MORPHOMETRIC AND MERISTIC VARIATIONS BETWEEN POPULATIONS OF WHITE-SPOTTED RABBIT FISH, *Siganus sutor* **(VALENCIENNES, 1835) FROM THE KENYAN COAST**

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DECLARATIONS

By the candidate:

I hereby declare that this dissertation has not been presented in any other institution and is my own original work.

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DEDICATIONS

I dedicate this Bachelors dissertation to my late father, Mr. Raphael Mutua Nzioka and to all my family, all of whom believed in diligence, science, art and the pursuit of academic excellence.

ABSTRACT

Morphometric and meristic variations were used to investigate the population structure of the White-spotted rabbit fish, *Siganus sutor* (Valenciennes, 1835) in order to determine what degree of isolation may result in notably phenotypic and genetic differentiation among its populations. Samples of the species were collected along the Kenyan coast from Malindi, Mombasa and Shimoni. A total of 31 morphometric measurements were taken from each of the 98 fish collected. 6 meristic characters were also counted in the specimens. All morphometric analysis was log-transformed and linear regression models used to estimate allometric coefficients of growth. The morphometric measurements were standardized to mean total length using an allometric formula and multivariate statistics used to analyse both morphometric and meristic characters. Principal component analysis revealed good discrimination between populations of the White-spotted rabbit fish from Shimoni, Mombasa and Malindi. Shimoni and Mombasa populations were different from one another while those from Malindi shared characteristics with both the Shimoni and Mombasa populations. Observed morphometric differences came from the ventral fin lengths and caudal peduncle widths for the Shimoni populations and mouth width, post-orbital length, head diameter and lower jaw width for Mombasa populations. From the results, discrimination observed can be attributed to diversity of environmental conditions, habitat characteristics and possibly food and feeding habits. Overall, morphometric characters were more useful than meristics in differentiating the three populations.

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LIST OF ABBREVIATIONS AND ACRONYMS

- BMU Beach Management Unit
- BTS Bartlett's Test of Sphericity
- DFA Discriminant Function Analysis
- DNA Deoxyribonucleic acid
- EBFM Ecosystem Based Fisheries Management
- KMFRI Kenya Marine and Fisheries Research Institute
- KMO Kaiser-Meyer-Olkin measure
- MNHN Museum National d'Histoire Naturelle
- MSA Measure of Sampling Adequacy
- NEM Northeast Monsoon
- NMK National Museums of Kenya
- PAST Paleontological Statistics Software Package
- PC Principal Component
- PCA Principal Component Analysis
- SEM Southeast Monsoon
- SPSS Statistical Package for Superior Scientist
- WIO Western Indian Ocean

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CHAPTER 1

1.0 INTRODUCTION

1.1 Background

The understanding of stock structure is vital and fundamental in fisheries management, especially where multiple stocks are exploited differently in terms of fishing effort and distribution of mortality (Ricker, 1981; cited in Begg and Waldman, 1999). When identifying and discriminating fishery stocks, the most effective approach considers various disciplines to gain multiple perspectives on patterns and processes in stock separation (Cadrin, *et al.*, 2005). Each discipline offers a unique view on stock structure relating to different definitions of the term "stock" (Templeman, 1983; Begg and Waldman, 1999).

Stocks have been referred to in relation to fisheries science as any group of fish species that are available for exploitation in a given area (Milton and Shaklee, 1987; cited in Begg and Waldman, 1999). Generally, though the term stock can be defined as a group of individuals that sustains itself over time, precise definitions vary with discipline (Booke, 1981; cited in Begg and Waldman, 1999). Hilborne and Walters (1992; cited in Begg and Waldman, 1999) define stock as arbitrary groups of fish large enough to be essentially self-reproducing, with members of each group having similar life history characteristics.

Inter-population variability within a stock has a theoretical and practical significance for determination of species plasticity and its ability to adapt to environmental changes. According to Begg and Waldman (1999), a lot of information is needed on the exploited species in order for identification of the stock to be compatible with the expected aims of its management. Several methods have been used in stock discrimination and include tagging and migration (mark-recapture), catch data, life history characteristics, parasites as indicators of fish stock, otolith microchemistry, morphology (meristics, morphometrics, scales and otolith analyses), genetics (protein variation, mitochondrial DNA, nuclear DNA) as well as serological and biochemical methods (Templeman, 1983; Begg and Waldman, 1999). Among these, morphology (morphometrics and meristics) have been widely used as a very important tool in identification of stocks (Murta, 2000; Abaunza, 2001; Albertson and Kocher, 2001; Armstrong and Cadrin, 2001; Aguilla-Perrera, 2004; Poulet *et al.*, 2004; Turan, 2004; Becktas and Belduz, 2009). Meristics are countable morphological structures (e.g. fin rays, etc) that have historically served as important basis for identifying fish stocks (Begg and Waldman, 1999). Count data is normally discrete allowing for statistical analyses. Since these characters are normally set early in life, they are controlled by both genetic and environmental factors, and remain stable throughout life; thus reflecting environmental influence over a relatively brief period of larval development (Begg and Waldman, 1999). Morphometric variations on the other hand can be used to discriminate "phenotypic stocks," defined as groups with similar growth, mortality and reproductive rates (Cadrin, 2000). Cadrin (2000) further suggests that variability in growth, development and maturation creates a variety of body shapes within a species as well as obscuring, or subsequent displacement by development of some ontogenic features persisting as a record of an individual's life history.

