



REDUNDANCY IN MORPHOLOGICAL COVARIATION PATTERNS BETWEEN *Colossoma macropomum* AND *Piaractus orinoquensis*

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AUTHORS' CONTRIBUTIONS

This work was carried out in collaboration among all authors. Authors DVR and MMP designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors YPP and SVR managed the analyses of the study. Authors ZGC, EDB, WRC, YVC and BDSC managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Currently, great importance has been given to the study of external morphology, especially in fish, when it is used as a means of identifying hybrids. However, in fish morphometry, multicollinearity, which refers to redundant information, that is, overlapping covariates (distances between homologous points) is a problem generally present in morphological covariation patterns. In order to have an overview of the existing collinearity diagnostic measures together with critical values commonly used to evaluate existence of collinearity among the covariates, this research considers the study of multicollinearity in the truss protocol on 92 specimens of *Colossoma macropomum* and *Piaractus orinoquensis*. The diagnostic measures used were those included in the R mctest package; classified into general and individual measures. Multicollinearity (redundant measurements

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in the "truss protocol") was detected in morphological covariation patterns between *C. macropomum* and *P. orinoquensis* specimens. Only distances associated with morphological covariation patterns that make a difference in the head area, in the area of the bases of the fins of the abdomen and in the anterior part of the fish, did not constitute redundant morphological information in the morphological covariation patterns of this species. The use of *VIF* is recommended as a measure to identify redundant information associated with the external morphology of *C. macropomum* and *P. orinoquensis*.

Keywords: Morphometry; truss protocol; fishes; redundant information; *VIF*.

1. REDUNDANCY IN FISH MORPHOMETRY

1.1 Introduction

The implementation of morphometric analysis in some species provides scientific knowledge that helps genetic improvement. Currently, the techniques that involve the use of data on the external morphology of individuals are not widely used, since phenotypic plasticity means that the evaluated characters cannot always be generalized for the identification and differentiation of species and hybrids [1,2]. It is clear that the morphological characters are physical evidence of the expression of the genotype. Therefore, the differences between specific body characteristics can become very important to establish patterns of differentiation and inheritance [3,4]. In continental fish, the morphometric characteristics referring to the anatomical shape have been used to evaluate the productive response in rearing both in natural environments and in captivity. Currently, there are more modern and precise morphometric analysis techniques, such as Geometric Morphometry [5,6,7,8], which together with multivariate statistical analysis and means of direct visualization, constitute one of the most useful tools to describe the biological form and its changes.

Generally, these techniques are based on a set of measured distances between identifiable points on the organisms. In most cases, the measurements (distances between homologous points) present a high correlation, which in the context of statistics, and especially of the models that are used to compare between species, this linear dependency relationship is called multicollinearity. In fish morphometry, multicollinearity can refer to redundant information, that is, overlapping covariates (distances between homologous points). This implies that multicollinearity can have serious consequences on models. Based on some theoretical considerations, the need to detect multicollinearity between covariates arises [9,10]. In this sense, this work presents an overview of the existing collinearity diagnostic measures together with the critical values commonly used to judge the existence of collinearity between the covariates considering the use of the truss protocol on

Colossoma macropomum and *Piaractus orinoquensis* specimens. These diagnostic measures are being implemented in R with the proposed *mctest* package [11].

2. MATERIALS AND METHODS

2.1 Diagnosis of Multicollinearity in Morphological Covariation Patterns between *C. macropomum* and *P. orinoquensis*

In this study, 46 specimens of *C. macropomum* (see Fig. 1) and 46 specimens of *P. orinoquensis* (see Fig. 2) with an average weight of 600g, respectively, from artificial ponds of a fish farm in Portuguesa state, Venezuela, were analyzed. The method "Truss protocol" or "trusses" Strauss and Bookstein [12] was used, which achieves an exhaustive reconstruction of the shape from the distances between the homologous anatomical landmarks (see Table 1 and Fig. 3). The distances connecting these landmarks form a series of continuous quadrilaterals with their respective internal diagonals (see Fig. 3), which allows detecting differences in shape in the vertical, horizontal, and oblique directions. The limitations in this study is the number of measures necessary to achieve better efficiency in estimating parameters related to the morphology of these species.

