

Article **Potential Use of Image Analysis in Breeding Programs for Growth and Yield Traits in Meagre (***Argyrosomus regius***)**

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Abstract: In the present work, we studied ten new productive traits in meagre (Argyrosomus regius), comprising three related to the carcass (cNiT) and seven related to morphometric (mNiT) characteristics. We harnessed non-invasive technology (NiT) by means of the IMAFISH_ML software. This tool's potential was leveraged on an industrial scale, encompassing the evaluation of 612 fish from two distinct rearing systems (marine cages and indoor tanks) at the time of harvest. Each fish underwent digital photography for morphometric measurements, manual weighing, and was manually eviscerated and filleted to calculate the carcass and fillet yield. Subsequently, the principal genetic parameters were estimated. The heritabilities for the growth traits were moderate (0.34 and 0.39 for TL and BW), whilst those for the cNiT traits ranged from medium to low (0.32–0.27). For the mNiT, they demonstrated a medium to low range (0.15–0.37), whereas the carcass and fillet yield heritabilities were considered to be medium to high (0.32 and 0.31). Most of the genetic correlations between the growth, NiT, and yield traits were not estimated accurately due to the limited data. As was expected, we observed predominantly high and positive correlations between the growth and mNiT. A genetic correlation to highlight was the fillet yield with the fish maximum height (0.87 \pm 0.23) and with the head height (0.87 \pm 0.24). This suggests that indirect selection using NiT could improve the growth and yield traits. Employing a multi-trait selection approach enables us to capture a broader spectrum of genetic variability and to potentially identify individuals with superior genetic potential. The use of image analysis software ensures objective and precise measurements, thereby reducing the potential for human error or bias during the selection process. Further studies should be carried out to improve the accuracy of the estimates, especially those of the genetic correlations.

Keywords: meagre; carcass yield; fillet yield; IMAFISH_ML; heritability; genetic correlations

1. Introduction

In recent years, demand has been growing for the inclusion of new aquaculture species to increase the diversity of species and the sustainability of the Mediterranean aquaculture sector, in order to expand the market for aquaculture products [\[1\]](#page-8-0). Within this context, meagre (*Argyrosomus regius*) has become a species of great interest. This species is a carnivorous fish belonging to the family Scianidae and is one of the new species being used in Mediterranean aquaculture to diversify the sector [\[2\]](#page-8-1). The main producing countries in the Mediterranean area are Egypt, Turkey, Spain, and Greece, which accounted for 64.9%, 14.2%, 10.6%, and 6.5% of the total production in 2021, respectively [\[3\]](#page-8-2). One of the main characteristics of meagre is its fast growth rate (about 800–1000 g in 18 months) [\[4\]](#page-8-3), with a low feed conversion ratio of 0.9–1.2 [\[5\]](#page-8-4), a low fat content in fillet (about 1–3%), a good attractive shape as a whole fish product and it has high processing

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yield power [\[6\]](#page-9-0). Variables such as growth, feed efficiency, and product yield are very important for the meagre industry, as they add great value to both the production and the economic performance. The product quality is valued both by intermediaries, who oversee the processing and must ensure that the product does not lose quality, as well as by the end consumer [\[7\]](#page-9-1). Meagre production industries must consider these quality characteristics, in addition to those of production, in order to satisfy their own needs and also those of the end consumer.

New key enabling technologies (KETs) are increasingly used to monitor the rearing of aquaculture species, with the aim of controlling variables such as the growth rate and animal welfare, and morphological variables such as the shape, size, and weight. Researchers have developed various indirect methods for estimating the carcass and fillet yield in live animals. León-Bernabeu et al. [\[8\]](#page-9-2) used photographic image analysis to evaluate various morphological variables in gilthead seabream (*Sparus aurata*), to see how they affected other productive traits and also to optimize the sampling time in companies. Freitas et al. [\[9\]](#page-9-3) developed a computer vision system, in which the body shape of pacus (*Piaractus mesopotamicus*) was included as a breeding target. Kankainen et al. [\[10\]](#page-9-4) used ultrasonography to measure the thickness of the fat and muscle layers in the live animal, which could then be used to predict the carcass yield. Similarly, body measurements such as the weight, length, and girth have been used to estimate the fillet yield in fish [\[11](#page-9-5)[,12\]](#page-9-6).

