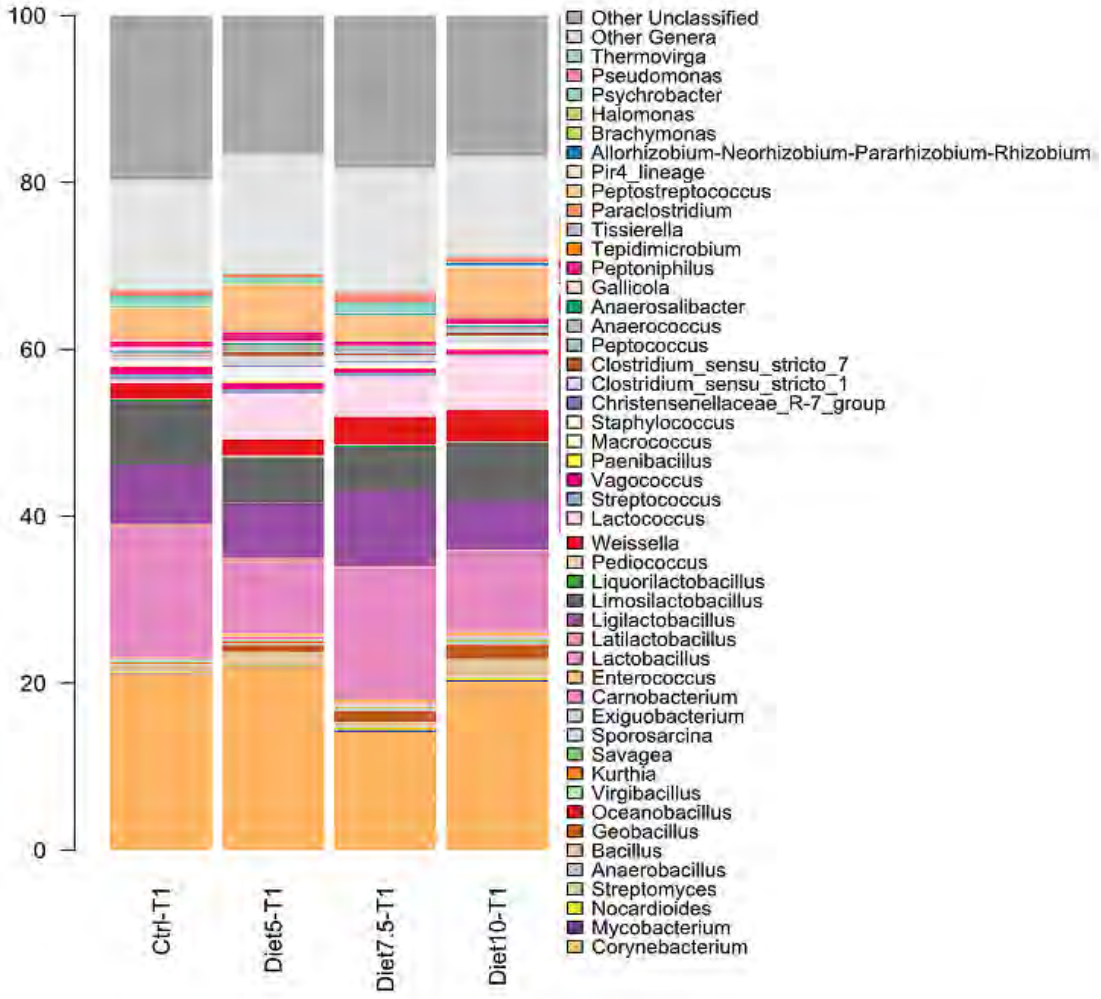
Results Microbiome - Trial 2 SCP/Torula yeast



