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EFFECTS OF DIETARY ETHYL ESTER OIL ON DIGESTIVE ENZYMES, INTERMEDIARY METABOLISM, AND MUSCLE FATTY ACID PROFILE IN EUROPEAN SEABASS (*Dicentrarchus labrax*)

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Introduction

Ethyl ester (EE) oil is a by-product of fish oil (FO) processing industry for the production of omega-3 for human consumption and still contains 4–10% n-3 long-chain polyunsaturated fatty acids (LC-PUFA), representing a potential sustainable and value-added lipid source for aquafeeds. However, the bioavailability of fatty acid ethyl esters may differ from that of triglycerides (TAG) found in conventional oils.

The main question is whether juvenile seabass can metabolize and utilize fatty acids (FA) delivered as EE to the same extent as FA supplied through conventional triacylglycerol (TAG)-based oils.

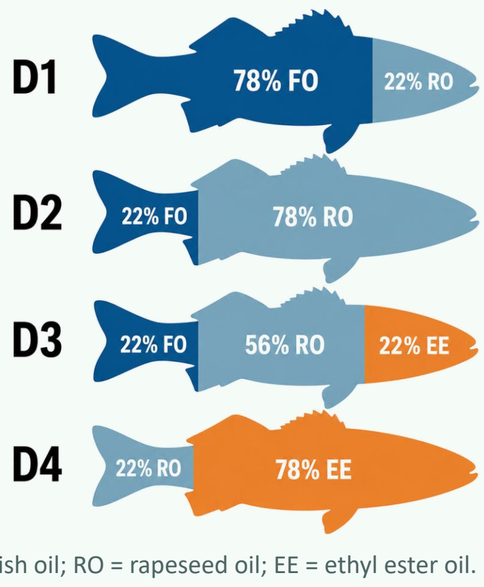
Objective

Evaluated the effects of replacing dietary FO with EE oil on:

- Growth performance and Feed utilization
- Digestive enzyme activity
- Hepatic intermediary metabolism
- Muscle FA profile

Diet formulation

Four isoproteic (48%) and isolipidic (16%) diets were formulated using different supplemental oil blends (%).



FO = fish oil; RO = rapeseed oil; EE = ethyl ester oil.
- Values represent the proportion of each oil source in the added oil blend, not the percentage of the complete diet.
- Four diets were comparable in analyzed composition, containing 47.5–48.7% crude protein, 15.9–16.4% crude lipid, 5.77–6.32% ash, and 24.3–25.4 kJ g^{-1} gross energy (dry matter basis); dry matter ranged from 91.5 to 95.8%.

Experimental design

- European seabass juveniles：
  62 ± 0.5 g initial body weight
- 12 tanks: 15 fish/tank; 3 tanks per diet
- 88-day feeding trial in RAS at 23 ± 1 °C
- Hand-fed to apparent satiation, 6 days/week
- One-way ANOVA + Tukey's test, $p < 0.05$

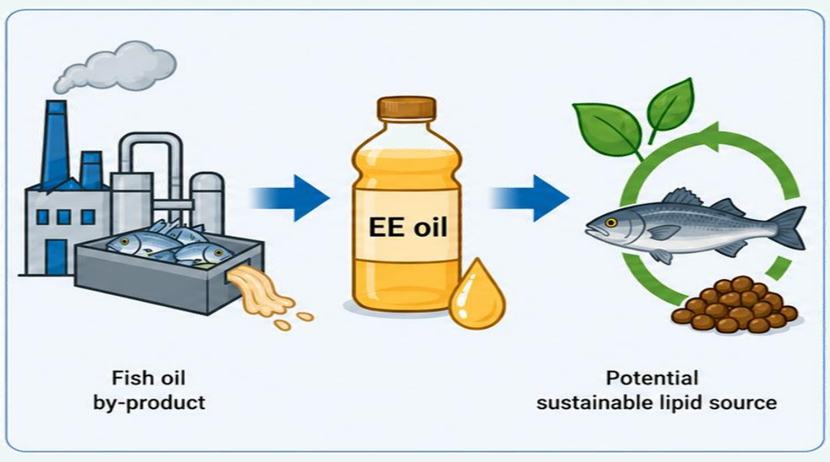
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BACKGROUND



Differences in Chemical Structure

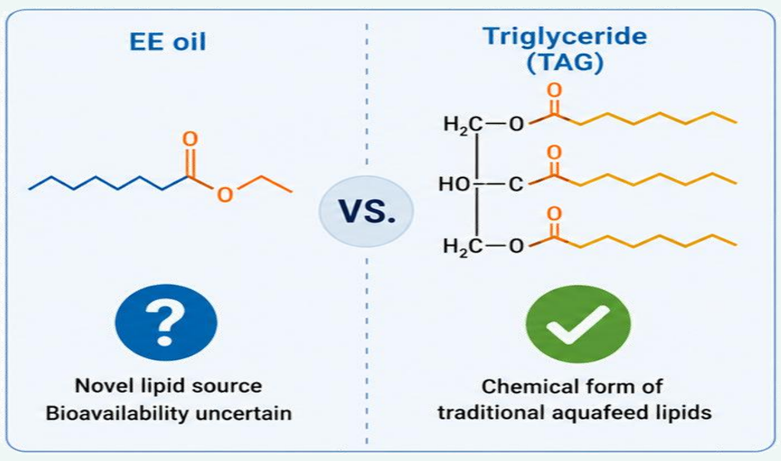


TABLE 1

Growth performance, feed utilization, nitrogen, and energy retention of European seabass fed the experimental diets.