Such population groups may just be demes characterized by members having similarities which are not heritable, but are induced by the environment, and may include several different sub-populations (Marr, 1957; cited in Begg and Waldman, 1999). These interactive effects with the environment, selection and genetics on individual ontogenies produce morphometric differences within a species (Cadrin, 2000). Begg and Waldman (1999) describe morphometrics to include analyses of body shape of particular morphological features of various body dimensions or parts and these data being continuous, must be corrected for size differences among specimens. Morphometric expression is also under simultaneous control of genetic and environmental factors, just as in meristics.

It should thus be noted that according to Meffe and Caroll (1994; cited in Begg and Waldman, 1999), isolated populations tend to reduce their genetic variability and, consequently, their ability to adapt to environmental variation, thereby restricting their evolutionary options and thus populations. As such, populations of the same species differ from one area to another. The phenotypic characters may thus be recognizable as a basis for separation and management of distinct populations.

1.2 Problem Statement

For effective fisheries management, knowledge of stock structure, distribution of fishing effort and mortality are essential, since each stock must be managed separately to optimize their yield (Begg *et al.*, 1999). The White-spotted rabbit fish is a fast growing, short-lived, important food fish in Kenya and the East African region. It is among the highly targeted, high commercial value and heavily fished yet abundant species on the Kenyan coast. In a survey of the composition of artisanal catch, Wakwabi *et al*. (2003) noted that 80% of the marine production comes from the shallow coastal waters and reefs. Nzioka (1984) and Fondo (2004) found that Siganids made up a huge percentage of the total catch after *Lethrinus* spp. (Kulmiye *et al.,* 2002), while *Lutjanus* spp. was the third most abundant group of fish in artisanal catch of the local fishery of Kenya. Since variations in fishing effort is evident, dramatic changes in growth, development and maturation of the species may result. Field observations and unpublished data show that variations in size already exist between populations of the species along the Kenyan coast, with some populations maturing earlier than others. It is therefore necessary to successfully separate populations of the White-spotted rabbit fish to aid in management, by determining what degree of isolation may result in notably phenotypic and genetic differentiation among its populations.

1.3 Justification

Fisheries management in the coastal region is largely focused on industrial fisheries (Robinson *et al*., 2008) with few artisanal fisheries having clearly defined harvesting strategies or management plans including controls on inputs and outputs. Already, the coastal fisheries are being overexploited and ecosystem impacts of fishing are widespread (De Young, 2006; cited in Robinson *et al*., 2008). This is evident with some areas practising beach seining thereby affecting the population structure of the species. Although of great commercial importance, little work has been done on the White-spotted rabbit fish in Kenya, with the exception of work done on age and growth parameters (Ntiba and Jaccarini, 1988), on their reproductive biology (De Souza, 1988; Ntiba and Jaccarini, 1990) and on their length-weight relationship and condition factor by De Souza (1988; cited in Ntiba and Jaccarini, 1990 and Wambiji *et. al.*, 2008) and by Wambiji *et al.* (2008). The study attempts to bridge gaps in management in terms of providing evidence that can shift focus to artisanal fisheries in order to develop clearly defined harvesting strategies or management plans that allows for conservation and Ecosystem-Based Fisheries Management (EBFM).

1.4 Objectives

1.4.1 Overall objective

The overall objective of the study is to determine whether morphometric and meristic variations exist in populations of the White-spotted rabbit fish and to separate the different populations of the fish along the Kenyan Coast.

1.4.2 Specific Objectives

The specific objectives were:

- 1. Use morphometric and meristic characteristics to separate populations of the Whitespotted rabbit fish from three locations along the Kenyan coast.
- 2. To identify the best set of morphometric and meristic characters for group separation.

1.5 Hypothesis

The general hypothesis for the study is that fragmentation of the White-spotted rabbit fish populations occurs along the Kenyan coast.

 H_0 – There is no difference between populations of the White-spotted rabbit fish from Malindi, Mombasa and Shimoni.

CHAPTER 2

2.0 LITERATURE REVIEW

The White-spotted rabbit fish, *Siganus sutor* (Valenciennes, 1835), is a member of the family Siganidae that is endemic to the Western Indian Ocean (Woodland, 1990; cited in Borsa *et al.*, 2007). This family in its present description are an economically important group of herbivorous fish consisting of a single genus, *Siganus* (Borsa *et al.*, 2007) and show uniformity in morphological characters and unique bright colour patterns that are usually exploited for defining species boundaries. However, higher level classification in this genus relies on gross body proportions, shape of tail and the snout length (Woodland, 1990; in Borsa *et al.*, 2007). It is a typical Indo-Pacific coral reef fish occupying all types of coastal habitats, occurring in shoals from estuaries and mangroves to the reef front, reef flat, seagrass and seaweed mats in the lagoon (Borsa *et al.*, 2007; Froese and Pauly, 2011). It is one of the most important commercial fish species in artisanal marine fisheries along the East African coast (Nzioka, 1984; Ntiba and Jaccarinni, 1988; Geets *et al.*, 1997).

