EFFECTS OF LYOPHILIZED DIETARY YEAST *Rhodotorula mucilaginosa* ON THE SKIN AND FILLET PIGMENTATION OF GILTHEAD SEABREAM *Sparus aurata*. A COMPUTER-BASED IMAGE ANALYSIS APPROACH.

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Introduction

Fish pigmentation assessment is commonly conducted via the CIE Lab model with a colorimeter. However, the non-homogeneous coloration of fish presents challenges for accurate measurement. Computer-based image analysis offers a promising alternative, providing homogeneous conditions without direct tissue contact. Gilthead sea bream, reliant on dietary carotenoids for coloration, can experience characteristic discoloration under intense rearing conditions. Carotenoid supplementation, including natural sources like yeasts, has been explored to mitigate this effect. *Rhodotorula mucilaginosa*, a yeast species capable of producing pigments, presents a potential solution.

Objective

The aim of this study was to evaluate the efficacy of *R.mucilaginosa* as a pigmentation source for gilthead sea bream using computer-based image analysis.

Experimental rearing

Potato Dextrose Agar

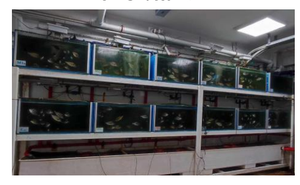
Lyophilized biomass
Rhodotorula mucilaginosa

Inclusion levels

0% (C)
1% (RM1)
2% (RM2)
3% (RM3)

Average initial weight
199.8 ± 1.56 gr
Feeding 1.2% BW

4 treatment (C, RM1, RM2, RM3)
3 replicates/ treatment
3 x 4 = 12 tanks (174L)
10 fish/tank



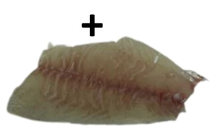
3-month rearing

Materials and methods

3 replicates /treatment
6 fish and fillets/ replicates
6 x 3 x 4 = 72 fish samples
6 x 3 x 4 = 72 fillet samples

Average final weight 308 ± 3.75 gr

+



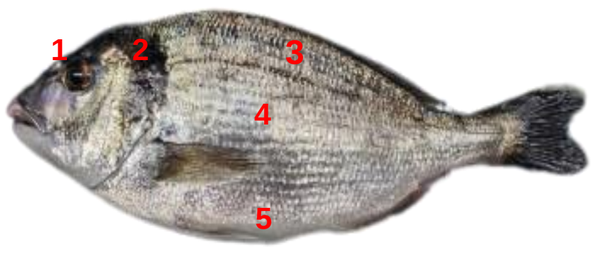
Puluz Photobox
60x60x60 cm

White background

Wide-angled camera
Resolution 12 MP
(4032 x 3024 Pixel)

Adobe Photoshop Version 23.5.1
was used for the image analysis

After color correction 5 spots across the specimen s' body and 3 spots on the fillet.



Chosen coordinates for color assessment

CIELAB color space

- L* (Lightness)
- a* (Redess)
- b* (Yellowness)
- Chroma = $\sqrt{(a^*)^2+(b^*)^2}$
- Whiteness = 100 - $\sqrt{(100-L^*)^2+(a^*)^2+(b^*)^2}$