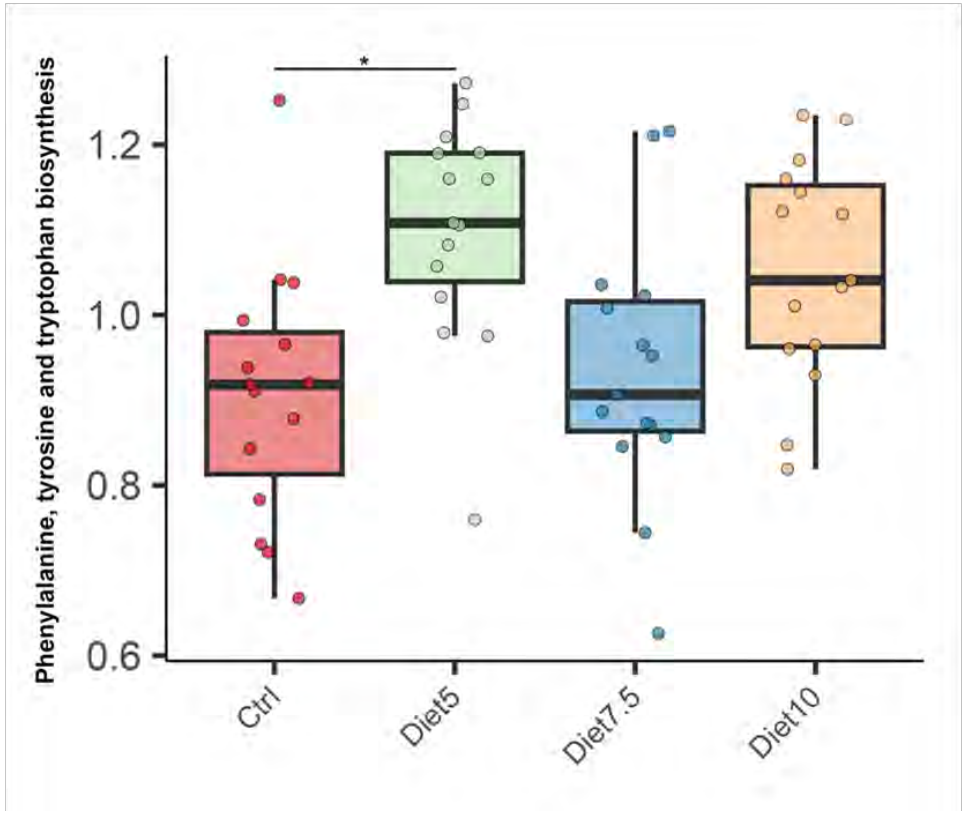


Results Microbiome - Trial 2 SCP - Torula yeast

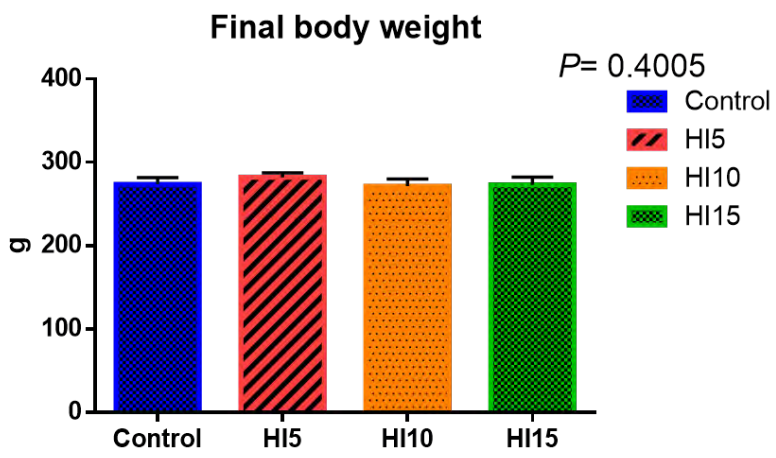
Prediction of metagenome functions

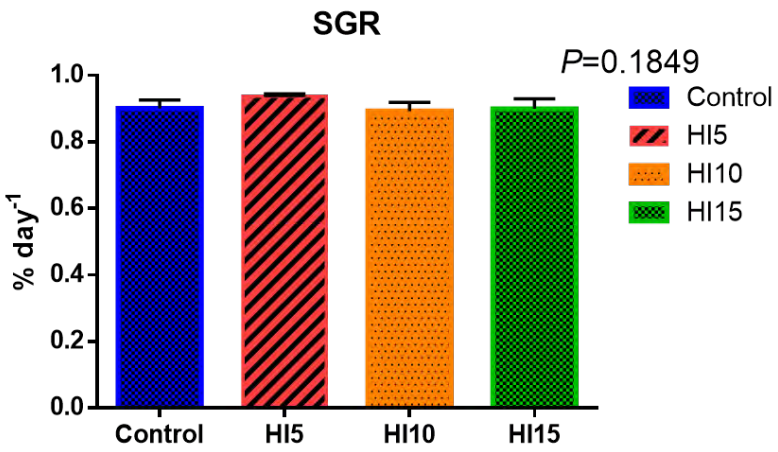


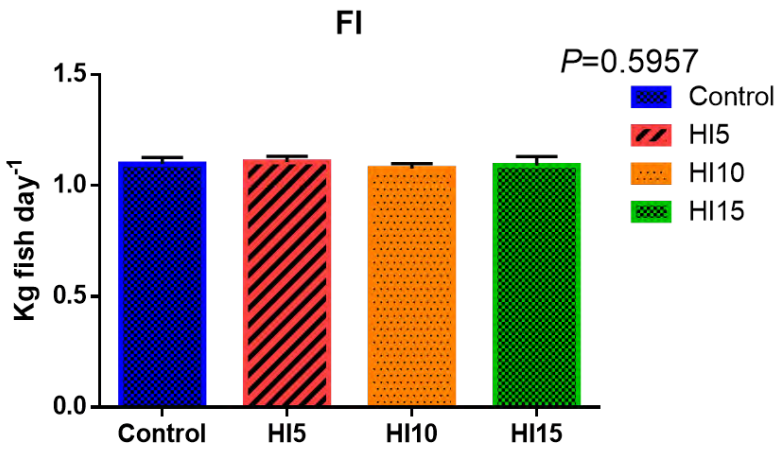
Significant variations Pathway within Timepoint1 Diet groups (MaAsLin2)