The species reaches to a maximum size of 45 cm (Froese and Pauly, 2011) and a maximum reported age of 3 years but is common at about 30 cm. It reaches sexual maturity at lengths greater than 20 cm (Kamukuru, 2006). It has a total of $13 - 14$ dorsal spines, 10 dorsal soft rays, 7 anal spines and 9-10 anal soft rays and has a pre-opercular angle of 88° – 98° . Colouration is highly variably, often being influenced by substrates colour and mood of the fish (Froese and Pauly, 2011). It is often green-grey to sandy dorsally, becoming paler ventrally in addition to about 30 large white spots that disappear on death. The spines are slender and venomous. The species is abundant throughout the WIO region from Somalia to South Africa and around islands in the Western Indian Ocean. The fish are normally caught with seines, set nets, and traps.

S. sutor is distinguished by its compressed oval body that is fairly slender and a body depth contained in 2.2 to 2.6 times in standard length. It has a pointed head and the anterior nostril having a flap in juveniles, which shortens progressively with age, the tip reaching to less than halfway to posterior nostril in specimens more than 12 cm in length (Fischer and Bianchi, 1984). A forward directed spine is present in front of the dorsal fin, with the longest dorsal spine being the $5th$ or 8th and the last dorsal spine the shortest, but not less than half the length of the longest. The $3rd$ or $4th$ anal spine is the longest, 1.3 to 1.5 times the length of the last anal spine. The spines are coated with toxic mucus. It has a forked caudal fin, median rays being half to two thirds the length of the longest ray (Fischer and Bianchi, 1984). *Siganus sutor* has very minute scales and about $26 - 31$ scale rows between the lateral line and bases of leading dorsal spines.

Recently published studies suggest that morphological variations occur within species (Murta, 2000; Poulet *et al.*, 2004; Turan, 2004). Morphometric and meristic variations have been used to investigate the population structure of the European hake in the Northeast Atlantic and Mediterranean Sea and the results showed that significant differences among areas (Abaunza *et al.*, 2001). This was more evident between the Mediterranean samples and those from the Atlantic. Abaunza *et al.* (2001) suggested that a Northern stock of European hake exist in addition to a Southern stock and that environmental conditions could possibly have an effect on the differences in stock for the European hake. Studies done in the Atlantic temperate waters on the Atlantic herring, *Clupea harengus*, by Armstrong and Cadrin (2001) found that morphometric differences exist between pre-spawning and post-spawning *Clupea harengus* and they were thus able to differentiate spawning groups of the Atlantic herring stocks.

Erguden *et al.* (2009) was also able to discriminate stock populations of the chub mackerel, *Scomber japonicus*, in the Black, Marmara, Aegan and Northeastern Mediterranean Seas whereby using discriminant function analysis, head morphometric characters played an important role in differentiating samples examined. Chub mackerel from the Mediterranean had larger heads and smaller mouths compared to the Northern groups in the Aegan, Marmara and Black Seas, a possible attribute to growth responses to the differing habitats arising from oceanographic and ecological conditions (Erguden *et. al.*, 2009).

In tropical Malaysian waters, multivariate discriminant analyses was able to differentiate two different species of Congeneric archer fishes *Toxotes chatareus* and *T. jaculatrix* based on their morphometric and meristic elements (Simon *et al.*, 2010). Other work done has been in freshwater fishes on *Saratherodon melanothoron* by Omoniyi and Agbon (2008) in Nigeria in which results revealed significant differences in body depth and caudal peduncle depth a month other morphological characteristics.

CHAPTER 3

3.0 METHODOLOGY

3.1 Study Area

The Kenyan coastline is about 600 km long from Kiunga in the North at about 1^0 41'S to Vanga in the South at 4^0 40'S, bordering Somalia at the North and Tanzania at the South, boasting an almost continuous fringing coral reef parallel to it (Kaunda-Arara *et al.*, 2003; Okemwa *et al.,* 2005). Two alternating seasons that strongly influence weather and productivity patterns of marine organisms at the Kenyan coast are experienced and include the southeast monsoon (SEM) running from April to October and the northeast monsoon (NEM) from November to March (McClanahan, 1988).

Three study sites (Figure 1) were selected – Malindi, Mombasa and Shimoni. Malindi is located at 3^0 15'S and 40^0 07'E and is unique in that it has a broken fringing reef 0.5 – 1.5 km offshore with shallow lagoons and a few reef flats that are exposed at some places during low tide and the area is also characterized by numerous patch reefs and rocky islands. Mombasa area located at 4° 04'S and 39⁰ 40'E also has a fringing reef 0.5 – 1.5 km offshore with shallow lagoons and a reef flat that is exposed at some places during low tide, sloping down to depths of greater than 30m. Shimoni on the other hand, is located at 4^0 38'S and 39⁰ 22'E about 75 km from Mombasa Island and lies within the Funzi Bay complex bordering Msambweni to the North and Vanga to the South. The area is characterized by numerous patch reefs and rocky islands.

Figure 1: Kenya coastline showing the study sites in Malindi, Mombasa and Shimoni.

