INTENSITY OF TREMATODE METACERCARIAE IN CYPRINOID FISH IN CHIANG MAL PROVINCE, NORTHERN THAILAND

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Abstract. The intensity of infection of trematode metacercariae was determined in five species of cyprinoid fish collected from Mae Ngud reservoir, Chiang Mai Province. These species were Thynnichthys thynnoides, Puntioplites proctozysron, Hampala macrolepidota, Puntius leiacanthus and Puntius gonionotus. T. thynnoides contained the highest number (83.0%) of metacercariae, whereas P. gonionotus had the fewest (0.19%). The caudal fin was the area of highest infection of metacercariae (49.39%), with the anal fin having the fewest (5.93%). Most metacercariae were isolated from Haplorchis taichui (63.27%), with an intensity variation of 0.3-165.2 metacercariae/fish.

INTRODUCTION

Trematode infections are the common helminthic infections in humans residing in northern Thailand (Radomyos et al, 1994; 1998; Pungpak et al, 1998). Haplorchis taichui (the heterophyid fluke of the Heterophyidae family) and Opisthorchis viverrini (the liver fluke of the Ophisthorchiidae family) were the most common flukes recovered from northern Thai patients treated with praziquantel. The prevalence of the former species found at 6 times more than that of the latter (Radomyos et al, 1998). Humans acquire fluke infection by ingesting raw and/or undercooked cyprinoid fish that harbor metacercariae. The most famous local fish dishes for Thai northerners are “Lab Pla” and “Pla Som” (WHO, 1995). The former dish is traditionally prepared by mixing the raw-chopped cyprinoid fish with spices, while the latter is a salted semi-fermented fish dish. The most common cyprinoid fish for both local dishes is Thynnichthys thynnoides (Pla Soy). Sukontason et al (1998) indicated that H. taichui was the only trematode metacercariae found in Lab Pla.

Recent fish surveys revealed that most trematode metacercariae were heterophyid flukes from several areas in northern Thailand and fish farms (Khamboonruang et al, 1997; Waikagul, 1998; Sukontason et al, 1999). The region where metacercariae infected cyprinoid fish varied according to the study area and species of fish (Komalamisra and Setasuban, 1989; Namue et al, 1998). Since there is no report concerning the distribution of metacercariae in northern Thai fish, the present investigation was undertaken to determine the intensity and distribution of trematode metacercariae in natural cyprinoid fish collected from Mae Ngud reservoir in Chiang Mai. This area represents the most important source of fish for the inhabitants of the province.

MATERIALS AND METHODS

Cyprinoid fish were collected from Mae Ngud reservoir, Mae Tang district, Chiang Mai Province. Five species of fish (10 fish for each species) were investigated for trematode metacercariae: Thynnichthys thynnoides, Puntioplites proctozysron, Hampala macrolepidota, Puntius leiacanthus and Puntius gonionotus. The fish were rinsed using tap water until clean and the internal organs were removed. Each individual fish was dissected into 5 portions for determining the metacercariae (ie caudal fin and muscle, pectoral fin and muscle, ventral fin and muscle, dorsal fin and muscle, and anal fin and muscle). Each portion from the same kind of fish was pooled and then digested using acid pepsin solution [conc hydrochloric acid 1 ml: porcine pepsin (Sigma®: Germany) 1 g: 0.85% sodium chloride solution 99 ml] in a mixer/blender at a ratio of 1 g of fish: 10 ml acid pepsin solution. The digested material was transferred into a water bath shaker for 1 1/2 hours at 37°C and subsequently passed through 2 layers of wet gauze. The digested material was rinsed with 0.85% sodium chloride solution and examined for metacercariae with a stereomicroscope. The identification of trematode metacercariae was carried out by the morphological examination based on Scholz et al (1991), and the identification of sclerites on the ventrogenital sac of Haplorchis species was based on Pearson and Ow-Yang (1982).

