# Reproductive biology of the barred sand bass (*Paralabrax nebulifer*)

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## **ABSTRACT**

he barred sand bass (Paralabrax nebulifer) is a common species in the nearshore marine environment and an important part of the marine recreational fishery of southern California. Related species of Paralabrax are either secondary gonochores or protogynous hermaphrodites. The objective of this study was to determine which reproductive strategy, if any, prevails in barred sand bass. A total of 437 specimens were collected from June 1996 through April 1997 from seven different locations along the southern California coast. Using the criteria outlined by Sadovy and Shapiro (1987), gonadal tissue from all individuals was examined for the presence of features strongly indicative of protogynous hermaphroditism. Two transitional individuals were found: a 273 mm standard length (SL) fish from Huntington Flats and a 306 mm SL fish from San Diego Bay. All males examined had a membrane-lined central cavity in the testes and a sperm sinus in the gonadal wall. These features are believed to result from individuals passing through a femalelike juvenile stage before maturing based upon the examination of juvenile individuals. Fifty-two percent of the males examined had atretic bodies. Males and females were equally distributed throughout the size classes collected. Although most barred sand bass examined appeared to be secondary gonochores, some individuals had the ability to change from female to male (protogynous hermaphroditism). This sexual strategy did not appear to differ between locations.

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#### INTRODUCTION

Barred sand bass (Paralabrax nebulifer), commonly species in the nearshore marine environment, is an important part of the marine recreational fishery of southern California. This species remains one of the most frequent catches for sport-fishers, ranking second in the number of fish taken in the state in 1989 (Oliphant 1990). Together with the kelp bass (Paralabrax clathratus), it forms more than 90% of the general "rock bass" recreational catch in the first half of this decade (Frey 1971). Barred sand bass ranges from Santa Cruz, California, south to Magdalena Bay, Baja California Sur, including Guadalupe Island (Miller and Lea 1972), occupying a variety of different habitats including kelp beds and sand flats on the open coast to inland harbors and bays. They are a benthic, relatively sedentary fish which are rarely found more than 3 m above the substrate, closely associated with bottom structure and sand (Turner et al. 1969, Feder et al. 1974, Larson and Demartini 1984). Barred sand bass are oviparous broadcast batch spawners that spawn from April to August with a peak in July (Love et al. 1996). They tend to form large breeding aggregations, "breeding balls," over mud flat areas during their spawning season. Their eggs and larvae are pelagic, drifting about in open water; juveniles appear in shallow water from late summer to early winter (Love 1991). In barred sand bass, 50% of males matured at 21.9 cm, and 50% of females matured at 23.9 cm. Males mature between the ages of 2 and 4 years, and females mature between the ages of 2 and 5 years (Love et al. 1996). Barred sand bass is a warm temperate member of the family Serranidae and one of the three species of Paralabrax in southern California. Many serranids have specialized reproductive strategies. Some species are protogynous hermaphrodites and hence spend the early part of their lives as females and later change sex to conclude their lives as males. Oda et al. (1993) suggested that barred sand bass might be protogynous hermaphrodites, but

other studies have been inconclusive. Studies on the other two temperate Paralabrax species have shown varying results. Kelp bass (Paralabrax clathratus) found in nearshore coastal waters usually associated with hard substrate and kelp are secondary gonochores and do not change sex (Smith and Young 1966). Spotted sand bass (Paralabrax maculatofasciatus) found in harbors and bay (Allen et al. 1995) have specific populations that are protogynous hermaphrodites (Hastings 1989, Hovey and Allen 2000). The presence of protogynous hermaphrodites was found to be related to population structure and location.

Knowledge of which reproductive strategy is practiced is important for proper management of the fishery. The objectives of this study are to determine if protogynous hermaphroditism occurs in barred sand bass, determine the sexual strategy(ies) practiced, and to determine whether this strategy varies between different locations.

### **METHODS**

A total of 437 specimens were collected by hook and line from June 1996 through April 1997 along the coast of southern California at Redondo Beach, Long Beach, Huntington Flats, Santa Catalina Island, San Onofre, Cardiff, and San Diego Bay (Figure 1). Each specimen was weighed to the nearest gram (g), measured to the nearest millimeter standard length (SL), and preliminarily sexed. Gonads were removed, weighed (g), and preserved in 10% formalin for later histological examination.