3.2 Experimental Design

A total of 3 landing beaches, one from each site, were sampled taking into account the varied topography of the East African/Kenyan reefs which limits distribution and seasonal migration of demersal fishes (Nzioka, 1979), the frequency of landings for the species and the presence of the beach management units (BMU's) to facilitate smooth sampling of species during landings at the beaches. Fish were caught by baited traditional *dema* traps common in the traditional artisanal fishery and obtained at the landing beaches as they were being landed by fishers. The traps are hexagonal in shape measuring about $0.9 - 2.0$ m diagonally, 20 cm in height and 10 cm at the mouth opening and are baited with seaweeds before setting daily. The fish samples were sorted to family level and all Siganid specimens identified and sorted to species level for morphometric and meristic analysis using identification keys by provided in FAO species catalogue (1984), Smith's Sea Fishes (Smith and Heemstra, 2004) and as described in FishBase (Froese and Pauly, 2011). These were compared to type specimens (paralectotype) from Kenyan coastal waters available at the National Museums of Kenya (NMK), Nairobi (catalogue number NMK/MW/1000/1 – 2) to ensure proper identification. The paralectotype is based on the designated lectotype *Amphacanthus sutor* (Valenciennes, 1835) from Seychelles which is at the Museum National d'Histoire Naturelle (MNHN) in Paris, France (catalogue number MNHN: A-1805 and paralectotypes from La Réunion and doubtfully Malabar (apparently lost). Current status is valid as *Siganus sutor* (Valenciennes, 1835).

At least 30 *S. sutor* fish samples were collected from each site, based on Reist's (1985) recommendation that at least 25 specimens be used for morphological analysis (Turan, 2004) and effort was made to obtain adult sized fish that were at least 20 cm in length so as to ensure that the characters were completely defined.

3.4 Data Collection

A total of 98 specimens were collected between December 2011 and January 2012. 32 adult specimens were randomly collected from Mayungu landing beach, Malindi, ranging from 25.4 – 39.2 cm TL and 30 adult specimens ranging from 17.1 – 31.9 cm TL were randomly collected from Bamburi landing beach in Mombasa. In Shimoni, a total of 36 adult specimens were randomly collected ranging from $19.0 - 35.8$ cm TL in size. Collected samples were transported in ice using a cooler box back to the laboratory and stored frozen.

Figure 2: *Siganus sutor* (Valenciennes, 1835) from FAO species identification sheets (Fischer and Bianchi, 1984).

31 Morphometric and 6 meristic characters are given in table 1 and illustrated in Fig. 3. Morphometric measurements were taken from the left lateral aspect, and measured to the nearest 0.05 cm using a vernier calliper (Mitutoyo, Japan). All meristic characters (table 1) were counted twice by the same observer. The numbers of teeth were excluded in the present study due to loss as a result of poor handling. Morphometric and meristic data was recorded in pre-printed numbered data forms and the sexes of the specimens were also determined and recorded.

	Character	Description	Acronym		
	31 Morphometric measurements				
1	Total Length	Tip of snout (upper jaw) to tip of longest caudal fin ray	TL		
$\boldsymbol{2}$	Standard Length	Tip of upper jaw to tail base	SL		
3	Head Length	Tip of upper jaw to posterior end of gill opening	HL		
4	Head Depth	Vertical measurement across anterior end of gill opening	HD		
5	Head Width	Distance between posterior ends of the opercular	HW		
6	Mouth Width	Greatest width of the mouth anterior to eye	MW		
7	Snout Length	Tip of upper jaw to anterior border of eye	SnL		
8	Eye Diameter	Greatest bony diameter of the orbit	ED		
9	Inter-orbital width	Width between the two eyes	IOW		
10	Post-orbital length	From posterior border of eye to posterior end of operculum	Porbl		
11	Body Depth	Maximum depth measured from the base of dorsal spine	BD		
12	Body Width	Greatest width just posterior to the gill opening	BW		
13	Pre-dorsal distance	Tip of upper jaw to anterior base of dorsal fin	PDD		
14	pre-pectoral distance	Tip of upper jaw to anterior base of pectoral fin	PPD		
15	Pre-ventral distance	Tip of upper jaw to anterior base of ventral (pelvic) fin	PVD		
16	Pre-anal distance	Tip of snout (upper jaw) to anterior base of anal fin	PAD		
17	Pectoral-anal fin	Distance from anterior base of pectoral fin to anterior base	PtAFD		
18	distance Ventral-anal fin	of anal fin Distance from anterior base of ventral fin to anterior base	VtAFD		
	distance	of anal fin			
19	Dorsal fin base length	Distance from anterior to posterior base end of dorsal fin	DFbL		
20	Dorsal fin ray length	Longest dorsal fin length	DFL		
21	Dorsal spine length	Longest dorsal spine $(5th$ or $8th$) length	GDspL		
22	Pectoral fin length	Distance from anterior to posterior end of the pectoral fin	PFL		
23	Ventral fin length	Distance from anterior to posterior end of the ventral fin	VFL		
24	Ventral spine length	Longest $(1st)$ ventral spine length	VspL		
25	Anal fin base length	Distance from anterior to posterior base end of the anal fin	AFbL		
26	Anal fin ray length	Longest anal fin length	AFL		
27	Anal spine length	Longest anal spine $(3rd$ or $4th$) length	GAspL		
28	Lower jaw length	Straight line measurement between the snout tip and			
		posterior edge of mandible	LwJL		
29	Lower jaw width	Distance between the posterior ends of the mandible	LwJW		
30	Caudal peduncle	Distance from posterior end of dorsal/anal fin to base of	CPL		
31	length Caudal peduncle	vertebral column Depth of caudal peduncle taken in middle of its length	CPW		
	width				
6 meristic counts					
1	Dorsal fin spine	Number of dorsal fin spines	Dspine		
$\boldsymbol{2}$	Dorsal fin ray	Number of branched rays on dorsal fin	Dray		
3	Anal fin spine	Number of anal fin spines	Aspine		
4	Anal fin ray	Number of branched rays on anal fin	Aray		
5	Pectoral fin rays	Number of pectoral fin rays	Pectray		
6	Gill rakers	Number of gill rakers on both upper and lower arms	Grakers		

Table 1: List with abbreviations and definitions of the measurements used.

