Deformity of agglutinated pelvic fin membrane in hatchery-reared black rockfish Sebastes inermis and its application for stock separation study

Tomoya Murakami,1,2* Satoshi Aida,1 Kouji Yoshioka,2a Tetsuya Umino2 and Heisuke Nakagawa2

1Hiroshima Prefectural Fisheries Experimental Station, Aki, Hiroshima 737-1207 and 2Graduate School of Biosphere Science, Hiroshima University, Higashi-hiroshima, Hiroshima 739-8528, Japan

ABSTRACT: This study is the first to report on the high occurrence of agglutinated pelvic fin membrane deformities in hatchery-reared black rockfish Sebastes inermis. For 5 years, this symptom was marked in hatchery-reared fish, with 58.7% of fish deformed on average (varying between 46.7 and 72.0%). The deformity was a peculiarity in hatchery-reared fish, but is negated in wild fish, and was not related to whether the fish broodstock originated from the wild or from a hatchery. Mark–release experiments showed that deformed fish were almost the same as normal hatchery-reared fish in growth and survival rates, and, theoretically, the recapture numbers of hatchery-stocked fish, estimated by deformity, almost coincided with actual recapture numbers, confirmed by otolith tagging. The results of the present study indicate that deformity in hatchery-reared black rockfish is useful as a stock separation tool.

KEY WORDS: black rockfish, deformity, otolith tagging, pelvic fin, stock separation.

INTRODUCTION

Since Blaxter took up the study of morphological differences between hatchery-reared and wild fish, much attention has been paid to the external appearance of hatchery-reared fish.1 Crowding in hatchery-reared conditions, deformity and abnormality have occurred among some species. For example, deformities of the pectoral fin and internostril epidermis have been reported in red sea bream Pagrus major.2,3 The latter deformity also occurred in black sea bream Acanthopagrus schlegeli, although its occurrence was much lower than in red sea bream.4 Abnormal eye location and pigmentation, such as ambicoloration and albinism, have also been documented in Japanese flounder Paralichthys olivaceus.5,6

The black rockfish Sebastes inermis is widely distributed along the coast of southern Hokkaido south to Kyushu and the southern coasts of the Korean Peninsula, where it feeds on small fishes and marine invertebrates in rocks and Zostera areas.8,9 This species is an important component of commercial fisheries and is popular in recreational angling in the Seto Inland Sea. The Hiroshima Prefectural Fisheries Experimental Station (HPFES) has produced the black rockfish since 1995,10 corresponding with stock enhancement. In the present study, hatchery-reared black rockfish were found to have a high annual occurrence of deformities of the pelvic fin.

Consequently, the purposes of the present study were twofold. First, the deformity of the agglutinated pelvic fin membrane was clarified. Second, the influence of the deformity on the growth and survival, and the usefulness of the deformity as a stock separation tool to discriminate by appearance hatchery-stocked fish from wild populations after stocking were evaluated.

MATERIALS AND METHODS

Definition of the deformity and its occurrence

The feeding schedule for seed production was as follows. The viviparous larvae were fed rotifers...
(0–50 days after extrusion), *Artemia* nauplii enriched with n-3 highly unsaturated fatty acids (15–100 days after extrusion), wild copepods (30–100 days after extrusion) and formula feed (more than 40 days after extrusion). Most of them were accommodated at a density of approximately 4000–6000 larvae/m³ initially and raised in 25-m³ octagonal concrete tanks for approximately 3 months at 10–18°C. The survival rate of seed production from 1995 to 2001 ranged between 33.9 and 73.6%. After that, they were kept in 5 m × 5 m × 5 m net pens until they were 1–6 years of age. To define the deformed type of pelvic fins, it was investigated preliminarily that 5-year-old fish originated from wild broodstock in January 1997. Based on the definition of the deformity, it was further investigated that the deformed type occurred among the hatchery-reared progeny produced between 1995 and 2001, except in 1996 and 1999 (Table 1).