RESULTS

A total of 3,136 metacercariae were isolated from the 5 species of cyprinoid fish examined (Table 1).
Among them, *T. thynnoides* harbored the highest number (83.0%: 2,603/3,136), whereas, *P. gonionotus* harbored the lowest (0.19%: 6/3,136). For the overall metacercariae isolated, the caudal fin was the area with the highest infection (49.39%: 1,549/3,136), with the anal fin being the lowest (5.93%: 186/3,136) (Table 1).

Since *T. thynnoides* harbored the highest number of metacercariae (83.0%), this species of fish was selected for determining the species of trematode metacercariae. *Haplorchis* spp was the predominant metacercariae found (75.45%: 1,964/2,603), with *H. taichui* being the highest found (63.27%: 1,647/2,603) (Table 2). The highest and lowest number of *H. taichui* metacercariae were localized in the caudal (53.36%: 1,389/2,603) and anal fm (4.69%: 122/2,603), respectively.

The intensity of *H. taichui* metacercariae varied considerably in the cyprinoid fish examined, with those in *T. thynnoides*, *P. proctozysron*, *H. macrolepidota*, *P. leiacanthus* and *P. gonionotus* having 165.2, 21.7, 9.5, 4.5 and 0.3 metacercariae per fish, respectively (Table 3).

**DISCUSSION**

The intensity of heterophyid metacercariae in cyprinoid fish collected from Mae Ngud reservoir was quite high, particular for *H. taichui*. An unidentified *Haplorchis* species as well as degenerated metacercariae were also found. This is probably due to underdeveloped worm and/or damage to the specimens during the process of metacercariae preparation. The metacercariae of *H. taichui* were isolated not only primarily from the *T. thynnoides* collected in this study, but also from *P. leiacanthus*, *P. gonionotus*, *P. proctozysron*, *H. macrolepidota*, *Danio regina*, *Aplocheilus panchax* and *Lariobarbus burmanicus* that were collected in other areas of Chiang Mai (Sukontason et al, 1999). According to Srisawangwong et al (1997), the metacercariae of *H. taichui* was predominant in 6 kinds of cyprinoid fish collected in Khon Kaen Province, northeastern Thailand, especially in *Hampala dispar*, which was similar to the study in Lao PDR (Ditrich et al, 1990). The study of Ooi et al (1997) in Taiwan also reported that 96.2% of *H. taichui* metacercariae were isolated from the freshwater fish, *Hemiculter leucisculus*. The metacercariae of *H. taichui*, therefore, showed broad ranges in host specificity, since it can live in several species of cyprinoid fish. In addition, the presence of this parasite in the definitive host also being found in several species of mammals (Faust and Nishigori, 1926; Pearson, 1964). For these reasons, this fluke is distributed easily in several geographical regions of Thailand, especially in the northeast and north. Due to the habit of people in the rural community of eating raw and/or improperly cooked freshwater fish, in combination with the broad ranges in host specificity, the occurrence of *H. taichui* is undoubtedly remains present in these regions of Thailand.

The present study showed that the areas most and least infected by metacercariae in fish were the caudal and anal fin, respectively, which resembled report by Tesana et al (1985) in that the distribution of *O. viverrini* metacercariae occurred abundantly in the caudal fin, but rarely in the anal fin. Vichasri et al
Table 2
Distribution of trematode metacercariae isolated from each separated organ of *Thynnichthys thynnoides* collected from Mae Ngud reservoir, Chiang Mai Province, northern Thailand.