In the case of meagre, it is still at an early stage in selective breeding programs. Little progress has been made, as it is a species which has only recently been included in selection programs that primarily focus on the growth performance. These strategies are already more consolidated in species such as salmon or gilthead seabream, where genetic improvement programs are more advanced [\[13\]](#page-9-7). The phenotypic and genetic variation in various product quality and yield traits have been estimated in salmon [\[14–](#page-9-8)[16\]](#page-9-9), Tilapia [\[17](#page-9-10)[,18\]](#page-9-11), sea bass [\[19\]](#page-9-12), and gilthead seabream [\[20–](#page-9-13)[22\]](#page-9-14). To the best of our knowledge, no work has been carried out on the genetic components related to other characteristics different from growth traits [\[6](#page-9-0)[,23\]](#page-9-15), which indicates a significant gap in the understanding of the genetic basis of the external morphology and yield traits in meagre.

The objective of this study was to estimate the phenotypic and genetic variability for the carcass and fillet yield, and for image analysis as a worthwhile tool to assess morphological traits. This approach is expected to incorporate meagre into a selective breeding program that encompasses not only productive factors but also quality attributes.

2. Materials and Methods

This study and analysis have been carried out within the framework of improving the competitiveness of the meagre sector through genetic selection (GENECOR); this Spanish project aims to optimize breeding programs from multiple fields, including the development and application of KETs.

2.1. Biological Material

The fish used in this study were obtained from one broodstock ($n = 9$; 4 males and 5 females), belonging to the company Alevines del Sureste S.L. (Cabo Cope, Águilas, Murcia, Spain) from the *Avramar* group. All broodstock were synchronized for egg release and embryo collection on four consecutive days (4DL model) to maximize family representation; 1.5–2.0 kg egg was incubated in cylindrical conical tanks (650 L). Water conditions were as follows: temperature 20.0 °C, salinity 36‰, and oxygen saturation was 100–120%. Thus, at 220 days post-hatching (dph), 612 offspring were individually tagged in the abdominal cavity for individual identification, with a Passive Integrated Transporter (PIT, Trovan Daimler-Benz, North Ferriby, UK), in a similar manner to that described by Vallecillos et al. [\[6\]](#page-9-0), and a sample of caudal fin was collected and preserved in absolute ethanol at room temperature for future DNA extraction. The fish were randomly separated and raised in two different systems (HS): 244 fish were raised in sea cages in Burriana (Castellón, southeast of Spain) and 368 fish in continental tanks in Águilas (Murcia, southeast of Spain), under different rearing conditions (Table [1\)](#page-2-0).

Table 1. Main features and physicochemical parameters for on-growing facilities. Feeding systems and water conditions.

2.2. Slaughtering, Image Capturing, and Sampling

The harvest was carried out at 549 dph. Fish were slaughtered by immersion in ice cold water (hypothermia) and stored at 4 °C for 24 h for further processing. Body weight (BW) and total length (TL) were measured according to the procedures described in Vallecillos et al. [\[6\]](#page-9-0). All the fish were photographed with a digital camera (Panasonic DMC-FZ200, Kadoma, Osaka, Japan) following the image capture protocol described by Navarro et al. [\[24\]](#page-9-16) for lateral images, in order to analyze 10 traits, i.e., non-invasive technology (NiT),
ecclentricity of the fishion of the including morphometric NiT (mNiT) and carcass NiT (cNiT) traits. Afterwards, visceral Incruding introprieties in the trip and direase in Fig. (EVIT) datas. Therwards, visceral fat deposits were manually removed, and all the fish were manually skinned and filleted. The weight of the carcass was obtained, as was that of both fillets, which were weighed together (fillet weight). Carcass yield (%) was determined as (carcass weight/BW) \times 100 and fillet yield (%) was determined as (fillet weight/carcass weight) \times 100.

2.3. Images Analysis

all images were analyzed using IMAFISH_ML software, developed in MatLab v.7.5 The Math-Works Inc., Natick, MA, USA) described in Navarro et al. [\[24\]](#page-9-16). After calibrating (The Math-Works Inc., Natick, MA, USA) described in Navarro et al. [24]. the camera to match the pixels to the actual distance, the image analysis first identifies the shape of the fish and then automatically performs a full set of 10 measurements (Table [2\)](#page-3-0). The images are also converted to grayscale to avoid the software from accidentally recognizing non-specific fish body areas such as shadows or fins.

Figure 1. Image from IMAFISH_ML software. Lateral view, for the determination of non‐invasive **Figure 1.** Image from IMAFISH_ML software. Lateral view, for the determination of non-invasive technological traits (NiT) of *Argyrosomus regius*: points X_1 , X_2 , X_3 , and X_4 of the horizontal axis will be used to determine longitudinal traits; Y_1 , Y_2 , Y_3 , Y_4 , and a, b, c, d, and e are dorsoventral axes, which will be used to determine height traits. which will be used to determine height traits.