No significant dietary effects were detected (one-way ANOVA; $P > 0.05$).

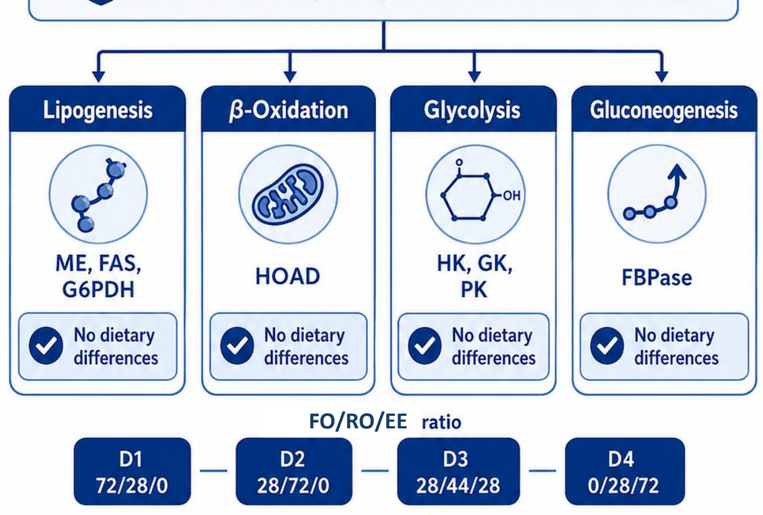
Parameter	D1 78/22/0	D2 22/78/0	D3 22/56/22	D4 0/22/78	SEM	*P*-value
1. Initial body weight (IBW, g)	61.7	61.7	61.7	61.7	0.00	0.963
2. Final body weight (FBW, g)	154.7	146.6	151.2	147.8	1.59	0.283
3. Weight gain (g kg ABW^{-1} day^{-1})	9.77	9.26	9.55	9.35	0.10	0.296
4. Daily growth index [1]	1.61	1.50	1.56	1.52	0.02	0.290
5. Feed intake (g kg ABW^{-1} day^{-1})	13.1	12.2	12.9	13.1	0.19	0.261
6. Feed efficiency [2]	0.75	0.76	0.74	0.71	0.01	0.340
7. Protein efficiency ratio [3]	1.55	1.62	1.55	1.45	0.03	0.051
8. Survival (%)	100.0	95.6	100.0	100.0	0.96	0.085
9. Nitrogen retention (% NI) [4]	28.4	27.9	27.1	26.5	0.37	0.719
10. Energy retention (% EI) [5]	22.9	23.4	24.2	22.6	0.31	0.977

** FO: fish oil; RO: rapeseed oil; FAEE: Fatty Acid Ethyl Esters oil.*
Values are presented as mean (n=3). SEM: Pooled standard error of the mean.
ABW: Average body weight = ((initial body weight + final body weight)/2).
[1] DGI: ((final body weight^(1/3) – initial body weight^(1/3)) / time in days) × 100.
[2] FE: wet weight gain / dry feed intake.
[3] PER: wet weight gain / crude protein intake.
[4] Nitrogen retention = ((FBW × FBN – IBW × IBN) / NI) × 100.
[5] Energy retention = ((FBW × FBE – IBW × IBE) / EI) × 100.
IBN/FBN or IBE/FBE: initial and final body nitrogen or energy content;
NI: nitrogen intake; EI: energy intake; IBW and FBW: initial and final body weight.

TABLE 2

Liver key enzyme activity in European seabass fed experimental diets
(mU mg^{-1} protein)

No significant dietary effects were detected for liver key enzyme activities ($P > 0.05$).