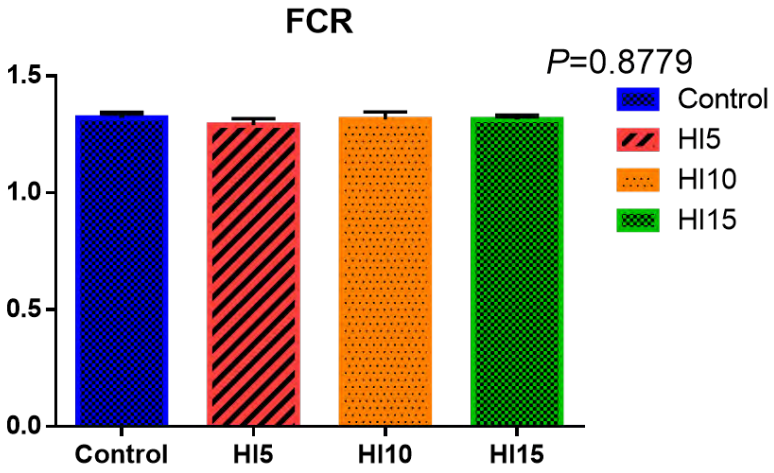


Results Growth - Trial 3 *Hermetia illucens* larvae meal

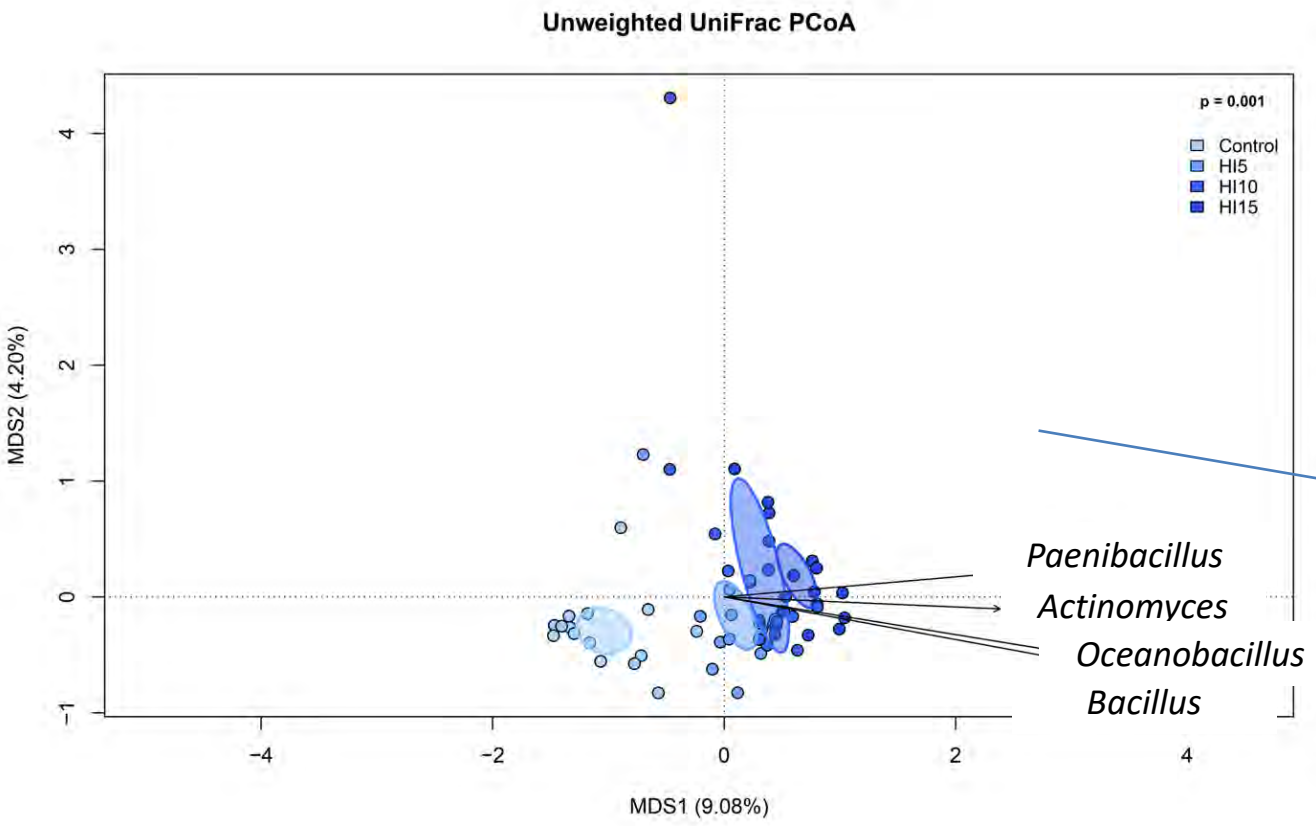


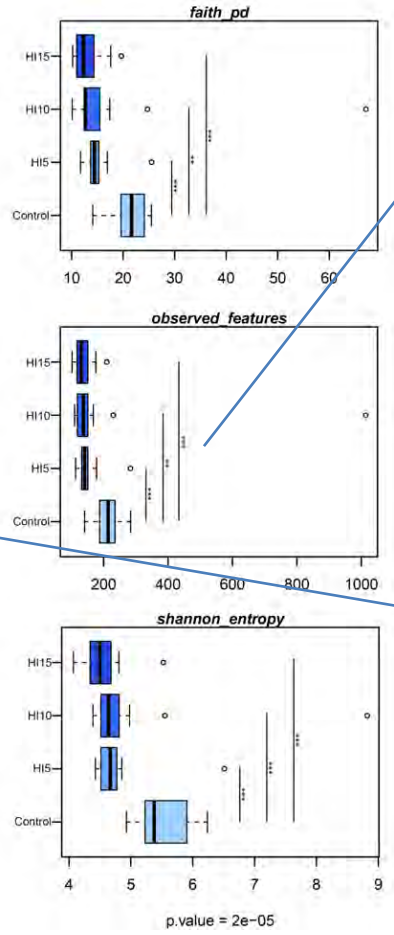