Figure 3: *Siganus sutor* from FAO species identification sheets showing some of the morphometric measurements that were taken (Modified from Fischer and Bianchi, 1984).

3.5 Data Analysis

To determine intraspecific variations in *S. sutor*, multivariate analysis was carried out separately for morphometric and meristic characters, since these variables are different statistically (meristics being discrete, morphometrics being continuous) and biologically (meristics being fixed earlier in development while morphometrics are more susceptible to the environment) (Erguden *et. al.,* 2009; Ihssen *et. al.,* 1981, cited in Costa *et. al.*, 2003; Hurlbut and Clay, 2008).

Because of the variation in size of fish from different areas, morphometric and meristic data were statistically adjusted to permit comparative analysis in terms of shape and counts independently of size (Costa *et. al.*, 2003; Thorpe, 1976, cited in Erguden *et. al.,* 2009). All morphometric analysis was transformed to common logarithms to obtain a better approximation to multivariate normality since linearity and normality are usually more closely approximated by logarithms rather than original variables (Hair *et. al.*, 1998, cited in Costa *et. al.*, 2003). Following Mekkawy and Mohammad (2011) method, the allometric coefficients of the raw morphometric characters and their relationship with fish size (TL) were estimated from the pooled combined-group sample using power function equation (common logarithms) and linear regression models $(y = bx + c)$ respectively. The type of allometry was evaluated by testing the significance of the allometric coefficients $b (b = 1, b > 1)$ 1 and $b < 1$ for isometry, negative allometry and positive allometry respectively) that serves as a criterion for the intensity of differential increase in morphological characters relative to a certain reference length (Mekkawy and Mohammad, 2011). Detected outliers in the morphometric regression analysis would have subsequently been withdrawn from further consideration. This procedure did not find any outliers, thus no specimen was removed from further consideration.

Morphometric character measurements were standardized to mean total length using the formula (Abaunza *et al.*, 2001):

$$
Log_{10} M_{adj} = Log_{10} M_{obs} - b (Log_{10} L_o - Log_{10} L_t)
$$

Where M_{adj} = standardized morphometric variable; M_{obs} = the uncorrected variable value (observed measurement); L_t = mean total length considered for all samples (from all locations); L_o = total length of each fish and *b* is the allometric coefficient for the respective character (slope of the relationship between log *Mobs* and log *Lo*). This regression model was chosen because none of these variables could be considered either independent or explanatory according to Murta (2000) hence; it is an appropriate procedure for objective analysis of the data when there is a size overlap among the groups being examined. The variable total length was used to standardize the rest of the variables and all statistical analysis was performed for combined sexes since size effects were removed for all samples. The collected samples were either predominantly male or female for two sites with one site having an almost equal distribution of both sexes (Malindi, male $n = 26$, female $n = 6$; Mombasa, male $n = 8$, female $n = 22$; Shimoni, male $n = 20$, female $n = 16$), therefore sex effects were not considered in the present study.

Stepwise discriminant analysis was performed using STATISTICA 7.0 Software to derive discriminant functions for the size corrected morphometric data and the untransformed meristic count data in order to identify those characters that contributed significantly and found useful in population differentiation. Populations were differentiated using Wilks' lambda test with values ranging from 1.0 (no discriminatory power) to 0.0 (perfect discriminatory power).

Correlation coefficients between each pair of characters were calculated using Minitab (Release 14) statistical software, to check if the data transformation (log transformation) was effective in reducing the influence of size in the measurements. It is expected that the absolute value of correlation coefficients would decrease after size correction and, values highly correlated after size effect removal would be considered redundant and the data set could be reduced (Murta, 2000). Bartlett's Test of Sphericity (BTS) was applied to test the null hypothesis that the correlation matrix is an identity matrix (i.e. all diagonal elements are 1 and all off diagonal elements decreased to 0 or close to 0). Highly correlated morphometric characters were thus eliminated from subsequent analysis. A Measure of Sampling Adequacy (MSA) was obtained for all the variables to ensure a value of greater than 0.5 that would allow for continuation of analysis (MSA values < 0.5 are unacceptable). MSA and BTS provided a minimum standard which should be passed before a PCA (or factor analysis) is conducted and were done using SPSS 17.0 Statistical Software.

The data obtained was then subjected to factor analysis using principal component analysis (PCA) from Paleontological Statistics Software Package (PAST) for education and data analysis to identify characteristics that are important in distinguishing population groups in the pooled sample i.e. the degree of similarity among samples in overall analysis and relative importance of each measurement for group seperation. Principal components were extracted from the variance-covariance matrix since all the variables were measured in the same units (milimeters). Population centroids (mean values) and 95% asymptotic confidence limits (ellipses) of the scores of individual White-spotted rabbit fish on the first two principal components were then computed for each sample.