Gonads were fixed by immersion in Davidson's solution for two weeks and then transferred to 70% ethyl alcohol. Crosssectional tissue samples (4 mm thick) were removed from the mid-area of each gonad and placed in Tissue Tek tissue cassettes. Using the Tissue Tek tissue fixing unit, cassettes were immersed in a series of increasing concentration (70, 80, 95, and 100%) ethyl alcohol and xylenes. The processed tissue was embedded with paraffin (56° C) and placed in a vacuum infiltrate for 1 h. The surface of the tissue block was exposed and samples were soaked in a detergent bath for 10 to 14 d. Samples were sectioned at 6-12 µm using an 820 Spencer microtome and mounted on slides. The slides were placed in an oven and allowed to dry for 7 d at 35° C. They were then stained using Mayer's alum hematoxylin and eosin, allowed to dry and sealed with cytosol and a cover slip. Gonad morphology was examined using a

compound microscope. Initial sex determination was confirmed based upon the finding of ovarian tissue (females), testicular tissue (males), and indeterminate tissue (immature individuals).

Sadovy and Shapiro (1987) outlined the criteria necessary for diagnosing hermaphroditism in fish. The following features are strongly indicative of protogynous hermaphroditism: (1) transitional individuals; (2) atretic bodies in Stages 1, 2, or 3 of oocytic atresia within the testes; (3) a membrane-lined central cavity in the testes; and (4) sperm sinuses in the gonadal wall. A fifth feature, population structure, can also add support to a diagnosis of hermaphroditism. All samples were examined to determine whether any of these features were present.

#### RESULTS

A total of 207 males, 198 females, 20 immature individuals, and 2 transitional individuals were examined (Table 1). The majority of the fish (males and females) were collected from Huntington Flats. Immature individuals were taken only from Cardiff, Catalina, San Diego, and San Onofre, with the majority being collected from San Diego. One transitional individual was taken from Huntington Flats and one from San Diego. The fish ranged in size from 78-460 mm SL. Size at first maturity for the barred sand bass is

FIGURE 1. Map of sampling locations along the southern California coast for barred sand bass (Paralabrax nebulifer) reproductive study. Locations include: Redondo Beach, Long Beach, Huntington Flats, Santa Catalina Island, San Onofre, Cardiff, and San Diego Bay.

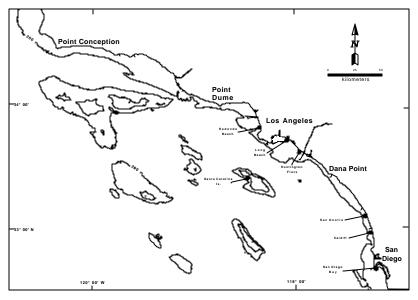


TABLE 1. Number, size, and sex of barred sand bass (*Paralabrax nebulifer*) collected in reproductive biology study, June 1996-April 1997.

Location	Total Fish		Immatures Size Range		Females Size Range		Transitional Size Range		Males Size Range			
	Size Range										Secondary Males	
	No.	(mm SL)	No.	(mm SL)	No.	(mm SL)	No.	(mm SL)	No.	(mm SL)	No.	(%)
Cardiff	80	170-310	1	183	33	170-310	0	na	46	200-290	33	72
Catalina	18	158-395	1	178	6	171-300	0	na	11	158-395	1	9
Huntington Flats	233	191-362	0	na	127	198-362	1	273	106	191-322	39	37
Long Beach	4	202-328	0	na	1	328	0	na	3	202-297	2	67
Redondo	6	250-405	0	na	1	285	0	na	5	250-405	4	80
San Diego	51	78-335	2,278-201		12	169-335	1	306	17	121-308	2	12
San Onofre	44	155-460	5,155-190		19	230-375	0	na	20	230-460	17	85
Total	436	78-460	2978	8-201	199	169-375	2,27	73-306	208	121-460	99	52

na = not applicable.

219 mm SL for males and 239 mm for females. Both transitional individuals were larger than the size at first maturity (273 and 306 mm SL).

Histological sections of a typical female show primary, secondary, and vitellogenic oocytes (Figure 2). Sections of a typical male show spermatocytes, seminiferous tubules, and the crypts containing the developing sperm (Figure 3). Sections of the immature individuals showed developing tissue. All immature individuals were found to pass through a female-like stage with undeveloped tissue resembling immature female features. Histological sections of a transitional individual show proliferating testicular tissue and degenerating ovarian tissue (Figure 4). Vitellogenic oocytes represent the degenerating female tissue, while the presence of developing sperm, sperm sinuses, and spermatocytes show the proliferating male tissue. The transitional individual shown was taken from Huntington Flats in August 1996 (during the later part of the breeding season). The second transitional individual was taken from San Diego in April 1997. This individual was taken outside of the breeding season and showed resting ovarian tissue rather than vitellogenic oocytes, and also contained proliferating testicular tissue. A membrane-lined central cavity (gonadal lumen) and the presence of a sperm sinus in the gonadal wall were found in all males examined (Figure 5). The sperm sinus was found to be used for the transportation of sperm in some of the males examined. Atretic bodies were found in 52% of all males examined (Figure 3).