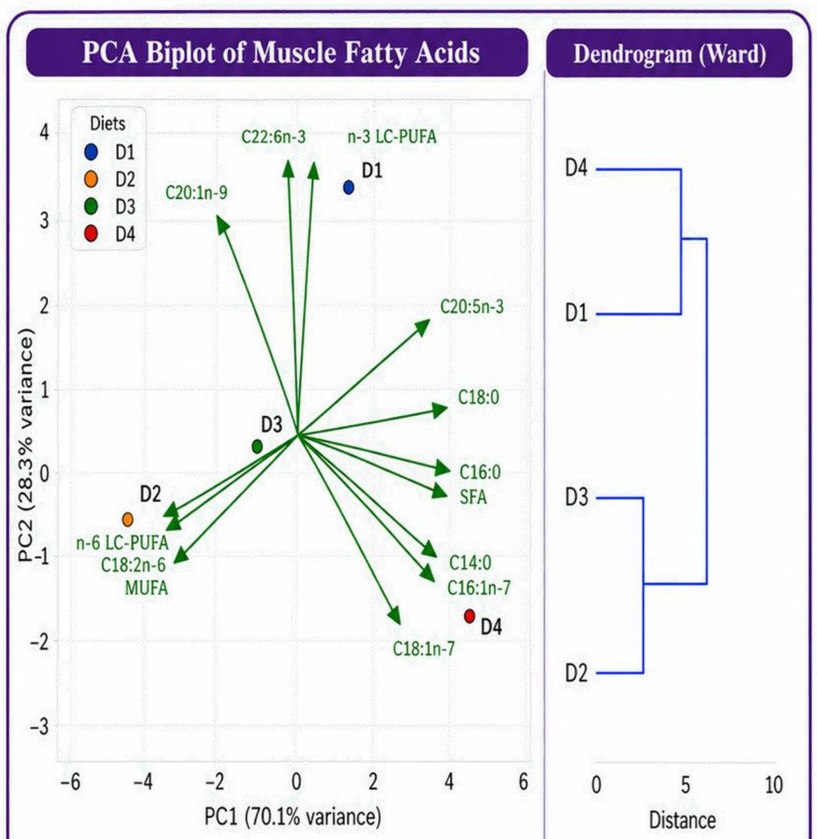


Nine livers were sampled from fish in each dietary treatment.
ME, malic enzyme; FAS, fatty acid synthase; G6PDH, glucose 6-phosphate dehydrogenase; HOAD, 3-hydroxyacyl-CoA dehydrogenase; HK, hexokinase; GK, glucokinase; PK, pyruvate kinase; FBPase, fructose bisphosphatase.

FIGURE 1

Principal Component Analysis (PCA) biplot and dendrogram of muscle FA composition of European seabass fed the experimental diets.

PCA and Ward clustering grouped D1 with D4, and D2 with D3.

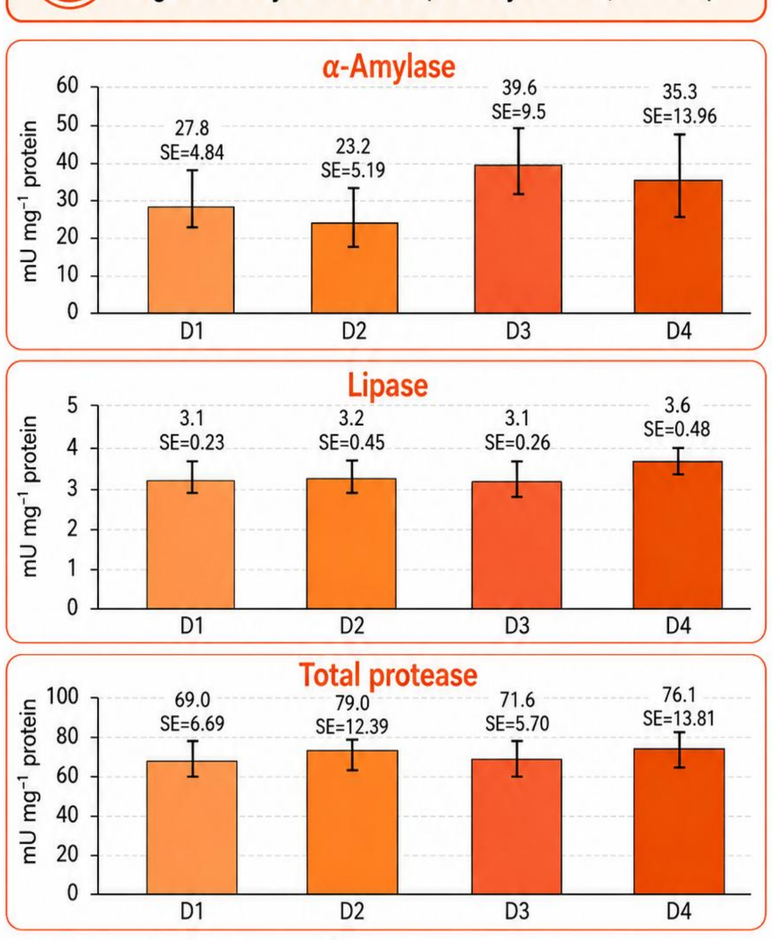


Nine fish muscles were sampled from each dietary treatment.

TABLE 3

Intestinal digestive enzyme activities (mU mg^{-1} protein) in the intestine of European seabass fed the experimental diets.

No significant dietary effects were detected for intestinal digestive enzyme activities (one-way ANOVA; $P > 0.05$).



Values are presented as means ± SE.

Nine fish intestines were sampled from each dietary treatment.

Conclusion

- Dietary EE oil did not impair growth or feed utilization in European seabass juveniles.
- Digestive enzymes and hepatic intermediary metabolism remained unchanged across diets.
- Muscle FA profile was modified: DHA and total n-3 LC-PUFA decreased, while the n-6/n-3 ratio increased in D2-D4 vs D1.
- Muscle EPA was maintained in D4 at levels comparable to D1, supporting EE oil as a promissing marine lipid source, although fillet DHA retention remains a challenge.