Potential of Grouper Hybrid
(Epinephelus fuscoguttatus × E. polyplekadion)
for Aquaculture

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Abstract
The marine grouper species are considered high value food fish in several countries. However, controlled breeding and hatchery production of grouper fingerlings for commercial farming is still in its infancy. Investigations on the growth performance of the brown marbled grouper Epinephelus fuscoguttatus (Forskal), camouflage grouper E. polyplekadion (Bleeker) and their hybrid (E. fuscoguttatus × E. polyplekadion) under hatchery and growout culture conditions indicate the potential of grouper hybrids for aquaculture.

Introduction
The marine grouper has a high market value in several countries, with the demand being greater than the supply from capture fisheries. Despite intensive research efforts, controlled breeding and hatchery production of grouper fingerlings for commercial farming is still in the early stages of development because of low larval survival in the hatchery and other such constraints. Groupers are conventionally farmed in sea cages or in earthen ponds. Because of the closing of a large number of shrimp farms due to disease and environmental problems in recent years, many farmers are trying to switch from marine shrimp farming to marine finfish farming and the grouper has received attention because of its high market potential. However, expansion of commercial grouper farming is constrained by the shortage of fingerlings of the desired grouper species either from the wild or from hatcheries. The majority of the farmers still get fingerlings collected from the wild due to constraints faced in hatchery production. This has encouraged many countries to initiate research on grouper breeding and larval rearing.

Although studies on breeding of groupers in captivity were initiated during the 1960s and 1970s in Japan (Ukawa et al. 1966), Kuwait (Hussain et al. 1975) and Singapore (Chen et al. 1977), reports on the successful hybridization of groupers are scarce. The only recorded hybridization of the grouper was achieved in Hong Kong between cultured female white-spotted green grouper E. amblycephalus and wild male red grouper E. australis (Tseng and Poon 1983). This attempt was made to obtain fast growing red grouper hybrids since red groupers are slow growing compared to white-spotted green groupers but fetch a high market value.

Rationale for Hybridization
Genetic improvement of fish for aquaculture is important for developing disease resistant, temperature and salinity tolerant fish species and strains with a high growth rate. Fish breeding programs frequently include crossbreeding between species/strains or lines to utilize the beneficial effects of the parent stocks. Basic areas of work include traditional fish breeding and quantitative genetic approaches, as well as more innovative techniques involving chromosomal manipulations, physiological alteration of normal sex determination to produce monosex populations, gene transfer, molecular discrimination and gene mapping. It is considered desirable to cross breed grouper species with a view to developing strains with enhanced performance under hatchery, nursery and growout conditions, for commercial farming.

Information on the intraspecific spawning/mating behavior among grouper populations is scarce. The taxonomy of grouper species is quite confused because of the similarity of the color pattern of some species and ontogenetic changes and other variations in color patterns (Heemstra and Randall 1993). The color patterns of most groupers are usually distinctive enough to identify the particular species. But there are also intraspecific variations in color pattern for each species. The camouflage grouper E. polyplekadion has often been confused with the brown marbled grouper E. fuscoguttatus which has a similar color pattern of irregular dark blotches superimposed on numerous small dark brown spots and a black saddle blotch on the peduncle (Al-Thobaiti and James 1996). These species are frequently distributed along the Red Sea coast. Productive studies undertaken at the Fish Farming Centre in the Kingdom of Saudi Arabia indicate that the brown marbled grouper grows

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significantly faster than the camouflage grouper under hatchery and growout culture conditions (James et al., in press). However, the camouflage grouper is more resistant to environmental stress and disease compared to the brown marbled grouper, especially under hatchery conditions (personal observation). Considering these factors and the high occurrence of these species along the Red Sea coastal waters, it was deemed necessary to cross breed these two species to obtain the beneficial effects of the parent stocks for enhancing the hatchery performance and the growth rate in culture operations. This paper is based on a study that investigated, for the first time, the growth performance of two species of groupers and their hybrid from hatchery to growout farming under hypersaline water conditions in the Middle East.

