

Behavior, Physiology, and Life History Comparisons in Four Species of Grouper: What do they mean for Grouper Management?

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ABSTRACT

The objective of this study was to conduct a preliminary investigation of the interplay between reproductive behavior and spawning strategy in territorial (*E. morio*, *E. guttatus*, and *M. microlepis*) and non-territorial (*E. striatus*) grouper species. Non-territorial males tend to have high gonadosomatic index (GSI) and are sperm competitors, while territorial males tend to have low GSI's and are resource (mate) competitors. *Epinephelus striatus* has a substantially higher GSI than the other three species, indicating its strategy as a sperm competitor. The physiology/anatomy/behavior relationship has been shown elsewhere between males within a species, but has not before been described across related species in which each species exhibits only one reproductive type. This relationship becomes important when we consider decreased overall male densities and associated skews of sex ratio in some groupers. This may lead to a disruption of the positive feedback loops and a physiological explanation for depensation/Allee effects in certain grouper species.

KEYWORDS: grouper, reproduction, physiology, life history, depensation

Comparaciones de Comportamiento, Fisiología, e Historia de Vida entre Cuatro Especies de “meros”: ¿Cuál es su significado para manejo?

El objetivo de este estudio fue el de conducir una investigación preliminar sobre la interacción entre el comportamiento reproductivo y las estrategias de desove en especies de meros territoriales (*E. morio*, *E. guttatus*, y *M. microlepis*) y no-territoriales (*E. striatus*). Los machos no-territoriales tienden a tener un alto índice gonadosomático (IGS) y son competidores de esperma, mientras que los machos territoriales tienden a tener un bajo IGS y compiten (se aparean) por recursos. *E. striatus* tiene un IGS sustancialmente más alto que las otras tres especies, lo que demuestra su estrategia como competidor de esperma. La relación entre fisiología / anatomía / comportamiento ha sido demostrada otras veces entre machos de una misma especie, pero hasta ahora no ha sido descrita entre especies relacionadas, donde cada especie presenta únicamente un tipo de reproducción. Esta relación es importante cuando se consideran disminuciones en las densidades totales e inclinaciones asociadas a la proporción sexual en algunos meros. Esto puede llevar a la disrupción del circuito de refuerzos positivos, y a una explicación fisiológica de los efectos de decompensación /Allée en ciertas especies de meros.

PALABRAS CLAVES: mero, reproducción, fisiología, historia de vida, decompensación.

INTRODUCTION

Groupers (Family Serranidae) of the southeast United States and Caribbean exhibit an array of reproductive strategies, from pair-spawning while in small aggregations of a few individuals to mass spawning in groups of tens of thousands, at least historically (Smith 1972, Colin et al. 1987, Gilmore and Jones 1992, Sadovy et al. 1994, Coleman et al. 1996). Group spawning groupers tend to be non-aggressive (while spawning), while pair-spawning species court females and defend territories against other males. More generally this reflects a sperm competition vs. mate competition life history strategy. The majority of grouper species studied have also been shown to be protogynous hermaphrodites. This means that there is a natural skew to sex ratio, with females being the dominant sex.

Reproductive strategy is reflected in relative testes size (gonadosomatic index, GSI). Males with high GSI tend to be the non-territorial and non-aggressive type, while males with low GSI are often territorial and aggressive. This relationship is seen across a wide range of vertebrate species (Harcourt et al. 1981; Cardwell and Liley 1991; Ku-

sano et al. 1991; Jennions and Passimore 1993; Harcourt et al. 1995).

Androgens, primarily testosterone and its derivatives, play a key role in these behaviors. In addition to stimulating the proliferation and maturation of gametes and the development of secondary sexual characteristics, androgens induce various reproductive behaviors including territorial displays and aggressive interactions (reviewed in Borg 1994). The trend is for androgen levels in territorial and aggressive males to be elevated above those in males that are subordinate or non-territorial. According to the challenge hypothesis (Wingfield 1984), dominant males experience positive feedback in the form of increased androgen levels upon successful defense of a territory. This positive feedback loop may be necessary for complete reproductive success in males.