CHAPTER 4

4.0 RESULTS

4.1 Fish Size and Sex

Original name for the species is *Amphacanthus sutor* (Valenciennes, 1835) designated lectotype from Seychelles, Western Indian Ocean (MNHN A – 1805) and paralectotypes are from Réunion and doubtfully Malabar (apparently lost), and current status is valid as *Siganus sutor* (Valenciennes, 1835). Overall, 98 specimens of *S. sutor* were examined for morphometric and meristic data. The mean total length of *S. sutor*, covering a wide sizerange (171.0 – 392.0 mm TL, across all populations amounted to 272.09 ± 46.994 mm (Table 2).

Table 2: Mean total lengths, standard lengths and percentages of males and females sampled.

Locality	Mean TL $(cm) \pm SD$ and $(range)$	Mean $SL \pm SD$ and (range)	Females $($ %)	Males $(\%)$
Shimoni	277.0 ± 48.70 (19.0 - 35.8)	226.3 ± 39.56 (15.4 - 29.4)	44.4	55.6
Mombasa	$244.2 \pm 44.45(17.1 - 31.9)$	190.5 ± 35.63 (13.3 - 24.9)	73.3	26.7
Malindi	$292.7 \pm 34.18(25.4 - 39.2)$	233.8 ± 27.84 (19.8 - 31.7)	18.8	81.3
Combined	272.1 ± 46.99 (17.1 - 39.2)	217.8 ± 39.16 (13.3 - 29.4)	44.9	55.1

4.2 Correlation coefficients

Correlation coefficients between characters before and after size effect removal are presented

in table 3.

Table 3: Morphometric correlation coefficients between characters, before and after removal of the size effect, are respectively shown below and

above the diagonal. Values higher or equal to 0.95 are shown in bold.

Most coefficients between morphometric characters were very close to 1 before size correction, and were considerably reduced after (values ≥ 0.95 highlighted in bold in table 3). The lowest correlation before the size effect removal was 0.54 whereas after removal, the highest was 0.71, with most coefficients changing to values close to zero after removal of the size effect. As such, the effect of body length had been successfully removed with allometric transformation with variables that were still highly correlated after size effect removal considered redundant and the data set reduced.

4.3 Discriminant Function Analysis

Derived Discriminant Functions in Stepwise DFA using 30 of the size corrected morphometric characters identified Caudal Peduncle Width (CPW) contributing the most in variations, followed by Lower Jaw Width (LwJW), Snout Length (SnL), Pre-Dorsal Distance (PDD), Head Diameter (HD), Standard Length (SL), Pectoral Fin Length (PFL) and Dorsal Fin base Length (DFbL) as shown in red in table 5 (Wilks' λ (lambda) = 0.05738, approx. F $(60, 132) = 6.9840$, $P < 0.00001$), while the rest of the characters were not significant (table 4).

Characters	Wilks' Lambda	Partial Lambda	F- remove	p-level	Toler.	1-Toler. $(R-Sqr.)$
Morphometric variables						
CPW	0.073579	0.779875	9.314456	0.000273	0.550805	0.449195
HD	0.065024	0.882482	4.394539	0.016154	0.730696	0.269304
PDD	0.065926	0.870409	4.913225	0.010253	0.392847	0.607154
SL	0.063320	0.906224	3.414820	0.038795	0.540187	0.459813
LwJW	0.068495	0.837758	6.390836	0.002903	0.538173	0.461827
SnL	0.068316	0.839956	6.287767	0.003166	0.525076	0.474924
PORBL	0.060576	0.947278	1.836670	0.167398	0.422265	0.577735
HL	0.059397	0.966083	1.158555	0.320241	0.283182	0.716818
ED	0.061869	0.927482	2.580214	0.083383	0.583090	0.416911
PFL	0.063262	0.907062	3.381189	0.039997	0.617517	0.382483
LwJL	0.060999	0.940711	2.079841	0.133064	0.498833	0.501167
MW	0.061400	0.934564	2.310580	0.107177	0.567913	0.432087
BW	0.059993	0.956490	1.501151	0.230384	0.592676	0.407324
DFbL	0.062903	0.912238	3.174773	0.048258	0.354873	0.645127
PAD	0.060104	0.954710	1.565475	0.216649	0.518334	0.481666
AFL	0.059247	0.968530	1.072245	0.348125	0.423314	0.576686
GDSpL	0.059386	0.966262	1.152214	0.322209	0.682805	0.317195
CPL	0.059273	0.968109	1.087088	0.343157	0.641598	0.358402
IOW	0.058955	0.973324	0.904433	0.409729	0.659159	0.340841
AFbL	0.059589	0.962969	1.269031	0.287872	0.339028	0.660972
VFL	0.058396	0.982633	0.583237	0.560939	0.471231	0.528769
PectAFD	0.058827	0.975440	0.830884	0.440169	0.468917	0.531083
VentAFD	0.058056	0.988398	0.387376	0.680369	0.445975	0.554025
DFL	0.058504	0.980826	0.645129	0.527871	0.539317	0.460684
HW	0.058123	0.987252	0.426125	0.654819	0.447412	0.552588
BD	0.057957	0.990085	0.330484	0.719758	0.553595	0.446405
PPD	0.057856	0.991805	0.272683	0.762188	0.571659	0.428341
GASpL	0.057675	0.994924	0.168356	0.845415	0.477599	0.522401
VSpL	0.057586	0.996465	0.117057	0.889719	0.594751	0.405249
PVD	0.057484	0.998224	0.058710	0.943030	0.477574	0.522426
			$F(60, 132) = 6.9840$			
Total	0.0573823					
			P < 0.00001			
Meristic variables						
Pect.Rays	0.637783	0.878178	6.519897	0.002230	0.021192	0.978808
Gill rakers	0.604034	0.927244	3.687821	0.028716	0.021192	0.978808
				$F(4, 188) = 15.80151$		
Total	0.5600870					
			P < 0.00001			

Table 4: Summary of discriminant function analysis for morphometric and meristic character.