**Mark–release experiment**

To compare the deformed fish with the normal hatchery-reared fish by growth and survival and to assess the usefulness of the deformity as a stock separation tool between hatchery-stocked and wild fish, a mark–release experiment was conducted. Black rockfish larvae (173 700) were born in January 2002 from 4- and 5-year-old hatchery-reared broodstocks. They had been raised in HPFES’ 25-m³ octagonal concrete tanks for 104 days after extrusion, with the above feeding schedule, at 10–18°C. The survival rate was 47.1%. Prior to stocking, all hatchery-stocked fish were marked by otolith tagging and the occurrence of deformity was also verified microscopically (n = 100). After stocking, the numbers of recaptured hatchery-stocked fish were estimated using the following two methods: (i) the actual number by tracing otolith tagging; and (ii) the theoretical number by deformity ((number of deformed fish captured/percentage of occurrence of deformity prior to stocking) multiplied by 100).

For otolith tagging, fish were immersed in a 40-mg/L solution of a florescent substance, alizarin complexone (Dojin Co. Ltd, Kumamoto, Japan), for 24 h at 15°C, which resulted in a daily florescent ring on the otolith detectable under ultraviolet light. Otolith tagging was 100% successful and the survival rate during the treatments was 98.9%. The otolith tagging remained and was detected for at least 3 years after tagging (T Murakami, unpubl. data, 2003).

**Release and recapture**

After otolith tagging, 81 000 fish (total length = 28.6 ± 2.9 mm [mean ± standard deviation]) were transported by boat and then released into the coastal area of Ikuno Island on 22 April 2002 (Fig. 1). The study area was 5–10 m deep and was occupied by the eelgrass *Zostera marina*. Recapture trials of stocked fish were carried out using a beam trawl 2.0 m wide and 0.3 m high with a mesh aperture of 4.2 mm and a towing speed of 0.3–0.5 m/s from 23 April to 20 October 2002. The age of the specimens was judged by year-ring on the sagittal otolith.11 Zero-year-old black rockfish was targeted for stock separation by tracing the deformity and otolith tagging.

**Statistical analysis**

For comparison between the theoretical number by deformity and the actual number by otolith tagging and between the percentage of occurrence of deformity prior to stocking and those of each sampling date, data were subjected to a $\chi^2$ test or Fisher’s exact probability test if the expectation was £5. The Mann–Whitney $U$-test was used for comparison of growth after stocking between normal

---

**Table 1** Percentage of occurrence of each type of deformed pelvic fin in hatchery-reared black rockfish

<table>
<thead>
<tr>
<th>Year produced</th>
<th>Age (years)</th>
<th>$TL$ (cm) Mean ± SD</th>
<th>No. specimens</th>
<th>Deformity (%)</th>
<th>Broodstock origin</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Type 1†</td>
<td>Type 2‡</td>
</tr>
<tr>
<td>1995</td>
<td>6</td>
<td>22.4 ± 1.4</td>
<td>169</td>
<td>16.0</td>
<td>30.8</td>
</tr>
<tr>
<td>1997</td>
<td>5</td>
<td>21.4 ± 1.3</td>
<td>100</td>
<td>12.0</td>
<td>62.0</td>
</tr>
<tr>
<td>1998</td>
<td>4</td>
<td>20.2 ± 1.0</td>
<td>100</td>
<td>14.0</td>
<td>39.0</td>
</tr>
<tr>
<td>2000</td>
<td>2</td>
<td>16.9 ± 1.4</td>
<td>103</td>
<td>18.4</td>
<td>37.9</td>
</tr>
<tr>
<td>2001</td>
<td>1</td>
<td>12.6 ± 1.3</td>
<td>100</td>
<td>19.0</td>
<td>53.0</td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td></td>
<td>572</td>
<td>15.9</td>
<td>42.8</td>
</tr>
</tbody>
</table>

†Perfectly agglutinated pelvic fins as shown in Fig. 2a.
‡Imperfectly agglutinated pelvic fins as shown in Fig. 2b.
SD, standard deviation; $TL$, total length.
Deformity and stock separation in rockfish

FISHERIES SCIENCE

841

fish and deformed fish in recaptured hatchery-reared fish. The correlation coefficient was evaluated by Student’s t-test. The criterion for statistical significance was \( P < 0.05 \).