The morphological patterns were studied using collinearity diagnostic measures commonly used and implemented in R with the *mctest* package proposed by Imdadullah et al., [11], among them; overall and individual diagnostic measures of multicollinearity.

2.2 Overall Collinearity Diagnostic Measures

2.2.1 Determinant

The matrix $X'X$ will be singular if it contains linearly dependent columns or rows. Therefore, determinant of normalized correlation matrix $R = X'X$ without intercept can be used to indicate existence of collinearity among regressors. However, determinant does not provide information about interdependence among regressors, it only provides information about

singularity (departure from orthogonality) of a correlation matrix. The determinant of $X'X$ on the scale is $0 \leq |X'X| \leq 1$ [13]. If $|X'X| \sim 0$, then collinearity exists among regressors [14].



Fig. 1. Cachama specimen (*Colossoma macropomum*)



Fig. 2. Cachama blanca specimen (*Piaractus orinoquensis*)

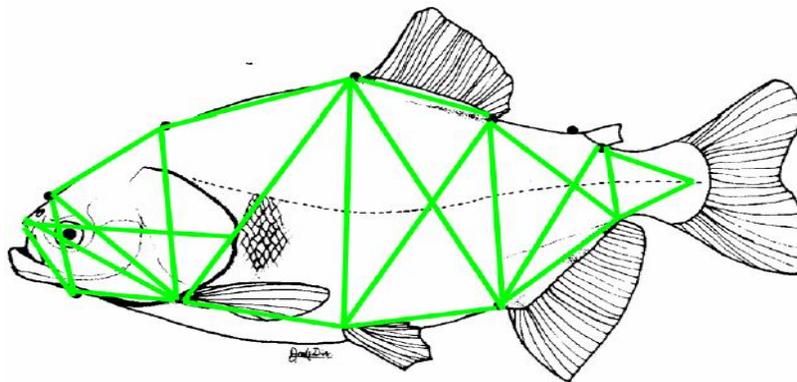


Fig. 3. Location of homologous points and distances measured on the left lateral profile of *C. macropomum* and *P. orinoquensis*

Table 1. Truss measurements from *C. macropomum* and *P. orinoquensis* specimens

Tip of snout to end of epiphyseal sulcus
Tip of snout to insertion of pectoral fin
Anterior edge of the epiphyseal sulcus to the end of the epiphyseal sulcus
Anterior edge of the epiphyseal sulcus at the insertion of the pectoral fin
Anterior edge of the epiphyseal sulcus when articulating
Articulate to insertion of pectoral fin
Posterior edge of epiphyseal sulcus to end of dorsal fin
Posterior edge of the epiphyseal sulcus at the insertion of the pelvic fin
Posterior edge of the epiphyseal sulcus to the insertion of the pectoral fin
Posterior edge of the epiphyseal groove when articulating
Insertion of pectoral fin to insertion of pelvic fin
Dorsal fin base
Anterior edge of dorsal fin to anterior edge of anal fin
Anterior edge of dorsal fin to insertion of pelvic fin
Anterior edge of dorsal fin to insertion of pectoral fin
Insertion of pelvic fin to end of anal fin
Posterior edge of dorsal fin to the fatty fin
Posterior edge of dorsal fin to posterior edge of anal fin
Posterior edge of dorsal fin to anterior edge of anal fin
Posterior edge of dorsal fin to insertion of pelvic fin
Anal fin base
Posterior edge of the fatty fin to the last scale of the lateral line
Posterior edge of fatty fin to posterior edge of anal fin
Posterior edge of the fatty fin to the anterior border of the anal fin
Posterior edge of the fatty fin to the anterior border of the anal fin
Eye diameter
Head length
Fat fin base

2.2.2 R-squared

Coefficient of determination (R^2) from regression of all x on y . The R^2 is a monotonic nondecreasing function of number of regressors included in the model, that is, R^2 indicates how well the regression fits the data [15]. On the other hand, higher the R^2 values, the more chances of regressors to be plagued with multicollinearity, since R^2 is affected by regressors sharing their variances [14].