4.4 Principal Component Analysis

PCA was applied to the morphometric data available because the MSA (Kaiser-Meyer-Olkin (KMO) measure) was 0.728 (values lower than 0.5 are unacceptable) and, Bartlett's Test of Sphericity (BTS) (test for the presence of correlations among variables) gave a $p \le 0.001$. 31 principal components (PCs), which contain a percentage of total variance of all variables, were produced, and 29.67% and 11.16% of the total variation were presented in the first and second PCs respectively (40.1% combine variance of PC1 and PC2), showing that the PCA was a success (Table 5).

PC	Eigenvalue	$\frac{1}{2}$ Variance
1	0.00937271	29.646
\overline{c}	0.00352490	11.149
3	0.00249871	7.904
$\overline{4}$	0.00224822	7.111
5	0.00181128	5.729
6	0.00153677	4.861
$\overline{7}$	0.00123969	3.921
8	0.00117604	3.720
9	0.00099868	3.159
10	0.00084800	2.682
11	0.00071291	2.255
12	0.00068936	2.181
13	0.00060972	1.929
14	0.00053766	1.701
15	0.00053239	1.684
16	0.00043637	1.380
17	0.00041135	1.301
18	0.00039897	1.262
19	0.00035568	1.125
20	0.00033018	1.044
21	0.00027147	0.859
22	0.00024148	0.764
23	0.00019327	0.611
24	0.00017022	0.538
25	0.00014394	0.455
26	0.00011386	0.360
27	0.00008395	0.266
28	0.00005857	0.185
29	0.00004338	0.137
30	0.00002536	0.080
31	0.00000000	0.000

Table 5: Principal components on the variance-covariance matrix showing significance of components**.**

The scree plot which measures the variance accounted for by corresponding components (variables) indicates the first four PCs being significant components accounting for 55.82% of the total morphological variations (Figure 4).

Figure 4: Scree plot shows that the eigenvalues for the 31 components (blue line) lie above random model (broken stick) values (red line) for the first four components. Those below represent non-significant components.

Population centroids (mean values) and 95% asymptotic confidence limits (ellipses) of the scores of individual White-spotted rabbit fish on the first two principal components are shown in the PCA scatter diagram. The red centroids represent the population from Shimoni, South Coast, the green represent population from Mombasa and Malindi populations are represented in blue (Figure 5).

Figure 5: PCA scatter diagram showing population centroids and 95% ellipses of scores of individual White-spotted rabbit fish (*S. sutor*) on the first two principal components from Shimoni (red), Mombasa (green) and Malindi (blue).

Shimoni and Mombasa populations appear different from one another while the Malindi populations show sharing of characteristics with both Shimoni and Mombasa.

Morphometric characters separating individuals of the White-spotted rabbit fish are shown in the PCA scatter diagram showing contribution of the variables to the principal component functions.

Figure 6: Contributions of morphometric variables to the component functions. Vectors indicate the loadings of the scores for each variable on the first two PCs.

PCA applied to meristic data on 5 principal components (PCs), resulted in 85% of the total variation being presented in the first PCs (Table 6).

Table 6: Principal components showing significance of components in meristic data.

PC		Eigenvalue % Variance
	3.23	85.54
2	0.52	13.75
3	0.02	0.61
4	0.00	0.10
5	0.00	0.00

Population centroids (mean values) and 95% asymptotic confidence limits (ellipses) of the scores of individual White-spotted rabbit fish on the first two principal components are also shown in the PCA scatter diagram (Figure 7). Shimoni and Mombasa populations appear different from one another while the Malindi populations show sharing of characteristics with both Shimoni and Mombasa.

Figure 7: PCA scatter diagram showing population centroids and 95% ellipses of scores of individual White-spotted rabbit fish (*S. sutor*) on the first two principal components from Shimoni (red), Mombasa (green) and Malindi (blue).

Meristic characters separating individuals of White-spotted rabbit fish are shown in the PCA scatter diagram (Figure 8) showing contribution of the variables to the principal component functions.

Figure 8: Contributions of meristic variables to the component functions. Vectors indicate the loadings of the scores for each variable on the first two PCs.

CHAPTER 5

5.0 DISCUSSION

The hypothesis raised in this study that morphometric and meristic variations of the Whitespotted rabbit fish population exists and that their population is fragmented along the Kenyan coast was fully confirmed. Morphometric and meristic analyses revealed a good discrimination between the White-spotted rabbit fish populations from Shimoni, Mombasa and Malindi. This trend indicates that morphology is a valid tool in stock identification for various fisheries as it has been adopted by many authors to identify and relate different fish races and/or populations (Murta, 2000; Abaunza, 2001; Albertson and Kocher, 2001; Armstrong and Cadrin, 2001; Cadrin, 2000; Aguilla-Perrera, 2004; Poulet *et al.*, 2004; Turan, 2004; Becktas and Belduz, 2009; Mekkawy and Mohammed, 2011).