**Method**

The broodstock of *E. fuscoguttatus* and *E. polyphekadion* used for breeding were collected from the hypersaline waters (42% salinity) of the Red Sea coast near Jeddah using handlines and fish traps locally known as gargoor or soquwa during 1994-95. The individual size of broodstocks of *E. polyphekadion* ranged from 3-5 kg and that of *E. fuscoguttatus* ranged from 12-16 kg. Breeding and larval rearing of these species were carried out using the method described by James et al. (1997). For cross breeding purposes, large size males (about 5 kg) of *E. polyphekadion* and females (about 12 kg) of *E. fuscoguttatus* were used. These species exhibited a typical lunar spawning rhythm as natural spawning occurs once in a month in the Red Sea. For cross breeding, the females of *E. fuscoguttatus* and the males of *E. polyphekadion* were maintained separately in fiberglass tanks of 25 m³ capacity with a water depth of 2 m at a stocking rate of 1-2 kg/m³. The water in the tanks was changed at about 50% turnover in a day. The broodstocks were fed daily to satiation with sardines enriched with cod liver oil by injecting 1 ml of the commercial product ‘Seven Seas’ into the gut of each sardine before offering them as feed for the broodstock. After confirmation of the gonadal development in females, male fish were introduced in the tank two days before the new moon phase. The spawning took place immediately after the new moon and lasted for three days. It was possible to obtain about 700 000-800 000 eggs per fish per spawning. About 71% of the eggs obtained from each spawning were good quality buoyant eggs. Fertilization was up to 96% and hatching averaged 80%.

After nursing *E. fuscoguttatus* and the hybrid for 90 days and *E. polyphekadion* for 120 days, investigations to assess growth of the two species and the hybrid under land-based growout culture conditions were undertaken using round fiberglass outdoor tanks (3 m diameter x 1.4 m depth x 10 m³ working volume). The water was changed about five times per day in the flow-through tank culture system. For comparative growth analysis, the data obtained from a stocking density of 15 fish/m³ in replicate was used. Locally formulated 4.5-6 mm size moist pellet feed with 40.9% crude protein (James et al. 1998) was used for feeding in the morning and trash fish, composed mainly of *Herklotsiichthys* spp., was used for feeding in the evening. The feed ration amounted to 5% of body weight during the first three months, 3% during the 4-6 months of the culture period and 2% from 7 months onwards until harvest. The fish were sampled monthly for growth monitoring.

The water quality was monitored daily. The water temperature ranged from 28-29°C and salinity 42-43%.

**Results**

**HATCHERY**

The size of the newly hatched larvae of the hybrid (*E. fuscoguttatus* female x *E. polyphekadion* male) ranged from 1.67-1.81 mm, which is smaller than that of *E. fuscoguttatus* (1.83-1.93 mm) and larger than that of *E. polyphekadion* (1.55-1.71). Although the newly hatched larvae of *E. polyphekadion* were smaller in size compared to those of *E. fuscoguttatus* and the hybrid, they grew rapidly to the size of others, within the first day after hatching. No significant difference (P>0.05) in growth was observed during the early part of the larval rearing up to day 20 between the
grouper hybrid and the parent fish species (Fig. 1). The hybrid and *E. fuscoguttatus* larvae grew faster compared to *E. polyphemus* 25 days after larval rearing period. On day 35, the size of the hybrid averaged 21.50±0.629 mm of *E. fuscoguttatus* 18.80±1.578 and of *E. polyphemus* 15.80±1.166 mm. After 35 days of rearing, growth of the hybrid was significantly faster (P<0.01) compared to the two parents. Also during this period (after day 35) the growth of *E. fuscoguttatus* larvae was significantly faster (P<0.01) than that of *E. polyphemus*. It was possible to obtain about 3 cm size grouper hybrid fingerlings for stocking in the nursery facility at the end of 40 days of larval rearing, whereas it took 45 days in the case of *E. fuscoguttatus* and 50 days for *E. polyphemus*. The fast growth and metamorphosis of the hybrid compared to the parents and other grouper species (Hussain and Higuchi 1980; Maneewong et al. 1986; Al-Abdul-Elah et al. 1996) is of great advantage for commercial applications since it could save considerable space, time and manpower utilization.