RESULTS AND DISCUSSION

State of deformity

The deformity types were categorized into two groups compared with wild fish (Fig. 2). Type 1 was an especially remarkable deformity in which the pelvic fin membranes were perfectly agglutinated. In the type 2 deformity, the degree of agglutinated fin membranes was imperfect compared with that in type 1. These two types of pelvic fins had one spine and five soft rays as with the wild fish.\(^8\) Deformities were not observed in the 100 wild fish collected from the Seto Inland Sea.

Based on these definitions of deformity, the percentages of occurrence of type 1 and type 2 deformities among the fish produced between 1995 and 2001, except in 1996 and 1999, are summarized in Table 1. Type 1 deformity appeared in nearly 15% of stock in each year, but type 2 deformity fluctuated between 30.8 and 62.0%. Consequently, the total percentage of the occurrence of type 1 and type 2 deformities ranged from 46.7 to 74.0%, with an average of 58.7%.

Similar deformities, including the lack or dwarfism of the ventral fin, have been reported in the red sea bream, but the cause is unknown.\(^12\) In the present study, it is noteworthy that each deformity rate of seed production from 1995 to 2001 ranged between 46.7 and 74.0%. The relationship between the percentage of occurrence of deformity in each progeny and their broodstock origin was not significant. This suggests that the deformity is caused by environmental factors in the hatchery-reared conditions rather than by parental effect. Further studies are needed to investigate the cause of the deformity.

---

Fig. 1  Release and sampling sites, Ikuno Island, Japan. (*) Release point; (dotted area) sampling area by beam trawl.

Fig. 2  The type of the deformity of agglutinated pelvic fin membrane in 5-year-old hatchery-reared black rockfish. Bar = 1 cm. (a) Perfectly agglutinated pelvic fins (type 1); (b) imperfectly agglutinated pelvic fins (type 2); (c) normal pelvic fins.
Table 2  Results of stock separation between hatchery-stocked and wild black rockfish using deformity and otolith tagging

<table>
<thead>
<tr>
<th>Trial</th>
<th>Days after stocking</th>
<th>Total</th>
<th>Otolith tagging</th>
<th>Deformity</th>
<th>P-value&lt;sup&gt;†&lt;/sup&gt;</th>
<th>Deformity rate (%)&lt;sup&gt;‡&lt;/sup&gt;</th>
<th>Growth of hatchery-stocked fish (with ALC)</th>
<th>TL (mm), mean ± SD</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Hatchery (with ALC)</td>
<td></td>
<td>Hatchery</td>
<td>Wild</td>
<td>Deformed</td>
<td>Normal</td>
</tr>
<tr>
<td>1</td>
<td>1</td>
<td>121</td>
<td>74</td>
<td>46</td>
<td>120</td>
<td>0</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>9</td>
<td>104</td>
<td>42</td>
<td>41</td>
<td>83</td>
<td>0</td>
<td>21</td>
<td>21</td>
</tr>
<tr>
<td>3</td>
<td>18</td>
<td>87</td>
<td>35</td>
<td>28</td>
<td>63</td>
<td>0</td>
<td>24</td>
<td>24</td>
</tr>
<tr>
<td>4</td>
<td>30</td>
<td>64</td>
<td>24</td>
<td>12</td>
<td>36</td>
<td>0</td>
<td>28</td>
<td>28</td>
</tr>
<tr>
<td>5</td>
<td>60</td>
<td>55</td>
<td>15</td>
<td>5</td>
<td>20</td>
<td>0</td>
<td>35</td>
<td>35</td>
</tr>
<tr>
<td>6</td>
<td>88</td>
<td>32</td>
<td>5</td>
<td>4</td>
<td>9</td>
<td>0</td>
<td>23</td>
<td>23</td>
</tr>
<tr>
<td>7</td>
<td>120</td>
<td>32</td>
<td>8</td>
<td>8</td>
<td>16</td>
<td>0</td>
<td>16</td>
<td>16</td>
</tr>
<tr>
<td>Total</td>
<td>495</td>
<td>203</td>
<td>144</td>
<td>347</td>
<td>0</td>
<td>148</td>
<td>148</td>
<td></td>
</tr>
</tbody>
</table>

*Number of stock separated significantly different between hatchery-stocked fish and wild fish by otolith tagging (P<0.05); **significant difference between deformed fish and normal fish (P<0.05).