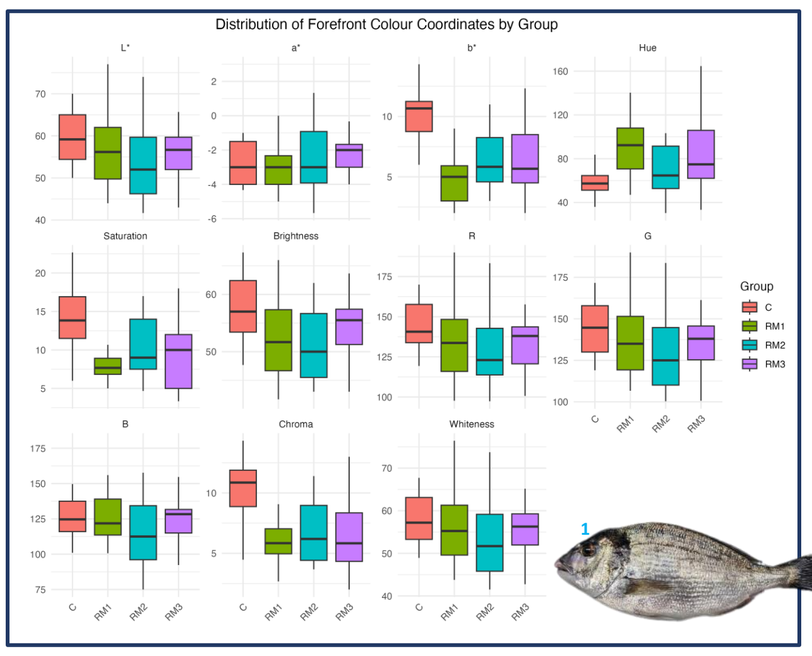
RBG Color space

- Red
- Blue
- Green

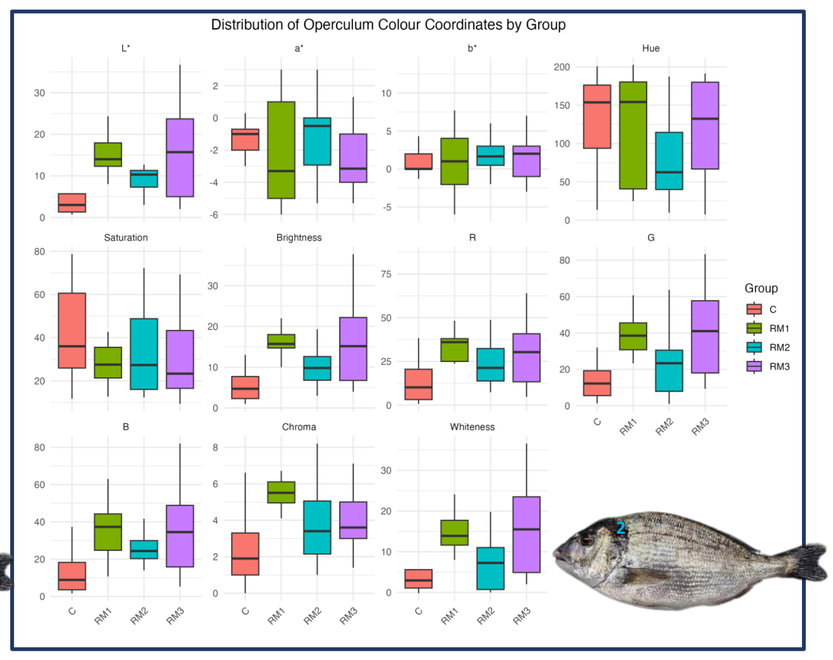
HSB (Alternative RBG Color space)

- Hue
- Saturation
- Brightness

Results - Fish



Average forefront b*, Chroma and Saturation of groups RM1, RM2 and RM3 were significantly decreased compared to the Control group.



Groups' RM1, RM2 and RM3 average R, G and B was significantly higher compared to the Control group.

- Spot 3 did not present any significant differences among the treatments.

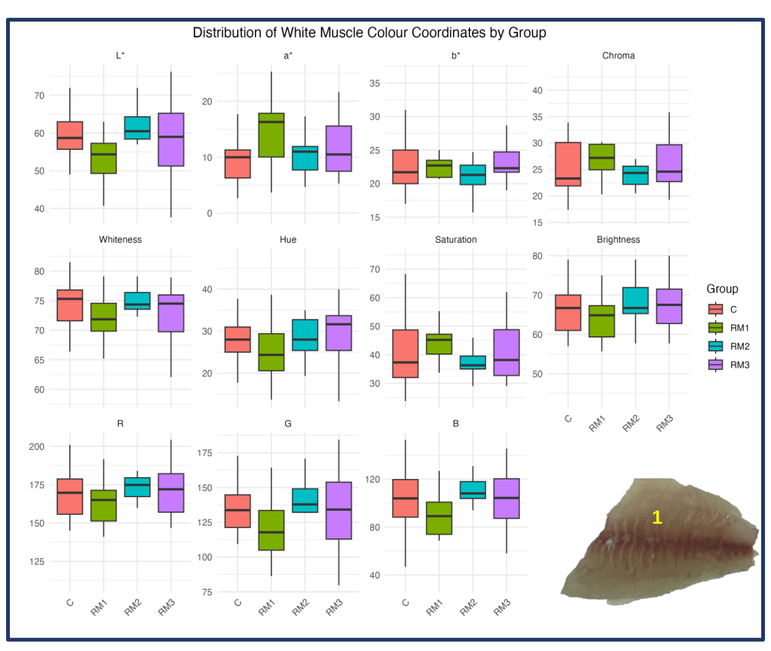
- The values of L* and Whiteness of the treatments RM2 and RM3 exhibited a statistically significant increase compared to the control group.