2.2.3 Farrar χ^2

It is the Chi-square test for detecting the strength of collinearity over the complete set of regressors.

$$\chi^2 = - \left[n - 1 - \frac{1}{6(2p+5)} \right] \times \log_e [X'X] \sim \psi_{\frac{1}{2}p(p-1)}^2.$$

Collinearity exists among regressors if $\chi^2 > \chi_{\frac{1}{2}p(p-1)}^2$ [16].

2.2.4 Condition index

$$CI_j = \sqrt{\frac{\max(\lambda_j)}{\lambda_j}} \quad j = 1, 2, \dots, p; \lambda_1 \geq \lambda_2 \geq \dots \geq \lambda_p.$$

Collinearity exists if any of $CI_j > 10, 15$, or 30 [9,17].

2.2.5 Sum of reciprocal of eigenvalues

In an orthogonal system $\sum_{j=1}^p \frac{1}{\lambda_j} = p$, therefore, for a sample based correlation matrix R with eigenvalues λ_j , comparing p with $\sum_{j=1}^p \frac{1}{\lambda_j}$ can be used to indicate collinearity. If $\sum_{j=1}^p \frac{1}{\lambda_j}$ is (say) five times larger than the number of regressors used in the model then collinearity exists among regressors [18,19].

2.2.6 Theil's indicator

Theil [20] proposed a measure of collinearity based on an incremental contribution ($R^2 - R_j^2$) to the squared multiple correlation, where R_j^2 is the R^2 from auxiliary regression of regressors.

$$m = R^2 - \sum_{i=1}^p (R^2 - R_{-i}^2).$$

If $m = 0$ then all X 's are mutually uncorrelated (no redundancy exists) as the incremental contribution all

add up to R^2 . However, if $m \sim 1$ then Collinearity exists among regressors.

2.2.7 Red indicator

Kovács et al. [21] presented a synthetic and new normalized indicator for diagnostic of collinearity by using eigenvalues or quantifying the average correlation of the data.

$$Red = \frac{\sqrt{\sum_{j=1}^p (\lambda_j - 1)^2}}{p \sqrt{p - 1}}$$

If value of the Red indicator is zero ($Red = 0$) then it indicates the absence of redundancy and value near to 1 ($Red \sim 1$) indicates maximum redundancy.

2.3 Individual Collinearity Diagnostic Measures

2.3.1 Klein’s rule

If R_j from the auxiliary regression is greater than the overall R^2 (obtained from the regression of y on all the regressors) then multicollinearity may be troublesome. The decision rule for detection of collinearity is, $R_{x_j, x_1, x_2, \dots, x_p}^2 > R_{y, x_1, x_2, \dots, x_p}^2$ [22].

2.3.2 VIF and Tol

VIF measures how much variances of the estimated regression coefficients are increased over the case of no correlation among p regressors. The diagonal elements of $(X'X)^{-1}$ matrix are considered as very important in detecting multicollinearity.

$$VIF_j = (X'X)^{-1}_{jj} = \frac{1}{1 - R_j^2} \text{ and } Tol_j = \frac{1}{VIF_j} = 1 - R_j^2.$$

The criticism on VIF is that $var(\hat{\beta}_j) = \frac{\sigma^2}{\sum x_j^2} VIF$ depends on σ^2 , $\sum x_j^2$ and VIF, which shows that a high VIF can be counterbalanced by a low σ^2 or high $\sum x_j^2$. So a high VIF is neither a necessary nor a sufficient measure of multicollinearity. The value of $VIF > 3, 5, 10$ or value of $Tol \sim 0$ indicates existence of collinearity among regressors [23].

