A Harpacticoid Copepod Parasitic in the Cultivated Brown Alga *Undaria pinnatifida* in Korea

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We have examined harpacticoid copepods inhabiting the phaeophyte Miyok, *Undaria pinnatifida* (Harvey), on the southern coast of Korea and cultured in the laboratory pieces of Miyok frond heavily infested by frond-mining nauplii to identify the harpacticoid copepod to which those nauplii belong. Of the harpacticoids found in Miyok-washings, only *Amenophia orientalis* Ho and Hong and an unidentified species of the genus *Scutellidium* occurred consistently in all Miyok samples examined. Many females of both species carried egg sacs. In the cultures the frond-mining nauplii developed, in 15 days at 15 °C, into copepodid stages or adults that were all identified with *Amenophia orientalis*. It is therefore concluded that this species is the causative agent for the so-called pinhole disease of the cultivated Miyok on the southern coast of Korea, which is diagnosed by the appearance on the thallus of numerous pinholes occupied by developing nauplii.

Introduction

The so-called pinhole disease of the cultivated Miyok (the brown alga *Undaria pinnatifida*) in Korea is a well-known plague inflicting a considerable damage to the Miyok farms in Korea. Because of the ever increasing demand for Miyok as a food item, Miyok cultivation in Korea has developed as a major mariculture industry in recent years. In the past few years, however, the industry has suffered greatly with the pinhole disease which is diagnosed by the appearance of numerous pinholes in the thallus of Miyok. Affected Miyok is therefore considered unfavorable for human consumption.

A scientific article reporting on the pinhole disease of Miyok was first published in Japan by Torii and Yamamoto (1975), who described a *Thalestris* sp. belonging to the harpacticoid copepod family Thalestridae as the causative organism. Subsequently, the same disease was reported from Miyok farms in Korea by Kang (1981), who also assumed

a *Thalestris* sp. as the causative agent, but no reliable evidence supporting his claim was presented. Recently, Ho and Hong (1988) described two new species of thalestrid harpacticoids from Miyok farms on the southwestern coast of Korea. As their species were collected from infested fronds, they considered them to be responsible for the infestation.

In studies on the harpacticoid copepods recovered in Miyok-washings, we have found not only those previously regarded as causative organisms for the pinhole disease but also several other harpacticoid species. One of which, *Scutellidium* sp., occurred consistently along with *Amenophia orientalis* Ho and Hong (1988) described as a causative agent.

Furthermore, *Parathalestris infestus* which Ho and Hong (1988) described as another causative species was found only occasionally. Therefore, we have reopened the question of what species is/are responsible for the Miyok pinhole disease in Korea.

Materials and Methods

For the studies of the harpacticoid fauna in Miyok beds, samples of heavily infested Miyok were collected from Miyok farms on the southern coast of Korea. Each sample (about 5 kg in wet weight), after draining excess water by suspending it in air about 20 seconds, was soaked in a 10% formalin. Later in the laboratory the preserved Miyok was washed to recover harpacticoids.

For the culture of harpacticoids, infested Mivok fronds were cut in pieces about 3×5 cm² containing about 100 pinholes occupied by developing nauplii. The pieces of fronds were individually cultured in covered petri dishes 10 cm in diameter at a constant temperature of $15\,^{\circ}$ C in an incubator. Water of the culture dishes was changed every other day by transferring the fronds as well as the free copepods to a new petri dish filled with filtered seawater. Protozoa were found on the frond throughtout the culture and their number seemed to increase with time. But no attempts were made to identify them or assess their abundance. The cultures were examined under dissecting microscope once every day, and one or two cultures were fixed in formalin for closer observations of the developing harpacticoids.

Results and Discussion

As shown in Table 1, Amenophia orientalis and Scutellidium sp. occurred consistently in all Miyok-washings, and these two species together comprised over 87% of the total number of harpacticoids found in every sample. Also found were about 10 other harpacticoids species, but none of them including Parathalestris infestus Ho and Hong (1988) originally described from Miyok beds on the south-western coast of Korea were common nor did they occur consistently in this study. Amenophia orientalis and Scutellidium sp., the two common species found in the study, were very characteristic by their bodies, which were highly depressed dorsoventrally while all other harpacticoid species found were cylindrical.

Table 1. Harpacticoids found in Miyok-washings

Location	Date of collection	Species found (% composition)			
Songjeong*	5/8/90	Amenophia orientalis	(49)		
		Scutellidium sp.	(43)		
		others	(8)		
-	5/24/90	Amenophia orientalis	(47)		
		Scutellidium sp.	(42)		
_		others	(11)		
	5/31/90	Amenophia orientalis	(33)		
		Scutellidium sp.	(61)		
		others	(6)		
Wando**	6/4/90	Amenophia orientalis	(54)		
		Scutellidium sp.	(33)		
		others	(13)		

^{*} on the southeastern coast of Korea

Table 2 shows the results of the culture experiment in which pieces of Miyok fronds containing pinholes (about 100 per piece) occupied by developing nauplii were cultured individually in coverd petri dishes at $15\,^{\circ}$ C in an incubator. The infested fronds used in the experiment were collected on May 24, 1990 in a Miyok farm in Songjeong on the southeastern coast of Korea. The collected fronds were brought to the laboratory and their culture was begun about two hours later on the same day.