Table 2. The traits measured by IMAFISH_ML software from lateral-side fish image based on the detected points depicted in Figure [1.](#page-2-1)

mNiT: morphometric non-invasive technological traits; cNiT: carcass non-invasive technological traits.

The importance of converting the images into grayscale must be stressed, as this step is critical for avoiding errors in the analysis. The images are carefully revised and modified if required, using image edition software to ensure accuracy (Adobe Photoshop CC. [2018], Berkeley, CA, USA, Peachpit Press).

2.4. Genotyping

The multiplex SMAr (Super Multiplex *Argyrosomuys regius*), was used as described in Vallecillos et al. [\[25\]](#page-9-17) for genetic characterization of the offspring and broodstock. The parental assignment was performed according to the criteria described by Vallecillos et al. [\[6\]](#page-9-0).

2.5. Statistical Data Analysis

Genetic parameters were estimated taking into account genealogies that had been previously obtained by microsatellite-based multiplex PCR panel as per Vallecillos et al. [\[25\]](#page-9-17), and with the same procedure used in Vallecillos et al. [\[6\]](#page-9-0) but changing the included covariate in the model depending on the trait to be analyzed.

Briefly, data were estimated using the following two-trait animal mixed model under a Bayesian approach solved with gibbs3f90 software (Ubuntu 13.10) [\[26\]](#page-9-18):

$$
y = X\beta + Zu + Wp + e
$$

where

y = vector of data;

 β = vector of systematic effects:

Housing system (HS) with two levels (cage or tank) for all traits, covariate body weight for all traits;

 $u =$ vector of additive genetic effects;

 $p =$ permanent environmental effect of family-HS;

e = residual;

X, *Z*, and *W* = incidence matrices relating data with systematic effects and random additive genetic and permanent effects, respectively.

Statistics were calculated using the R package "BOA" [\[27\]](#page-10-0). For further information about the distributions and processes see Vallecillos et al. [\[6\]](#page-9-0).

3. Results

3.1. Phenotyping

The effects of the housing system on the growth (BW and TL), on the yield (carcass and fillet yield), and on the morphometric traits are shown in Table [3.](#page-4-0)

Table 3. Phenotypic data for growth, morphometric non-invasive technological traits, and yield, for meagre at 549 days post-hatching, raised in two different housing systems.

BW (body weight), TL (total length), TLA (total lateral area), FilA (fillet area), FilAr (fillet area ratio), TLL (total lateral length), FilML (fillet maximum length), TaEL (tail excluded length), HeH (head height), FMH (fish maximum height), FEc (fish eccentricity) and HeEc (head eccentricity). Variables were adjusted to BW (mean weight of 998 g). b is the regression coefficient (units/gram).

The fish in the cage were 37.9% heavier than those in the tank, when the difference is calculated as a percentage with respect to the average of both HSs. There were no differences between the HSs for other traits when adjusted by BW. The BW had a meaningful effect on the morphometric traits, those related to the length, height, and especially to the area. Thus, for every 100 g weight increase, the TL increased by 1.39 cm, and the TLA and FilA increased by 19.3 and 12.4 cm2, respectively. The BW had no effect on the carcass yield, FilAr, FEc, and HeEc, highlighting that the yield and the shape remain the same throughout the slaughter BW range. Moreover, no significant HS effect was observed for the fillet yield (Figure [2\)](#page-5-0), FEc, and HeEc. However, the carcass yield for the tank fish was around 2% higher than for the cage fish.

3.2. Heritability and Correlations

The heritabilities and genetic and phenotypic correlations for the growth and analyzed NiT traits calculated using IMAFISH_ML from images. The area traits (TLA, FilA, and FilAr), length traits (TLL, FilML, and TaEL), height traits (HeH and FMH), and shape traits (FEc and HeEc) are shown in Table [4.](#page-6-0) The heritability values from the NiT traits were medium and only low for the EFc trait, and most of them showed high standard deviations. As a whole, the heritabilities for growth, carcass, and fillet yield were considered mediumhigh (0.34 and 0.39 for TL and BW; and 0.32 and 0.31 for carcass yield and fillet yield, respectively). The heritabilities for the area cNiT trait were medium (0.32 for TLA and

0.27 for both FilA and FilA%, respectively). The mNiT was medium-low, with a minimum *value for the FEc (0.15) and a maximum value for the HeH (0.37). However, these estimates value for the FEC (0.15) and a maximum value for the HeH (0.37). However, these estimates* lacked accuracy due to the limited amount of data.

Figure 2. Posterior distributions of the carcass and fillet yield estimate in cage (red) and tank (blue) generated from Bayesian models. generated from Bayesian models.