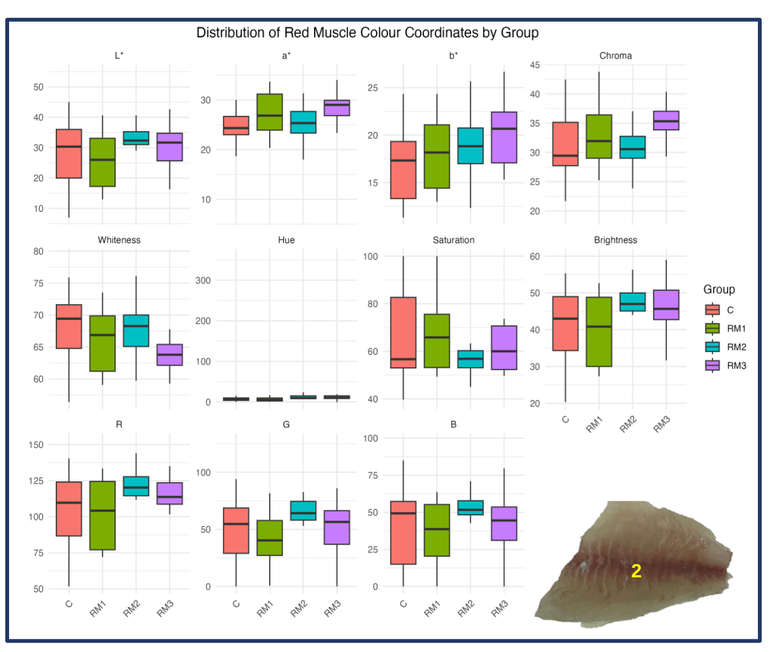
- All RM treatments exhibited a trend for lower RGB values compared to the control group.

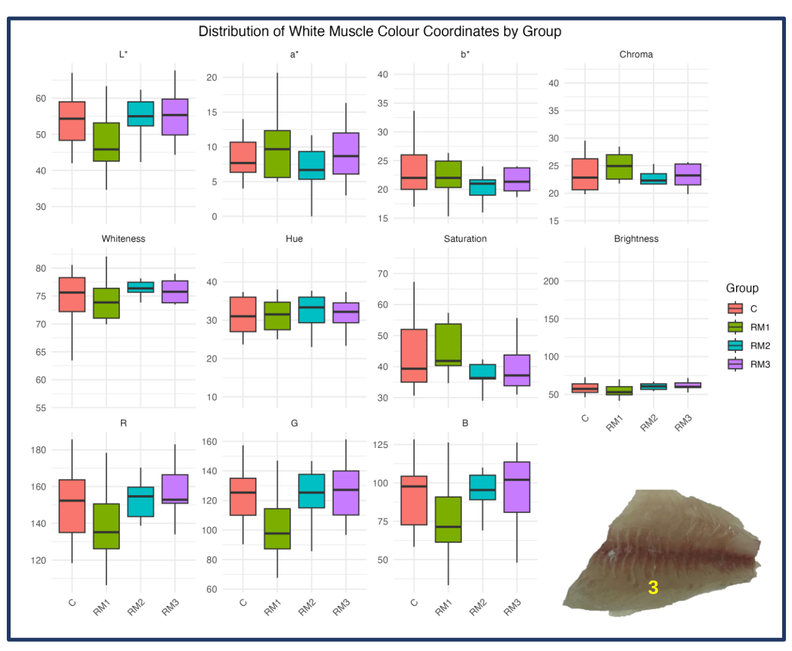
Results - Fillets

Fillet's white muscle exhibited a statistically significant difference only at spot 1. More specifically, b* (yellowness) was significantly higher at groups RM1 and RM3 compared to the control group.

Fillet's red muscle of group RM3 demonstrated significantly higher a* (redness) and Chroma values compared to the control group and significantly lower whiteness values.







Discussion

Consumers choose fish based on their visual appearance and they associate bright and intense colors with freshness, flavor, and higher quality (Pulcini et al., 2021).

The lower b* (yellowness) values of the RM treatments are not in accordance with previous studies where carotenoid-rich microalga *Chlorela vulgaris* enhanced the color of the forefront line (Gouveia et al., 2002). Meanwhile, lower Saturation and Chroma values also are not with accordance with previous studies, however, these results may be linked with the camera angle. Indeed, frontal view photos lead to a better recognition of the yellow pixels of the forefront line by CBI softwares (Pulcini et al.,2020). In fish with grey/silver pigmentation, such as sea bream and sea bass, the variable L* is of particular importance as it is correlated with the fish quality (Erdağ & Ayvaz, 2021).

Fish fed with *R. mucilaginosa* showed increased L* and Chroma values compared to the control group, confirming the presence of a color difference. As far as the fish fillet is concerned, the red muscle of group RM3 appeared significantly more red that the control group. White muscle of groups RM1 and RM3 appeared to be more yellow that the control group. It is known that through CBI, a comprehensive assessment of coloration can achieved (Pulcini et al., 2021).

The results of this study indicated that the inclusion of the red yeast *Rhodotoroula mucilaginosa* could influence the skin pigmentation of gilthead seabream. Results become more noticeable at inclusion levels above 2%. Changes in the flesh pigmentation were noticeable mainly only on the red muscle part of the fillet and at an inclusion level of 3%.

Acknowledgments

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