Results microbiome - Trial 3 *Hermetia illucens* larvae meal

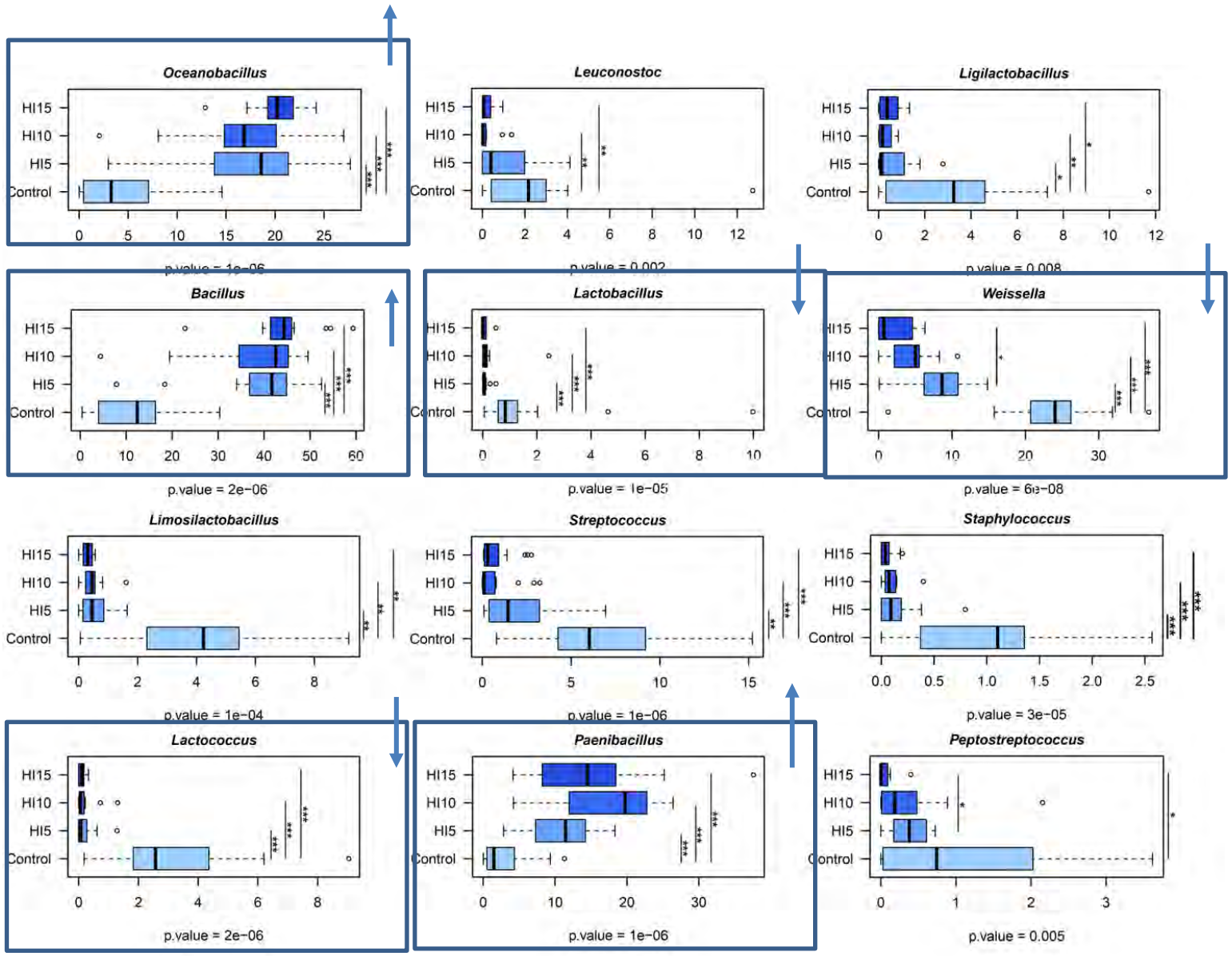




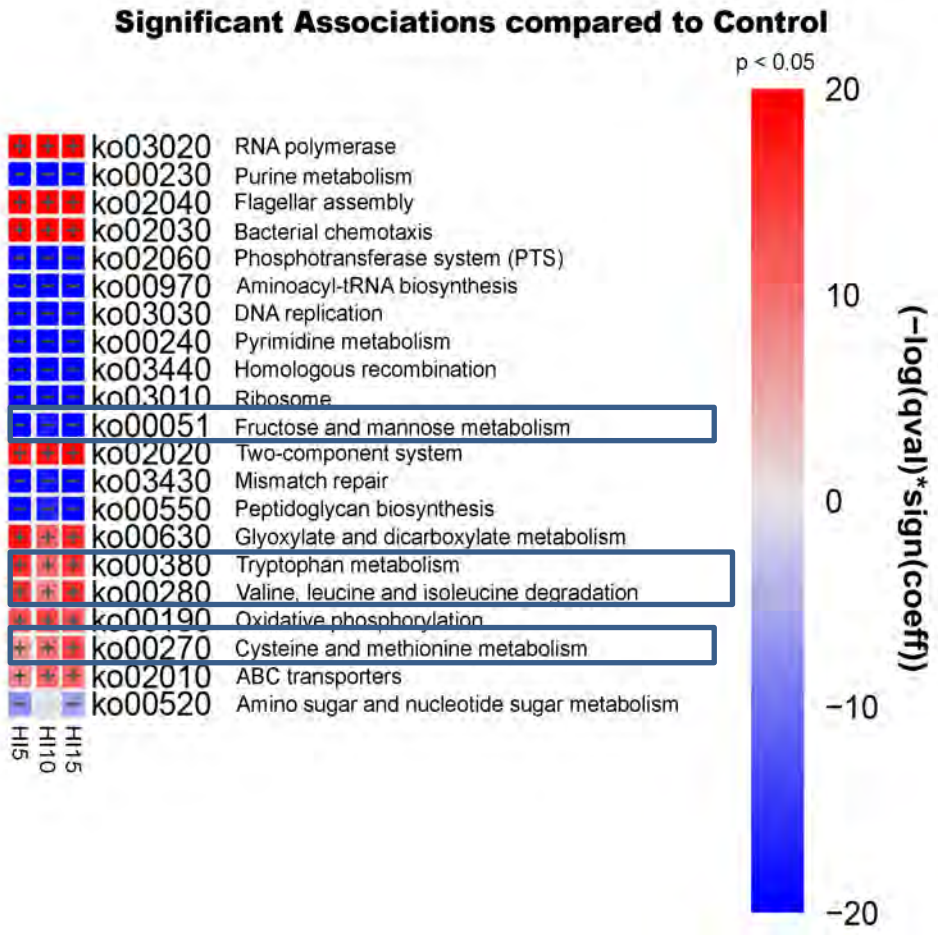
Inclusion of Hermetia Illucens showed a reduced alpha diversity indexes