The Wilks' lambda tests in the derived DFA indicated a small difference between the 3 population groups when their morphometric characters were compared by means of discriminant analysis (table 3) but, intraspecific morphological variation elicited from DFA was mainly related to measures related to head characteristics and fin-related characteristics. This could mean that inter-population variability within stocks of *S. sutor* is as a result to adaptations to the environment and habitat with respect to feeding habits and swimming behaviour respectively.

PCA revealed that the most important components PC1 (size component) and PC2 (shape component) were related fin features (swimming behaviour) and head features (feeding adaptation) respectively. This is in agreement with the earlier seen application of a forward stepwise DFA.

Morphometric discrimination between the three studied populations of *S. sutor* along the Kenyan coast is probably as a result of the feeding strategy adopted in each of the locations and the swimming behaviour. The Shimoni population (negative PC1 values) is quite distinct

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morphometric characters separating these two populations are shown in fig. 6, with VFL and CPW being responsible for separation of Shimoni populations and MW, PORBL, HW and DFL being responsible for the Mombasa populations. This can be attributed to the varying habitat substrates and environment in terms of topography in the two regions since Shimoni is characterized by patch reefs whereas Mombasa is characterized by a fringing reef, for which *S. sutor* have environmentally adapted to. Being a demersal fish species, studies by Kaunda-Arara and Rose (2004) on out-migration of tagged fishes from marine National Parks to fisheries in coastal Kenya suggest that there is an unwillingness to cross sand and deep water habitat patches surrounding patch reefs by *S. sutor* (net swimming distance of about 0.67 ± 1 0.51 km). The same study however suggests that in fringing reefs *S. sutor* travels further distances (1.59 \pm 1.07 km) since such reefs provide continuity. This can explain difference in morphometrics between Shimoni (patch reef) populations and Mombasa (fringing reef) populations since Mombasa populations tend to swim further compared to Shimoni populations as evidenced by the characters VFL and CPW. Furthermore, feeding strategies in patch reefs may be different than in fringing reefs hence supporting the variations in head features that are responsible for feeding/foraging. Mombasa populations predominantly feeding on seagrass in the lagoon while those in patch reefs having to feed on algae that is more common compared to seagrass. This is supported by MW, PORBL, HW and DFL that are separating the Mombasa population from the Shimoni population. It is interesting to note that the Malindi population shares morphometric characters similar to the Shimoni and Mombasa populations as seen by the overlap in fig 5. This can be explained by the fact that the Malindi area is comprised of both patch and alternating fringing reefs. This further validates the fact isolated populations tend to reduce their genetic variability and, consequently, their ability to adapt to environmental variation, thereby restricting their evolutionary options resulting in intraspecific species variation (Begg and Waldman, 1999). As such, morphometric expression is under simultaneous control of genetic and environmental factors.

When it comes to meristics, not much difference is observed in PCA except for the pectoral fins and gill rakers contributing to variations in populations. Meristics may thus not be a precise tool to be used in separating populations of *S. sutor* since according to Begg and Waldman (1999) meristic characters are set early in life, are controlled by both genetic and environmental factors, and remain stable throughout life; thus reflecting environmental influence over a relatively brief period of larval development.

CHAPTER 6

6.0 CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

The results obtained in the present work from PCA showed that morphology can be used to separate stock populations of the White-spotted rabbit fish since isolated populations tend to reduce their genetic variability and, consequently, their ability to adapt to environmental variation, as environmental factors have an influence on the morphology of the species. This restricts their evolutionary options and thus populations, thereby supporting the fact that populations of the same species differ from one area to another.

DFA identified 9 characters that could be used for group seperation of *S. sutor*: Caudal Peduncle Width (CPW) contributing the most in variations, followed by Lower Jaw Width (LwJW), Snout Length (SnL), Pre-Dorsal Distance (PDD), Head Diameter (HD), Standard Length (SL), Pectoral Fin Length (PFL) and Dorsal Fin base Length (DFbL). Head features HD and LwJW showed the most contribution towards separating Mombasa from Shimoni populations while CPW and VFL were responsible for separating Shimoni populations from Mombasa populations as depicted by PCA. The Malindi population has characters evident in both the Shimoni and Mombasa stocks. Meristic characters were not good tools for group separation since the White-spotted rabbit fish shows uniformity in morphological characters.

The results may have implications for the management of the fishery for *S. sutor*. If heavy fishing pressure reduces the stock in one of its main areas of geographic distribution to low levels, significant replenishment by immigration from other areas such as from fringing reefs to adjacent patch reefs or vice versa, may take a long time as the species is sedentary and unwilling to spill over from patch reefs to nearby existing fringing reefs. This is particularly evident in that populations of the White-spotted rabbit fish show no migratory behavior for feeding or spawning. The species matures at an earlier age/size for the Mombasa population compared to those from Shimoni, Malindi and from studies done by Kamukuru (2006).

6.2 Recommendations

Following results of the study, the following recommendations are advanced:

- 1. The species has a high commercial value hence analysis of the morphological variation on a broad geographic scale would be particularly important in order to evaluate its population structure.
- 2. Habitat characteristics need to be studied to determine specific environmental factors responsible for variations.
- 3. For reliable identification, inferences of stock identity from this work should be compared with data obtained from other stock identification methods such as genetics and fluctuating asymmetry.

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