Problems may arise when the natural spawning structure is disrupted through fishing. In addition to potential direct disruption of behavior on the aggregation, loss of males through compression of age structure (Huntsman and Schaaf 1992, Heppell et al. 2006) may result in a decrease

in circulating androgen levels, a change in behavior, and a decrease in sperm production levels or quality. This would then indirectly impact reproductive success and the long term productivity of the population.

The objective of this study was to conduct a preliminary comparative analysis of life history, spawning behavior, morphology and physiology across four species of grouper. Results are considered in light of similar correlations seen in other fish species and in terms of the implications spawning life history strategy might have on the vulnerability to overexploitation of different grouper species. This work follows on modeling efforts to explore the impact of fishing on protogynous species and evaluation of potential management strategies for sex ratio recovery (Heppell et al. 2006).

METHODS

Samples (blood and gonad) from male gag (*Mycteroperca microlepis*) were collected between December 1995 and March 1996 between Jacksonville, FL and Cape Canaveral, FL. Nassau grouper (*Epinephelus striatus*) and red hind (*E. guttatus*) samples were collected in early February, 1996 near the spawning aggregation on Glover's Reef, Belize. All samples were collected from fish caught in the course of commercial fishing operations. Androgen and gonadosomatic index (GSI) data for red grouper (*E. morio*) were taken from Johnson (1995).

Gonads were fixed, embedded, and then sectioned on a rotary microtome. Sections were H&E stained, and staged at 100X magnification on a light microscope (Jackson and Sullivan 1995; Heppell and Sullivan 2000).

Blood was collected to measure circulating levels of androgens. Testosterone (T) and 11-ketotestosterone (11KT) were determined following the protocols of Woods and Sullivan (1993) and Heppell and Sullivan (2000). Data were analyzed by ANOVA followed by Tukey's Honest Significant Difference (HSD) Test at an alpha level of 0.05.

Published data on gonadosomatic index, spawning style, and sex ratio were assembled from the literature and author observations.

RESULTS

Males of all four species were at similar stages of gonadal development (data not shown). Plasma T levels showed a significant difference between the four species (ANOVA $p < 0.05$), and further analysis showed a slight elevation in T for *E. guttatus* relative to *E. striatus* ($p < 0.05$), but no significant differences were seen between the other species. Levels of 11KT also showed a significant difference among means between the four species ($p < 0.05$). 11-ketotestosterone levels were significantly lower in *E. striatus* than in the other three species ($p < 0.05$, Figure), but there were no differences in 11KT levels between the latter three species ($p > 0.05$).

Gonadosomatic indices (from published data) demonstrate that *E. striatus* has substantially larger testes than the other three species (Hood and Schleider 1992, Tucker et al. 1993, Sadovy et al. 1994, Sadovy and Colin 1995, Johnson 1995, Collins et al. 1998, Alan Collins, NOAA Fisheries, unpublished). *Epinephelus striatus* is a documented group spawner while the other three species are territorial