SCP groups showed a significant variation compared to the, control group in terms of overall microbiome composition

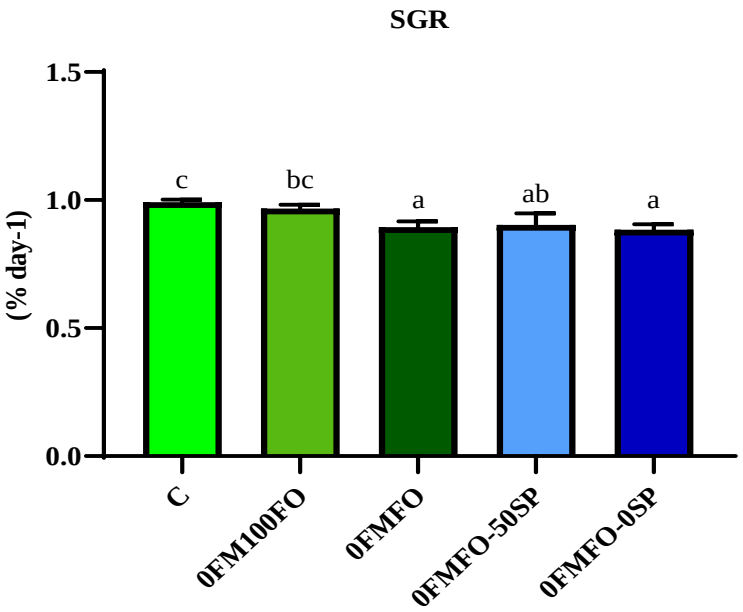
Results microbiome - Trial 3 *Hermetia illucens* larvae meal