2.3.3 Eigenvalues

Kendall [24] and Silvey [25] suggested the use of eigenvalues of $X'X$ (correlation matrix) to check the presence of multicollinearity and set the criteria that small eigenvalues (near to zero) are indication of high

collinearity, however, they did not mentioned how much small it should be. One or more smaller eigenvalues of $X'X$ or its related correlation matrix indicate collinearity.

2.3.4 CVIF

Curto and Pinto [26] proposed new measure of multicollinearity to evaluate the impact of the correlation among regressors in the variance of the OLSEs.

$$CVIF_j = VIF_j \times \frac{1 - R^2}{1 - R_0^2}$$

where, $R_0^2 = R_{yx_1}^2 + R_{yx_2}^2 + \dots + R_{yx_p}^2$. Collinearity exists if $CVIF_j \geq 10$.

2.3.5 Leamer’s method

Leamer (in Green [10]) suggested a measure of the effect of multicollinearity for the j th variable;

$$C_j = \left\{ \frac{\left(\sum_{i=1}^n (X_{ij} - \bar{X}_j)^2 \right)^{-1}}{(X'X)^{-1}_{jj}} \right\}^{\frac{1}{2}}$$

This measure is the square root of the ratio of variances of estimated coefficients ($\hat{\beta}_j$) when estimated without and with the other regressors. If X_j is uncorrelated with the other regressors C_j would be 1 otherwise will be equal to $(1 - R_j^2)^{\frac{1}{2}}$, i.e., $C_j \sim 0$ indicates existence of collinearity among regressors.

2.3.6 F and R² relation

The relationship of F-test and R^2 from regressing X_j on the other remaining regressors can be used to detect multicollinearity. The relationship is described as:

$$F_j = \frac{\frac{R_{x_j, x_1, \dots, x_p}^2}{p - 2}}{\frac{1 - R_{x_j, x_1, \dots, x_p}^2}{n - p + 1}} \sim F_{(p-2, n-p+1)},$$

where $F^* = F_{p-2, n-p+1}$. If $F_j > F^*$, then it means that the regressor X_j is collinear with other regressors and it should be dropped from the model [27].

2.6.7 Farrar w

It is an F -test for locating the regressors which are collinear with others and it makes use of multiple correlation coefficients among regressors.

$$w_j = \frac{R_j^2}{1 - R_j^2} \left(\frac{n - p}{p - 1} \right) \sim F_{(n-p, p-1)}$$

If $w_j > F_{(n-p, p-1)}$, there is indication of considerable collinearity [16].

Most of the overall and individual measures to detect multicollinearity described above are included in the R mctest package, which mainly implements functions for detecting multicollinearity between covariates using the omcdiag () functions in the case of general measures and imcdiag () for individual measurements [11].

3. RESULTS

In Table 2, most of the diagnostic measures, except the Theil indicator, suggest that there are characteristics associated with the morphological covariation patterns between specimens of *C. macropomum* and *P. orinoquensis* redundant, that is, there is multicollinearity, which can contribute to the entropy of the models used to identify patterns of morphological covariation of these species. In Table

3, the VIFs suggest that most of the distances measured on the lateral profile of these specimens are attributed to redundant morphological characteristics. Only morphological characteristics such as; posterior edge of epiphyseal groove to pectoral fin insertion variable, anterior edge of dorsal fin to anterior edge of anal fin, anterior edge of dorsal fin to pectoral fin insertion, pelvic fin insertion to anterior edge of the anal fin, posterior border of the dorsal fin to the anterior border of the anal fin, posterior border of the adipose fin to the last scale of the lateral line and base of the adipose fin do not constitute redundant morphological information, that is, they are not causing multicollinearity (see Fig. 4). These covariates are associated with morphological covariation patterns that make a difference in the area of the head, in the area of the bases of the abdomen fins and in the anterior part of the fish. The results of the Farra-Glauber test (individual diagnostic measure of multicollinearity) did not show a good performance in relation to the identification of the origin of multicollinearity, since it was not able to identify non-redundant covariates associated with the morphology of the specimens. of *C. macropomum* and *P. orinoquensis*.