The distribution of males, females, and immature individuals throughout the size classes was examined. Males and females were found to be equally distributed throughout the different size classes (Figure 6).

FIGURE 2. Female morphology in barred sand bass (*Paralabrax nebulifer*) showing primary oocytes (P), secondary oocytes (S), and vitellogenic oocytes (V).

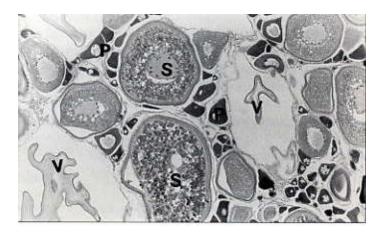


FIGURE 3. Male morphology in barred sand bass (*Paralabrax nebulifer*) showing seminiferous tubules (ST), spermatocytes (SP), and developing sperm (DS). Also pictured is an atretic body (AB) showing the characteristic yellow-brown pigment, and granulose cells (GC).

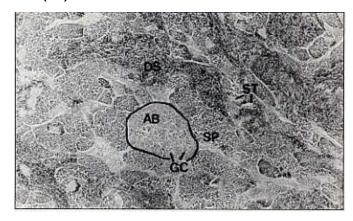


FIGURE 4. Transitional individual in barred sand bass (Paralabrax nebulifer) showing degenerating ovarian (DO) tissue surrounding proliferating testicular tissue (PT).

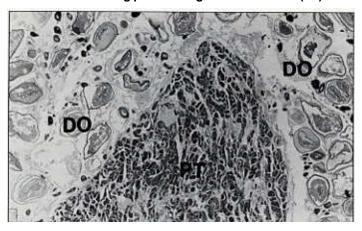


FIGURE 5. Gonadal lumen in male tissue of barred sand bass (Paralabrax nebulifer) showing gonadal (G) lumen and squamos (SQ) epithelium cells. Also pictured is a sperm sinus (SS) (containing developing sperm) in the gonadal wall of a male showing the muscle (M) layer and gonadal wall (GW).

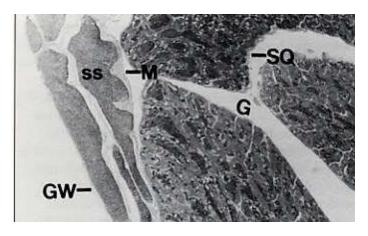
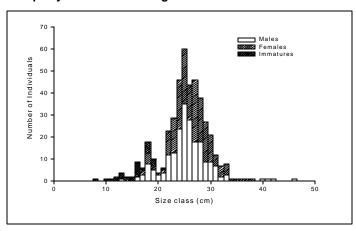


FIGURE 6. Population structure of barred sand bass (Paralabrax nebulifer) showing males and females equally distributed throughout the size classes.



#### DISCUSSION

The increasing presence of the barred sand bass in the sportfish industry (Oliphant 1990) demonstrates a need to study the life history of this species as a means to effectively manage the fishery. The objective of this study was to determine what reproductive method(s) is practiced by the barred sand bass. This objective was accomplished by examining the specimens to see whether protogynous hermaphroditism occurs. The five criteria used to diagnose this condition were the presence of: (1) transitional individuals; (2) a membrane-lined central cavity (gonadal lumen) in males; (3) a sperm sinus in the gonadal wall of males; (4) atretic bodies in Stages 1, 2, or 3 of oocytic atresia (in males), and (5) population structure (Sadovy and Shapiro 1987).

The first and strongest criterion used to indicate sex change is the presence of transitional individuals (i.e., individuals that are in the process of changing sex). The gonadal tissue of protogynous hermaphrodites will show a degeneration of the initial ovarian tissue and a proliferation of testicular tissue. Two transitional individuals were found in this study. While this number may seem relatively small, transitional individuals are relatively uncommon in field studies. They are more typically seen in laboratory studies where sex change is induced experimentally (Sadovy and Shapiro 1987). The fact that transitional individuals were found at all supports the theory that at least a small portion of the population of barred sand bass has the ability to change sex.