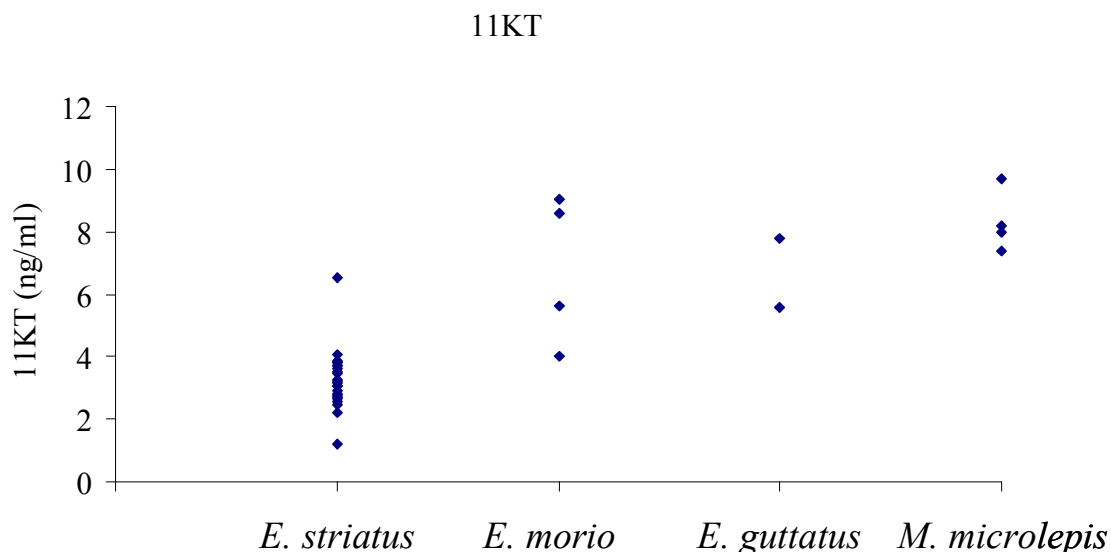


Figure 1. 11-ketotestosterone levels in four species of grouper.

(Coleman et al. 1996 and references therein). Sex ratios are more skewed towards females in the territorial species than in the group spawning *E. striatus* (data not shown).

DISCUSSION

Reproductive strategies evolve to maximize the reproductive fitness of the individual, and comparative life history analysis shows that there is more than one strategy to solve the fitness equation. Male *M. microlepis* defend territories and display aggressive behavior within the confines of a localized spawning aggregation (Gilmore and Jones 1992). Male *E. guttatus* cluster into groups of several females and a male (Shapiro et al. 1993), and males actively defend territories while on the spawning aggregation (Colin et al. 1987). *Epinephelus morio* aggregates in small polygynous groups (Sadovy et al. 1994). While the data are less clear for *E. morio* (Sadovy et al. 1994, Coleman et al. 1996), these observations are more in line with behaviors for *M. microlepis*, and *E. guttatus* than for *E. striatus*, which is a non-territorial group spawning species (Sadovy and Eklund 1999).

Epinephelus striatus males have substantially larger testes than any of the other three groupers examined here (Hood and Schleider 1992, Tucker et al. 1993, Sadovy et al. 1994, Sadovy and Colin 1995, Johnson 1995, Collins et al. 1998, Alan Collins, NOAA Fisheries, unpublished). An increase in gonad size allows production of larger quantities of gametes, which potentially allows a male to swamp his competitors' sperm in mass spawning events. Evaluation of GSI has been suggested as a means of identifying the occurrence of sperm competition in fish (Sadovy et al. 1994). On the other hand, *E. guttatus*, *E. morio*, and *M. microlepis*, all have GSI values that rarely exceed 2%, suggesting sperm competition is less important for these species. In these three species males compete to establish and defend territories and therefore monopolize access to females, potentially limiting the importance of sperm competition (Taborsky 1998).

Epinephelus striatus males have significantly lower levels of 11KT than male *E. guttatus*, *E. morio*, or *M. microlepis*, while T levels differ only between *E. striatus* and *E. guttatus*. This is consistent with what has been observed within other species, where 11KT levels are correlated with male reproductive tactics in fish while the pattern of T levels between male reproductive types is more variable (Brantley et al. 1993).

These results demonstrate, for the first time, that the relationship between androgens, morphology, and male reproductive tactics holds between closely-related species with different spawning behaviors. *Epinephelus striatus*, *E. guttatus*, and *M. microlepis* all aggregate to spawn (and *E. morio* may do so), while male *E. striatus* are the only one of the four species that do not establish or defend territories while spawning. This difference in reproductive behavior is correlated with significantly elevated levels of 11KT in the plasma of males from territorial species, and

with substantial differences in GSI between the reproductive types.

Finally, sex ratios differ between *E. striatus* and the other three species. Sex ratios are naturally skewed in favor of females for *E. guttatus*, *E. morio* and *M. microlepis*, while the sex ratio in *E. striatus* approaches unity or skews slightly towards males. A female skew is in part a function of protogynous reproductive development in the first three species. Problems arise because this female-skewed sex ratio can become significantly amplified in the presence of increased mortality through fishing (Huntsman and Schaaf. 1992, Coleman et al. 1996, Heppell et al. 2006), and there exists the potential for sperm limitation in these populations. Furthermore, a disruption in the number (and therefore density) of males setting up territories on spawning aggregations may remove the androgen positive feedback loop because of decreased intra-specific aggressive interactions. This could subsequently lead to adverse affects on sperm development and maturation. Ultimately this has the potential to reduce reproductive capacity below that which would be predicted simply based on estimates of spawning stock biomass.

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