Table 2. Overall collinearity diagnosis in patterns of morphological covariance between *C. macropomum* and *P. orinoquensis*

Index or test	Collinearity diagnosis
Determinant	*
Farrar-Glauber	*
Red indicator	*
Sum of lambdas	*
Theil indicator	NS
Condition number	*

* (Collinearity identified); NS (unidentified collinearity)

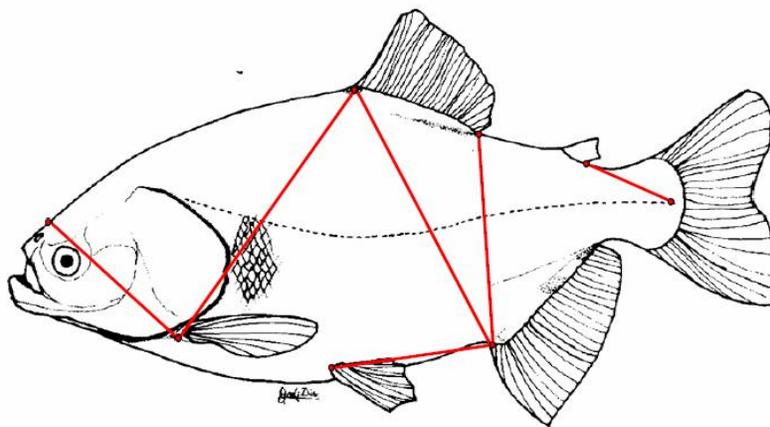


Fig. 4. Covariates (distances) not redundant in the “truss protocol” on *C. macropomum* and *P. orinoquensis*

Table 3. Individual collinearity diagnosis in patterns of morphological covariance between *C. macropomum* and *P. orinoquensis*

Landmarks	VIF	Fi
Tip of snout to end of epiphyseal sulcus	*	*
Tip of snout to insertion of pectoral fin	*	*
Anterior edge of the epiphyseal sulcus to the end of the epiphyseal sulcus	*	*
Anterior edge of the epiphyseal sulcus at the insertion of the pectoral fin	*	*
Anterior edge of the epiphyseal sulcus when articulating	*	*
Articulate to insertion of pectoral fin	*	*
Posterior edge of epiphyseal sulcus to end of dorsal fin	*	*
Posterior edge of the epiphyseal sulcus at the insertion of the pelvic fin	*	*
Posterior edge of the epiphyseal sulcus to the insertion of the pectoral fin	NS	*
Posterior edge of the epiphyseal groove when articulating	*	*
Insertion of pectoral fin to insertion of pelvic fin	*	*
Dorsal fin base	*	*
Anterior edge of dorsal fin to anterior edge of anal fin	NS	*
Anterior edge of dorsal fin to insertion of pelvic fin	*	*
Anterior edge of dorsal fin to insertion of pectoral fin	NS	*
Insertion of pelvic fin to end of anal fin	NS	*
Posterior edge of dorsal fin to the fatty fin	*	*
Posterior edge of dorsal fin to posterior edge of anal fin	*	*
Posterior edge of dorsal fin to anterior edge of anal fin	NS	*
Posterior edge of dorsal fin to insertion of pelvic fin	*	*
Anal fin base	*	*
Posterior edge of the fatty fin to the last scale of the lateral line	NS	*
Posterior edge of fatty fin to posterior edge of anal fin	*	*
Posterior edge of the fatty fin to the anterior border of the anal fin	*	*
Posterior edge of the fatty fin to the anterior border of the anal fin	*	*
Eye diameter	*	*
Head length	*	*
Fat fin base	NS	*

* (Collinearity identified); ns (unidentified collinearity)