**GROWOUT**

Under similar growout culture conditions, a significantly faster growth rate (P<0.01) was observed in *E. fuscoguttatus* and in the hybrid compared to that of *E. polyphemus* (Figs. 2A-C). The difference in the hybrid and *E. fuscoguttatus* was not statistically significant (P>0.05) during the first five months of the growout culture period (Fig. 3). However, the higher growth observed in the case of the hybrid as compared with that of *E. fuscoguttatus* during the sixth month of the culture period was statistically significant (P<0.05). Highly significant difference in growth rate (P<0.01) of the hybrid compared to that of *E. fuscoguttatus* was observed on the seventh month of the growout. It was possible to

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*Fig. 2. Farm raised brown marbled grouper *E. fuscoguttatus* (A), typical hybrid (*E. fuscoguttatus* x *E. polyphemus*) (B), and camouflage grouper *E. polyphemus* (C).*
achieve marketable size hybrid grouper (mean size 695.8±15.20 g) and *E. fuscoguttatus* (mean size 598.0±36.00 g) at the end of the seven month growout period. Whereas, it took about 12 months of rearing for *E. polycephalus* to attain a marketable size of 529.4±28.64 g. Irrespective of initial stocking size, an average daily growth rate of 3.02 g/fish/day was observed in the grouper hybrid, whereas the daily growth rate averaged 2.34 g/fish/day for *E. fuscoguttatus* and 1.31 g/fish/day for *E. polycephalus*. The growth rate of the hybrid observed in this tank culture system is higher than that observed by Tacon et al. (1991) for *E. tauvina* (2.87 g/fish/day) reared in floating net cages. The growth rate achieved for *E. polycephalus* during this investigation is also higher than the growth rate (1.13 g/fish/day) observed for *E. microdon* in French Polynesia (AQUACOP et al. 1990). Previous investigations also indicated that the growth rate of groupers is species specific and depends on the culture conditions (Chua and Teng 1979; Lee 1982; Sakares and Sukhanteang 1985; Sakares and Kumpang 1988; Kohno et al. 1989; Tacon et al. 1991; Chao et al. 1993). The food conversion ratio (FCR) in hybrid (average 1.39±0.04) was significantly higher (P<0.05) as compared to *E. fuscoguttatus* (average 1.49±0.25) and *E. polycephalus* (2.9±0.85) indicating efficient utilization of feed by the hybrid. This implies a reduction in the feed costs for commercial farming.

Wide size variations among farmed grouper species under growout culture conditions are very common. It is interesting to note the occurrence of two phenotypes among the grouper hybrid (Fig. 4). The small-sized fish among the hybrid had a color pattern similar to *E. polycephalus*, whereas the large-sized individuals were similar to that of *E. fuscoguttatus*. However, the grouper hybrid differed from their parent stock in several taxonomic and meristic characteristics. The identifying characteristics of the hybrid compared to that of their parent stock are summarized in Table 1.

### Conclusion

Grouper hybrids have demonstrated better growth under growout conditions. However, the techniques developed here for the grouper hybrid (*E. fuscoguttatus* × *E. polycephalus*) should be further improved for breeding and selection of fast growing grouper hybrid traits for commercial applications. The maintenance of pure genetic lines is easier for the present grouper hybrid crosses since the physical appearance and meristic characteristics allow hatchery workers to determine easily the presence of contaminated broodstock in the culture facilities. Further research efforts could be directed towards obtaining more information on the visual and chemical cues which determine spawning compatibility between species as well as on other factors influencing the frequency with which two grouper species hybridize. Studies are needed to determine which hybrid crosses respond best to feeding and have higher growth rates and disease resistance so that there are better choices of grouper hybrids for commercial rearing.
References


James, C.M., S.A. Al-Thobaiti, B.M. Rasem and M.H. Carlos. Comparative growth of brown-marbled grouper Epinephelus fuscoguttatus (Forsskål) and camouflage grouper E. polyphekadion (Bleeker) under hatchery and growout culture conditions. Asian Fish. Sci. (In press).


Table 1. Differences in taxonomic characteristics of parent and hybrid groupers.

<table>
<thead>
<tr>
<th>Taxonomic characteristics</th>
<th>Parent stock</th>
<th>Grouper hybrid phenotype</th>
</tr>
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<tbody>
<tr>
<td>E. fuscoguttatus (female)</td>
<td>10-12</td>
<td>14</td>
</tr>
<tr>
<td>E. polyphekadion (male)</td>
<td>17-21</td>
<td>16-17</td>
</tr>
<tr>
<td>Pectoral fin-ray counts</td>
<td>18-20</td>
<td>20-23</td>
</tr>
<tr>
<td>Number of pyloric caeca</td>
<td>20-30</td>
<td>14*</td>
</tr>
</tbody>
</table>

*14 main caeca each branch out to 13-14 minor caeca.

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