3.2. Heritability and Correlations studied, while some genetic correlations were less precise due to the reduced sample size and the limited number of families (see in Vallecillos et al. [\[6\]](#page-9-0)). As expected, the phenotypic correlations between the BW and TL were positive and high (0.94 \pm 0.00), and correlations between these traits (BW and TL) and the NiT traits that refer to the length, height, or area were also positive and high with values around 0.7. However, the correlations of the BW or TL with the carcass or fillet yield were low, and those with the fish shape (FEc and HeEc) and FilAr were almost null. With respect to the carcass and fillet yield, the correlation was low and negative (-0.29 ± 0.1), and between these traits and the NiT they were low or almost null. For the NiT traits, the area with respect to the length was mostly high and positive. However, the correlations between the FilAr and NiT traits showed null values, except for the HeH and HeEC in which negative correlations were observed; low for the HeH (−0.28 \pm 0.10) but higher for the HeEc (−0.71 \pm 0.04); and for the FiML they showed a positive correlation (0.40 \pm 0.08). Thus, when the FilAr increased, the HeH decreased and tended to obtain rounder heads. The same situation occurred with the FiML and HeEc in which a negative correlation (-0.27 ± 0.10) was observed, revealing the trend that rounder heads (lower values) were related to a longer fillet and this was related to a higher FilAr. heads (lower values) were related to a longer fillet and this was related to a higher FilAr.
As we have stated before, most of the genetic correlations were not estimated accu-In this study, the phenotypic correlations were accurately estimated for the traits

rately. Therefore, only the significant correlations will be discussed. The genetic correlation between the BW and TL was very high (0.96 \pm 0.15). The TLA and FilA showed a high positive genetic correlation with the length and height NiT, but not with the FilAr and fish shape NiT (HeEc and FEc), for which the genetic correlations were almost null. The fillet yield showed high genetic correlations with the TLA, FilA length, and height NiT, and the correlation with the FMH (0.87 \pm 0.23) and HeH (0.87 \pm 0.24) was particularly notable. Concerning the length and height NiT, high and positive genetic correlations were mostly observed (range from 0.62 to 0.99) between them, but not with the FilAr and fish shape (HeEc and FEc). However, it was observed that for the phenotypical correlation, the FilAr and HeEc showed a high negative genetic correlation (−0.87 \pm 0.25), confirming that a high FilAr was related to a rounder head. Furthermore, as in the phenotypical correlations, high FilAr was related to a rounder head. Furthermore, as in the phenotypical correlations, a positive genetic correlation was found for the FilAr and FilML and a negative genetic correlation was found between the FilML and HeEc, but in those cases the standard deviation was very high; more data are therefore needed to make a more accurate estimate.

Table 4. Heritabilities (in bold at diagonal, with standard deviation), genetic correlations (below the diagonal in italics, with standard deviation), and phenotypic correlations (above the diagonal, with standard deviation) for growth, carcass yield, fillet yield, and non-invasive technological (NiT) traits, in meagre at harvest size (549 dph).

BW: body weight (g); TL: total length (cm); TLA: total lateral area (cm²); FilA: fillet area (cm²); FilAr: fillet area ratio; TLL: total lateral length (cm); FilML: fillet maximum length (cm); TaEL: tail excluded length (cm); HeH: head height (cm); FMH: fish maximum height (cm); FEc: fish eccentricity; HeEc: head eccentricity.

4. Discussion

As the first step of this assay, we explored the impact of two different rearing conditions on the carcass quality traits in meagre. We only found significant differences for the body weight and carcass yield. The fish raised in cages exhibited a 1.46-fold higher weight compared to their tank-reared counterparts. This result is likely attributable to the higher population density in the tank, as it has been observed in other works [\[28,](#page-10-1)[29\]](#page-10-2). The phenotypical variability for the carcass and fillet yield was low. Despite this fact, the fish reared in tanks exhibited a higher carcass yield than those in cages, which was potentially linked to the improved access to feed, limited movements, or a lower stress level in the tank environment. For the morphometric traits, no effect was observed when these variables were adjusted to a constant weight. A detailed discussion of the growth traits (BW and TL) in both rearing systems can be found in Vallecillos et al. [\[6\]](#page-9-0). Elalfy et al. [\[30\]](#page-10-3) identified weight disparities; this is similar to the findings of our research. For the yield, similar results have been observed in related studies. For instance, Vandeputte et al. [\[31\]](#page-10-4) compared five wild sea bass populations, noting similar carcass and fillet yields among them. In a study by García-Celdrán et al. [\[32\]](#page-10-5), the fillet yield in three populations of gilthead seabream demonstrated consistency at approximately 36% across all the populations. Furthermore, image analysis techniques, including the use of the IMAFISH_ML software, have been employed in studies focusing on estimating the quality traits, particularly in gilthead seabream $[8,30]$ $[8,30]$, in which no differences between the populations studied have been observed.