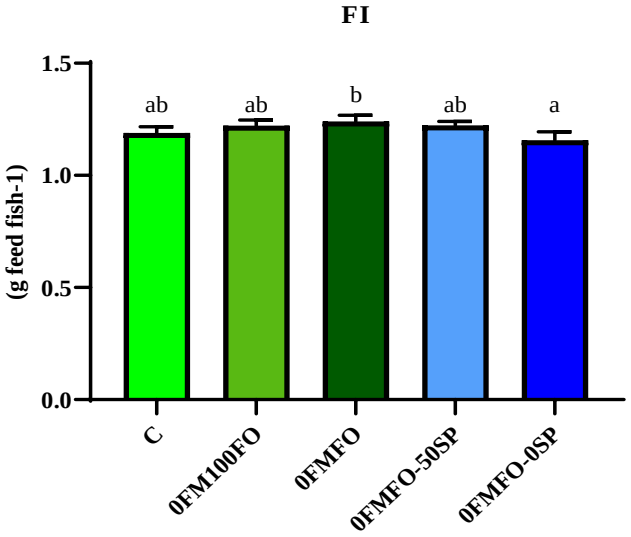


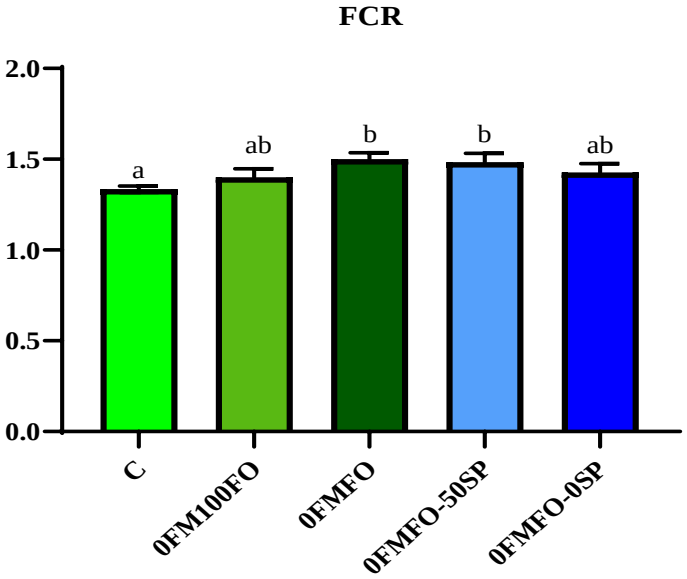
Prediction of metagenome functions



Results growth - Trial 4 By-products from fisheries and aquaculture