4. DISCUSSION

The *VIF* as a measure of collinearity is not without criticism. As is known, the $var(\hat{\beta}_j)$ depends on three factors: σ^2 , $\sum x_j^2$ and VIF_j . A high *VIF* is offset by a low σ^2 baja or a high $\sum x_j^2$. Otherwise: a high *VIF* is neither a necessary nor a sufficient condition to obtain high variances and standard errors. Consequently, high multicollinearity, as measured by a high *VIF*, may not necessarily lead to high standard errors. Throughout this analysis, the terms high and low are relative [27]. Given the importance of the *VIF* as indicated in the first instance by Mandell [28], who shows that the standard error of the *j*-th regression coefficient can be expressed as the product of the residual standard error of the regression by the variance inflation factor (*VIF*), widely used to detect multicollinearity; In particular, it shows that the *VIF* is severely affected by the smallest eigenvalues of the *R* matrix, and Glantz and Slinker [29], who indicate that this coefficient measures the increase that occurs in the variance of b_j with respect to the minimum

value that is would be achieved in the total absence of collinearity of the corresponding covariate X_j with respect to the remaining covariates, that is why the *VIF* should be considered as a diagnostic measure of collinearity, fundamentally as an individual measure, especially when the objective is to identify the origin of multicollinearity.

Regarding the individual diagnosis of multicollinearity in the morphological covariation patterns presented in Table 3, these results show, in the first place, the advantages of *VIF* compared to the other multicollinearity measures as referred by Belsley [30], Gujarati and Porter [27] and Ramirez et al., [31] also verify the criticisms made by some authors regarding the Farrar and Glauber test, who point out the ineffectiveness of this procedure, especially when identifying the origin of multicollinearity. On the other hand, the results of the morphological covariation patterns between *C. macropomum* and *P. orinoquensis* coincide with those reported by Pineda et al., [32] who used principal component analysis for the morphometric comparison

between males and females of *C. macropomum* maintained in ponds, and those reported by Villegas et al., [33] in a multivariate analysis that allowed a morphometric comparison of a hybrid originated from *C. macropomum* and *P. orinoquensis*. The foregoing reveals what was indicated by Porras-Rivera and Rodríguez-Pulido [34] and Conte-Grand et al., (2015), who point out that external morphology is not always reliable when used as the only means of identification, particularly for hybrid individuals beyond the first generation.

5. CONCLUSIONS

Multicollinearity (redundant truss protocol measures) was evidenced associated with morphological covariation patterns between *C. macropomum* and *P. orinoquensis* specimens, which contribute to the entropy of the models used to study the morphology of these species. Much of the distances measured on the lateral profile of these specimens is attributed to redundant morphological characteristics. Only the distances associated with patterns of morphological covariation that make a difference in the area of the head, in the area of the bases of the abdomen fins and in the anterior part of the fish, do not constitute redundant morphological information in the morphological covariation patterns between *C. macropomum* and *P. orinoquensis*. Finally, based on the diagnosis of multicollinearity in the morphological covariation patterns of this species, the use of *VIF* is recommended as a measure to identify the origin of multicollinearity, namely, redundant information associated with the external morphology of these specimens compared to the other multicollinearity measures, which should be considered given the importance given to the study of external morphology, especially in fish, when it is used as a means of identifying hybrids. However, further measurements need to do for a more exhaustive study of the morphological covariation patterns between these species.

ETHICAL APPROVAL

As per international standard or university standard ethical approval has been collected and preserved by the authors.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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APENDIX 1

R code for the overall and individual diagnosis of general multicollinearity in morphological covariation patterns between *C. macropomun* and *P. orinoquensis* specimens.

```
> library('mctest')
> x <- Data.morfometria[ , -1]
> y <- Data.morfometria[ , 1]
> omcdiag(x, y, detr = 0.001, red = 0.6, conf = 0.99, theil = 0.6, cn = 15)
> omcdiag(x, y, Inter = FALSE)
> omcdiag(x, y)
> imcdiag(x, y, corr = TRUE)
> imcdiag(x, y)
```

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