The importance of measuring easily quantifiable traits, such as the body weight and length, lies in their role in assessing fish performance and guiding breeding decisions [\[33\]](#page-10-6). The genetic parameters for the growth traits (BW and TL) showed a medium heritability with a high phenotypic and genetic correlation between them, in accordance with Nousias et al. [\[23\]](#page-9-15) and Vallecillos et al. [\[6\]](#page-9-0). A similar situation has been seen in other teleost species, such as gilthead seabream, in which the genetic parameters for growth traits have shown medium heritability from 0.20 to 0.37 [\[8](#page-9-2)[,30](#page-10-3)[,31](#page-10-4)[,34\]](#page-10-7).

However, some traits, including the carcass and fillet yield, pose challenges in their measurement, and are often overlooked in breeding programs [\[11\]](#page-9-5). The estimated heritabilities were medium for both the carcass and the fillet yield. Similar results were observed by Vandeputte et al. [\[31\]](#page-10-4) in sea bass (0.48 for carcass yield and 0.38 for fillet yield), and by Franslin et al. [\[11\]](#page-9-5) in different species for the fillet yield (sea bass 0.41, gilthead seabream 0.32 in cage and 0.42 in tank, and rainbow trout 0.34). García-Celdrán et al. [\[32\]](#page-10-5) reported low heritabilities (0.11) for the fillet yield in gilthead seabream, and Prchal et al. [\[12\]](#page-9-6) showed a high heritability for the carcass yield (0.48) and fillet yield (0.63) in common carp. Positive medium genetic correlations were shown between the BW-carcass and BW-fillet yield, albeit with low consistency due to the very high standard deviation. In seabream, García-Celdrán et al. [\[32\]](#page-10-5) observed an almost null genetic correlation for growth traits (BW and TL) with the carcass yield, and a negative genetic correlation with fillet yield $(-0.58 \pm 0.09$ BWfillet yield and -0.59 ± 0.50 for length-fillet yield), although the latter correlation was not estimated accurately. In common carp, Prchal et al. [\[12\]](#page-9-6) also showed a negative genetic correlation for performance traits with respect to body weight $(-0.15 \pm 0.15$ for carcass yield and -0.29 ± 0.13 for fillet yield). Researchers have explored indirect and genomic selection methods to tackle the complexities of improving the carcass yield and fillet yield, particularly in species such as rainbow trout [\[35\]](#page-10-8). However, the fillet yield is not clearly associated with the BW or length, and direct selection remains the preferred approach for its improvement in various fish species [\[11\]](#page-9-5). Notably, advancements in technology, including computer vision [\[8,](#page-9-2)[36](#page-10-9)[,37\]](#page-10-10) and machine learning [\[38\]](#page-10-11), offer the potential for the non-invasive and automated measurement of morphometric traits, saving time and minimizing measurement errors, which is essential for large-scale breeding programs. This approach empowers fish breeders to make more informed decisions about selecting future fish to breed without relying solely on offspring characteristics [\[30\]](#page-10-3). In this study, the IMAFISH_ML software automatically generated a total of 6120 morphometric measurements in just a few hours,

covering 10 different technological traits from 612 fish. In general, the morphometric traits showed medium heritabilities and positive genetic correlations with the growth traits, although these correlations were not estimated accurately. The genetic correlations of some morphometric measurements (TLA, FilA, FMH, and HeH) with the fillet yield were particularly interesting and indicate the potential for using the easily recorded FMH as a selection criterion to improve the fillet yield of meagre. Similar results were found in common carp [\[12\]](#page-9-6) and in gilthead seabream [\[8\]](#page-9-2), with a high heritability and genetic correlation for the growth and morphometric traits. Therefore, image analysis has been shown to save time and be an accurate tool to be included in a breeding program for growth improvement. Due to the limited amount of data available, extensive analysis with more data should be continued for the yield traits in order to obtain more accurate estimates.

5. Conclusions

Non-invasive technological (NiT) traits from IMAFISH_ML image analysis have been revealed as a potential tool in meagre breeding programs, defining promising candidates for genetic selection, since these traits showed medium-high heritabilities and strong genetic correlations with growth traits. Indirect selection for the fillet yield could also be carried out through these non-invasive measurements, especially for the fillet maximum height. However, further studies should be supported to obtain more accurate estimates, especially when focusing on indirect selection.

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