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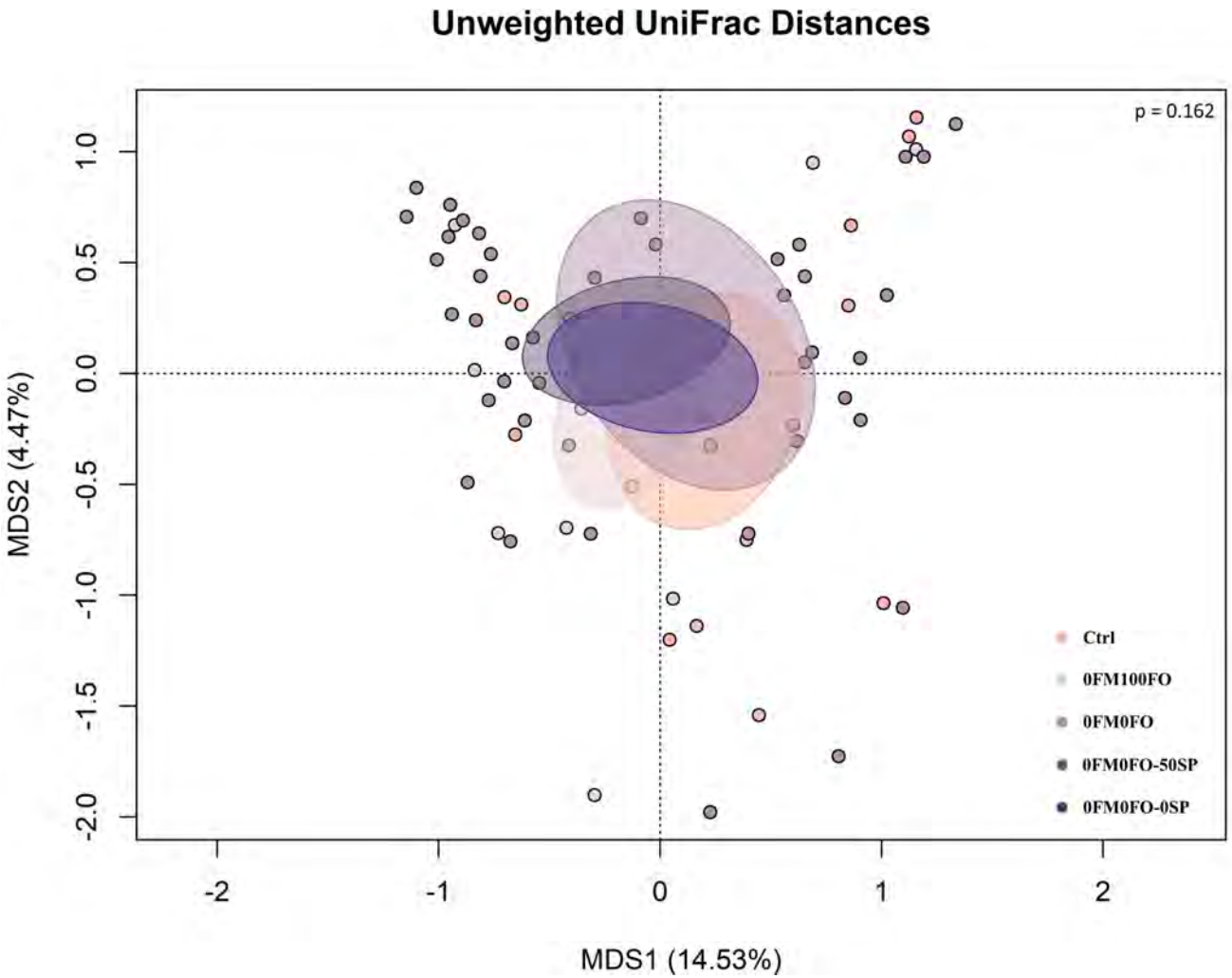
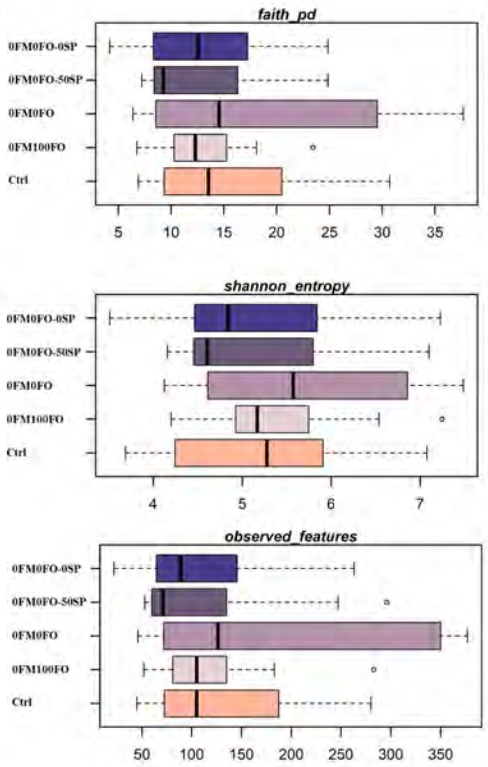
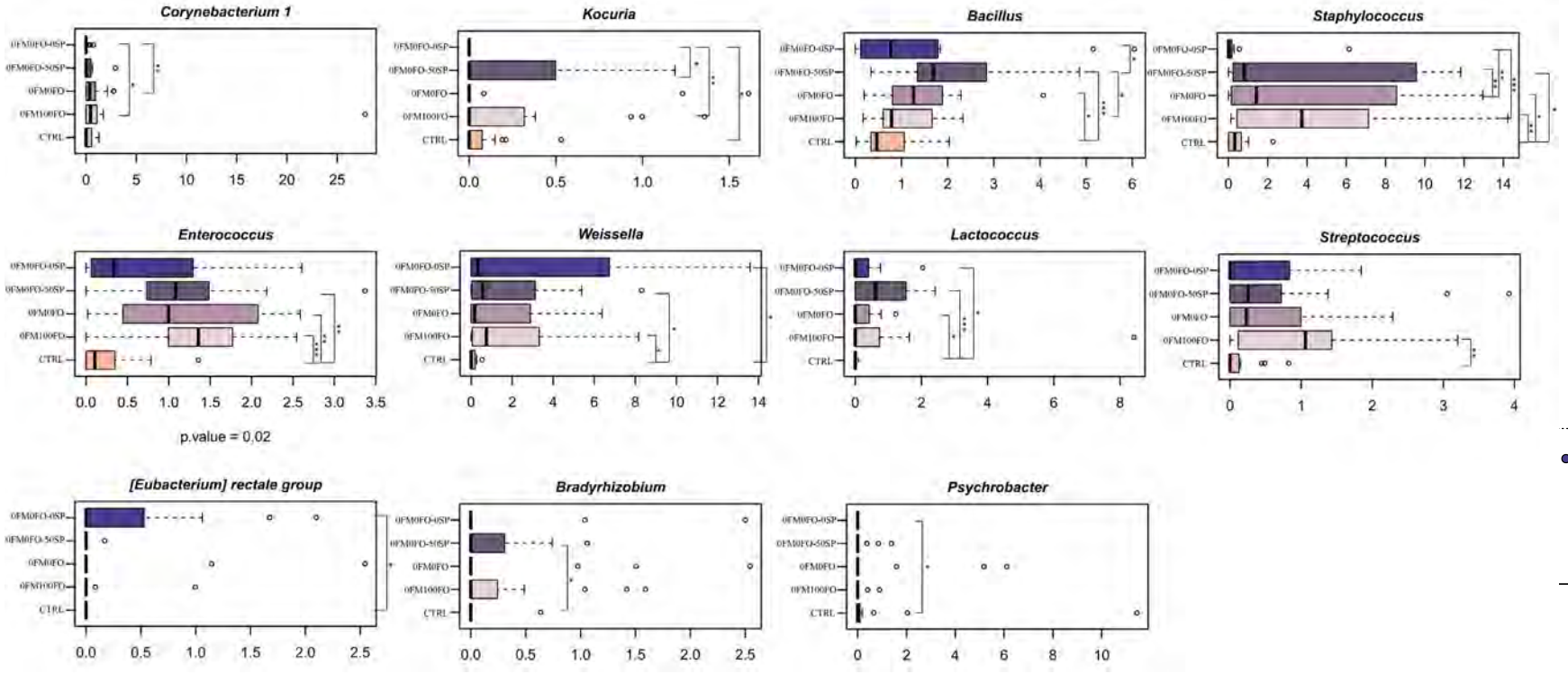
Results microbiome - Trial 4 By-products