<table>
<thead>
<tr>
<th>Species of metacercariae</th>
<th>Caudal fin</th>
<th>Pectoral fin</th>
<th>Ventral fin</th>
<th>Dorsal fin</th>
<th>Anal fin</th>
<th>Total (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Haplorchis</em> spp</td>
<td>1,028</td>
<td>396</td>
<td>309</td>
<td>144</td>
<td>87</td>
<td>1,964 (75.45)</td>
</tr>
<tr>
<td>- <em>H. taichui</em></td>
<td>838</td>
<td>350</td>
<td>260</td>
<td>125</td>
<td>74</td>
<td>1,647 (63.27)</td>
</tr>
<tr>
<td>- Unidentified <em>Haplorchis</em> spp</td>
<td>190</td>
<td>46</td>
<td>49</td>
<td>19</td>
<td>13</td>
<td>317 (12.18)</td>
</tr>
<tr>
<td>Degenerated metacercariae</td>
<td>361</td>
<td>125</td>
<td>99</td>
<td>19</td>
<td>35</td>
<td>639 (24.55)</td>
</tr>
<tr>
<td>Total</td>
<td>1,389</td>
<td>521</td>
<td>408</td>
<td>163</td>
<td>122</td>
<td>2,603 (100.00)</td>
</tr>
</tbody>
</table>

Table 3
Intensity of *Haplorchis taichui* metacercariae in cyprinoid fish collected from Mae Ngud reservoir, Chiang Mai Province, northern Thailand.

<table>
<thead>
<tr>
<th>Species of cyprinoid fish</th>
<th>No. fish examined</th>
<th>Intensity a</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Thynnichthys thynnoides</em></td>
<td>10 (302)</td>
<td>165.2 (59.2)</td>
</tr>
<tr>
<td><em>Puntioplites protozysron</em></td>
<td>6 (34)</td>
<td>21.7 (12.1)</td>
</tr>
<tr>
<td><em>Hampala macrolepidota</em></td>
<td>10 (10)</td>
<td>9.5 (9.5)</td>
</tr>
<tr>
<td><em>Puntius leiacanthus</em></td>
<td>8 (49)</td>
<td>4.5 (181.7)</td>
</tr>
<tr>
<td><em>Puntius gonionotus</em></td>
<td>20 (58)</td>
<td>0.3 (80.3)</td>
</tr>
</tbody>
</table>

aIntensity is given as the mean number of metacercariae per infected fish. Number in parentheses referred to Sukontason *et al* (1999).

This study showed a higher intensity of *H. taichui* in Chiang Mai Province than that of *O. viverrini* which was in accordance with recent studies in Chiang Mai (Poolpol, 1995; Sukontason *et al*, 1999) and Khon Kaen Province (Srisawangwong *et al*, 1997). However, it was in contrast to those of Sujuanun and Thitasut (1971) in Chiang Mai and Kamalamisra and Setasuban (1989) in Khon Kaen. Waikagul (1998) suggested that *O. viverrini* metacercariae was very high in the northeast region of Thailand, particularly in those reports before 1990, which contrasted to *H. taichui*. Up until now, the situation in both regions of Thailand regarding the intensity of *O. viverrini* and *H. taichui* has changed. Several factors may be involved in this change such as the existence of the first intermediate snail host, the second intermediate host of fish as well as the definitive host, usage of sanitary latrines, custom of eating raw and/or undercooked fresh water fish and the presence of infected reservoir hosts. Efforts should be made to determine those factors involving parasitic transmission in order to clarify the cycle of the parasite and eventually prevent humans from acquiring trematode infection.

**REFERENCES**


Haas W, Granzer M, Brockelman CR. *Opisthorchis viverrini*: Finding and recognition of the fish host (1982) also reported that the anal fin was the least infected area of *O. viverrini* metacercariae. Moreover, the metacercariae of *H. taichui* was observed mostly in the tail region of fish (Ooi *et al*, 1997). Since fish use the caudal fin in swimming more vigorous than other fins and the attachment of the fish host is stimulated by water current (Haas *et al*, 1990), these may be the reasons why cercariae can recognize and attach to the caudal fin more than others sites. Nevertheless, the site of attachment of *Haplorchis* metacercariae in this study was in contrast to the scales of fish reported by Namue *et al* (1998).


Ooi HK, Chen CI, Lin SC, Tung KC, Wang IS, Kamiya M. Metacercariae in fishes of Sun Moon Lake which is an endemic area for *Clonorchis sinensis* in Taiwan. *Southeast Asian J Trop Med Public Health* 1997;28(suppl 1):222-3.