The second criterion examined was the presence of a membrane-lined central cavity (gonadal lumen) in the testes. The gonadal lumen is thought to be a remnant of the ovarian lumen, which is used for egg transportation in females. All males examined were found to have gonadal lumens. The presence of a gonadal lumen is only a valid criterion if it is non-functional in males. Sperm was found in this cavity in some of the males examined. This may indicate that the cavity was used for the transportation of sperm in some individuals. If so, this criterion would not be met.

The third criterion examined was the presence of the sperm sinus in the gonadal wall of the males. The sperm sinus in the gonadal wall is thought to develop from the splitting of the muscle layers of the ovarian capsule. All males examined were found to have a sperm sinus in the gonadal wall.

All males examined in this study contained gonadal lumens and sperm sinuses in the gonadal wall. Two theories other than hermaphroditism can account for these two features being present in all males examined. The first is that these features are evolutionary remnants of a prior

historical era when the species was hermaphroditic (Smith and Young 1966). Species that fall into this category are termed secondary gonochores. The second theory is that these features result from all individuals passing through a female-like juvenile stage (Sadovy and Shapiro 1987). An examination of juvenile individuals in this study supported the later theory.

The next criterion examined was the presence of atretic bodies. Atretic bodies are germ cell remnants of the initial sex that are retained after the sex change has occurred. These remnants can be caused by degenerating oocytes (going through atresia), but may also result from the degeneration of other types of cells (Smith and Young 1966). Alternative explanations for the presence of atretic bodies other than oocytic atresia include parasitic encystation (Atz 1964), sperm degeneration (Warner 1975b), and nonspecific tissue degeneration (Smith 1965). Atretic bodies were found in 52% of the males examined.

An attempt was made to estimate the number of individuals that had undergone sex change. Since the gonadal lumen and the sperm sinus in the gonadal wall were common characteristics among all males examined, the presence of atretic bodies was used as an indication that sex change had occurred. Males possessing atretic bodies are termed secondary males based upon the assumption that they began their lives as females and secondarily changed to males. The percentage of secondary males varied among collection sites on the southern California coast (Table 1). While some of the percentages appear rather high, it is important to note that a small number of males were examined at some of those locations. Also, the percentages for San Diego Bay and Huntington Flats are not among the highest; however, these were the only two locations with transitional individuals. The number of individuals undergoing sex change may have been underestimated because atretic bodies are not retained indefinitely. As time passes following the change of sex, fewer germ cell remnants are present. On the other hand, the number may be overestimated since some atretic bodies are formed by methods other than oocytic atresia (Smith and Young 1966). It is important to remember that this estimate is being based solely upon one characteristic.

The final criterion examined was population structure. Typical protogynous populations have a bimodal distribution, with females making up a greater portion of the smaller individuals and males making up a greater portion of the larger individuals. In this study, an equal distribution of males and females was found throughout the size classes (Figure 6). Similar results were found for individual locations. However, factors that may obscure bimodality in protogynous populations include: (1) lack of sexual dimor-

phism; (2) alternative pathways of male sexual development; (3) bisexual juveniles; and (4) variation in size at sex change (Sadovy and Shapiro 1987). Thus, protogyny was not ruled out on the basis of the length-frequency distribution graph. In addition, barred sand bass generally form large breeding aggregations during their spawning season and many of the individuals used in this study were obtained during this period. Individuals were grouped based upon their capture location. One sampling location may have contained aggregating individuals from several different subpopulations or locations.

Protogyny is advantageous where larger males are able to dominate access to females (Warner 1975a). In the large breeding aggregations that are formed by barred sand bass, size does not appear to be an issue. Individuals group together and all release their gametes (regardless of their size). This spawning behavior may explain why protogynous hermaphrodites were infrequently found at any of the sites. However, this study did not look at the individual populations that occur outside of the breeding season (where a larger size may hold an advantage). Kelp bass and barred sand bass are known to gather in large schools during spawning (Limbaugh 1955) and both species are thought to be gonochoristic. Studies conducted on the spotted sand bass showed protogynous hermaphroditism to be most prevalent in dense, isolated populations (Hovey and Allen 2000).

In conclusion, barred sand bass does possess the ability to change sex, as shown by the presence of transitional individuals and supported by males possessing atretic bodies. However, few individuals actually do so. Instead, this species exhibits a combination of two different reproductive strategies: protogynous hermaphroditism and gonochorism, with the latter appearing to be the most prevalent. Based upon the sites examined in this study, location does not appear to be a determining factor.

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