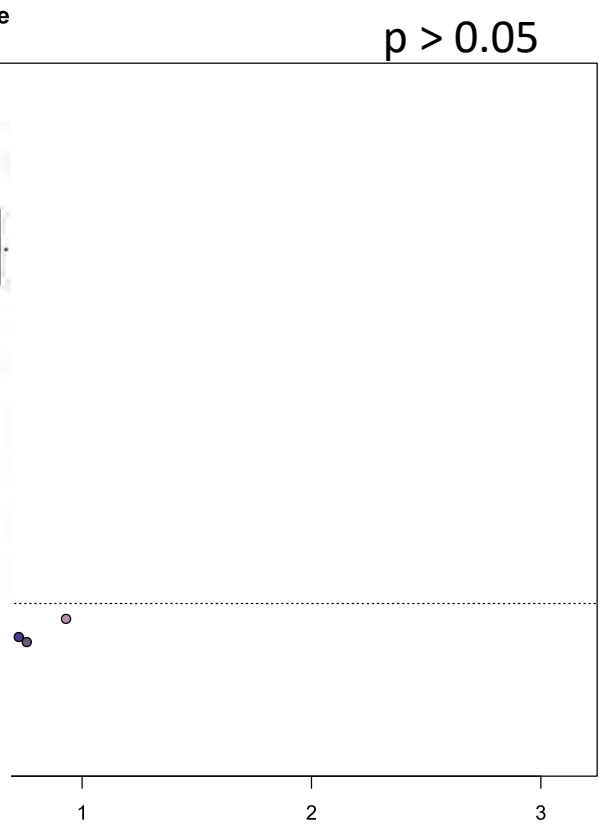




Results microbiome - Trial 4 By-products from fisheries and aquaculture

Prediction of metagenome functions





Conclusion

- Inclusion of SCP from *C glutamicum* (*up to 20%*) and insect meal (*up to 15%*) led to equal growth and feed utilization in comparison to control diets

- In both trial fish were associated to a gut microbiome reconfiguration enriched by potential beneficial taxa *Bacillus, Oceanobacillus,* (*both*)*; Paenibacillus (HI*)

- Predicted metagenome functions indicated an increased in amino acid metabolism such as tryptophan, valine, leucine and isoleucine (both) histidine (SCP), methionine (HI)

- Bacterial tryptophan metabolites, such as indole and indolic acid derivatives, are potent bioactive metabolites that affect intestinal barrier integrity and immune cells in mice

Future perspective

Development of welfare indicators microbiome-based
in farmed fish species

Thanks for the attention!

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Microbiome Science and Biotechnology unit – Dept. of Pharmacy and Biotechnology

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