At the beginning of the culture, the developing nauplii in pinholes of the fronds were all at the early nauplius stage. In four days, about 80% of them developed into late nauplii or beyond; in seven days, about 90% developed into early copepodids or beyond; in 11 days, about 90% developed into late copepodids or adults, and in 15 days, 31% reached the adult stage.

The adults obtained in the culture agreed in morphological details with the descriptions of *Amenophis orientalis* given by Ho and Hong (1988), and all developmental stages found in the cultures belonged to the same species.

The Miyok frond, from which infested pieces were cut out for the culture experiment, was found to harbor *Amenophia orientalis*, *Scutellidium* sp. and other harpacticoid species in a ratio of 47:42:11,

^{**} on the southwestern coast of Korea

Date L	Length of culture	No. of pinhole in the culture	No. of copepodids recovered	% composition of development stages found**				
	in days			I	II	III	IV	V
5/24***	0			100				
5/28	4	346	167	21	71	8		
5/31	7	398	144		9	75	16	
6/4	11	278	79		1	8	86	
6/8	15	330	56			5	64	3

Table 2. Development of Amenophia orientalis reared from early nauplius stages in laboratory*

and in the first two species all stages including ovigerous adults were common. However, all late copepodids and adults, stages that permit a positive species identification, developed from the nauplii in pinholes cultured were of only one species, *Amenophia orientalis*.

According to the ratios of the number of copepods obtained in the culture and that of pinholes in the fronds, about a half of the cultured harpacticoids survived after four days, about 28% survived after 11 days, and only 17% survived after 15 days. These low survival rates resulted from the death of all copepods in a number of cultures. In some cultures, however, up to 60% of the nauplii survived to develop into late copepodids or adults.

As the nauplii molted into copepodids, they crawled out of the hole and started moving actively on the surface of the frond. They began eating algal tissue on the surface usually without digging through the fronds. The fronds therefore showed increasing numbers of scratch marks on the surface as the copepodids actively consumed the algal tissue.

In all of the cultures, we have observed many protozoa crawling on the frond. Their number seemed to increas with time. We were not able to determine, however, whether the copepods actually preyed on these protozoa.

We have not observed newly hatched nauplii started digging into the algal tissue, but we have actually observed nauplii in the pinholes that were actively nibbling the algal tissue. The nauplii had massive, jaw-like antennae similar to those found in other frond-mining nauplii described by Harding (1954) and Fahrenbach (1962). Initially the hole was round and big enough to accommodate just one nauplius, but it became elongate as the nauplius dug in one directions. By the time when the nauplius was ready to molt into a copepodid, the hole was as large as about seven times the residing copepod, and the surrounding algal tissue was dark. The edge of the hole, however, showed no obvious growth forming a gall as in other frond-mining harpacticoids.

As shown by Fahrenbach (1962) and Ho and Hong (1988), several harpacticoid species parasitic to red algae have been descrived, including all nauplius and copepodid stages in some species. In two species, *Diarthrodes feldmanni* and *Diarthrodes cystoecus*, both larvae and adults are known to feed on the algal tissue (Bocquet, 1953; Fahrenhach, 1962).

All of these parasitic harpacticoids are known to mine and produce galls or pinholes on the frond. Similiar galls or pinholes occupied by nauplii were first found in the phaeophyte Miyok *Undaria pinnatifida* in Japan by Torii and Yamamoto (1975) and in Korea by Kang (1981). Adult harpacticoid copepods to which these mining nauplii were believed to belong have been described for the first time in Korea by Ho and Hong (1988). Of the two species, *Amenophia orientalis* and *Parathalestris in-*

^{*}Pieces of Miyok frond containing about 100 pinholes occupied by developing nauplii were cultured in covered petri dishes at 15 °C in an incubator.

^{**} Developmental stages: I = early nauplii, II = late nauplii, III = early copepodids, V = late copepodids, V = adults.

^{***} The starting date of the culture.

festus, described by Ho and Hong (1988), however only Amenophia orientalis was found in the study to be conspecific with the nauplii mining the frond of Miyok in Korea. In our knowledge, our study is the first to identify the minign nauplii by rearing them in the laboratory up to the adult stage.

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要 約

우리나라 養殖미역에 바늘구멍을 뚫는 橈脚類 幼生을 同定하기 위하여 南海岸의 養殖미역에 棲 息하는 橈脚類를 調査하고 미역에 바늘구멍을 뚫 는 橈脚類 幼生을 實驗室에서 直接飼育했다. 南海 岸의 養殖미역 葉體에 棲息하고 있는 橈脚類는 여 러 種이 있는데, 이들 中 Ho와 Hong이 記述한 Amenophia orientalis와 다른 未同定種인 Scutellidium sp.가 이번 研究中 미역葉體 洗滌液속에 繼 續的으로 많은 量이 出現하였다. 이 두 種에 속하는 암컷中에는 抱卵個體가 많았다. 飼育實驗 結果를 보면, 15℃에서 구멍을 뚫는 幼生은 15일만에 成體 또는 成體에 가까운 未成體까지 成長했다. 이들 成 體 혹은 未成體는 모두 Amenophia orientalis로 同 定되었다. 그러므로 우리나라 養殖미역에 寄生하면 서 미역葉體에 구멍을 뚫는 種은 Amenophia orientalis로 밝혀졌다.

Received November 31, 1990 Accepted December 8, 1990