†Otolith tagging was 100% successful, so it was assumed that the number of stock separated between hatchery-stocked fish and wild fish by otolith tagging was the actual number.

‡Results were theoretically estimated by deformity rate prior to stocking (63.0%).

§(Number of deformed fish/number of otolith tagged fish [i.e. actual number of hatchery-stocked fish]) multiplied by 100.

*Comparison between the deformity rate prior to stocking and those of each sampling date.

††Fisher's exact probability test.

ALC, alizarin complexone; TL, total length; SD, standard deviation.
Growth, survival and usefulness of the deformity as a stock separation tool

Based on the mark–release experiment, 495 wild and hatchery-stocked juveniles were captured during seven trials. Two hundred and three of the total number of captured fish had a deformity (Table 2).}

In preliminary investigations prior to stocking, the occurrence of the deformity was 63.0%. Afterward, it was confirmed that the percentage of the occurrence of deformity had been almost a constant value (60%) from 3 to 5 cm in total length in a terrestrial tank (T Murakami, unpubl. data, 2003). Therefore, the theoretical number of hatchery-stocked fish judged by the deformity can be estimated in each sampling date. In contrast, hatchery-stocked and wild fish were discriminated by otolith tagging. Otolith tagging was perfectly successful prior to stocking. Hence, the results for stock separation by otolith tagging can be attributed to the actual numbers of hatchery-stocked and wild fish.

When the results of stock separation by the two methods were compared, the theoretical numbers coincided with the actual numbers ($\chi^2$ test), except at 9 days after stocking (Table 2). Consequently, the high occurrence of deformity in the pelvic fins of black rockfish can be useful as a stock separation tool, as can deformities of the pectoral fin (51.0–97.7%) and inter-nostril epidermis (61.4–90.5%) in red sea bream.3 This view also supports the significant linear relationship ($n = 7$, $r = 0.988$, $P < 0.001$) between the actual numbers and the theoretical numbers of hatchery-stocked fish as shown in Fig. 3.

However, there is a possibility that the pelvic fin deformity affects the growth and selectivity between normal fish and deformed fish for recapture-trawl or survival rate after stocking, through the reduction of swimming activity and escaping speed against predators. If the deformity did have such negative effects, differences in growth between the normal and deformed fish and in percentages of the occurrence of the deformity pre- and post-stocking should be evident. Table 2 shows that there was no significant difference in growth between the normal and deformed fish, except that the total length of the deformed fish was significantly larger than that of the normal fish at 120 days after stocking. Table 2 also shows the percentages of the occurrence of recaptured hatchery-stocked fish, given as (number of deformed fish/number of otolith-tagged fish [i.e. actual number of hatchery-stocked fish]) multiplied by 100. Results show that the percentage of the occurrence of each sampling date ranged from 50.0 to 75.0%, with an average of 58.5%. These values were not significantly different, with 63.0% prior to release ($\chi^2$ test or Fisher’s exact probability test, $P > 0.3$). Therefore, it was confirmed that the deformity had no influence on the growth and survival after stocking during the research period.

In conclusion, the deformity of agglutinated pelvic fins has a negligible influence on the growth and survival after stocking, and has potential as a stock separation tool to assess the stocking efficiency of hatchery-stocked black rockfish. Further research is needed to investigate the cause and influence of the deformity before the fish become adults.

ACKNOWLEDGMENTS

The authors would like to thank Emeritus Professor Fumio Yamazaki, Hokkaido University, for his valuable suggestions. We also express our sincere gratitude to staff members of the Hiroshima Prefectural Fisheries Experimental Station for their continuous assistance. This study was supported in part by a Grant-in-Aid from the Ministry of Agriculture, Forestry and Fisheries, Japan.

REFERENCES


