

Fine Structure of the Silk Spigots in the Spider *Dolomedes sulfureus* (Araneae: Pisauridae)

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닷거미과의 황닷거미 (*Dolomedes sulfureus*)의 실크 분비장치의 미세구조

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ABSTRACT

Dolomedes spiders of the family Pisauridae are one of free wandering spiders with semi-aquatic habitation. They do not build web for prey-hunting but build a nursery web for spiderlings. This paper describes the fine structure of the silk-spinning spigots of the fishing spider *Dolomedes sulfureus* revealed by the field emission scanning electron microscope (FESEM). The fishing spider *Dolomedes sulfureus* possesses only three types of silk glands which connected through the typical spinning tubes on the spinnerets. The silk spigots of this spider were identified as three groups: ampullates, pyriforms and aciniforms. Two pairs of major ampullate glands send secretory ductules to the anterior spinnerets, and another two pairs (or 1~2 pairs in males) of minor ampullate glands supply the middle spinnerets. In addition, the pyriform glands feed silk into the anterior spinnerets (62~68 pairs in females and 45~50 pairs in males), and the aciniforms send ductules to the middle (33~40 pairs in females and 18~25 pairs in males) and the posterior spinnerets (42~50 pairs in females and 24~28 pairs in males). Among these, the ampullate one is the most predominate gland in both sexes.

Keywords : Fine structure, Silk, Spigot, Spider, *Dolomedes sulfureus*

INTRODUCTION

Dolomedes spiders of the family Pisauridae are also known as fishing spiders or dock spiders. Almost all *Dolomedes* species are semi-aquatic, with the exception of the tree or grass dwelling spiders *D. albineus* in USA and *D. minor* in New Zealand (Gertsch, 1979; Foelix, 1996). They run over the surface of ponds or streams. They are known

to capture small fish and frogs (Gorb & Barth, 1994).

Dolomedes spiders do not use silk for capturing prey. These spiders usually wait for their prey on grass, on the edges of ponds or streams. Like all pisaurids, female *Dolomedes* carry their egg sacs in their chelicerae, and use silk to build a nursery web in which the young hatch. These nursery webs are built shortly before the spiderlings emerge from the eggsac (Suter et al., 1997).

Although, the wandering spiders do not produce webs for

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prey-catching (Coddington & Levi, 1991; Foelix, 1996), they also possess silk apparatus even though the functions are still not fully defined (Kovoor, 1987; Peters & Kovoor, 1991). Moreover, It now seems that all spiders who actively hunt their prey, or who use little or no silk in prey-catching, are descendant of web building spiders (Shear, 1994). Thus, the silk glands and their functional spinning apparatuses are regarded as important characteristics of the modern spiders.

Previous reports have also demonstrated that the functional specialization of silk producing apparatus involves precise modifications of the shape of the spinnerets, the number and morphology of spigots, and anatomical characteristics of the silk glands (Peters, 1987; Shear, 1994; Moon & Tillinghast, 2004). Accordingly, the variations in the composition of the spinning apparatus of araneid spider have studied by many workers (Coddington, 1986; Kovoor, 1987; Nentwig & Heimer, 1987; Peters & Kovoor, 1991; Park & Moon, 2002; Moon & An, 2006).

Although, the *Dolomedes* spiders are one of the most popular spiders in temperate areas, little is known about their silk-spinning systems (Suter & Gruenwald, 2000). Thus, the fine structural organization of the silk spigots in the fishing spider *Dolomedes sulfureus* especially its functional microstructure of the silk-spinning apparatus were analysed using field emission scanning electron microscopy (FESEM).

MATERIALS AND METHODS

Adult individuals of the fishing spiders *Dolomedes sulfureus* (Araneae: Pisauridae) were collected in a local area near Cheonan campus of Dankook University, Cheonan, Korea. All spiders were maintained under ambient conditions with natural lighting in wooden frames with glass plates on the front and back, and fed insects and water daily. Six females and five males were anesthetized with CO₂ and dissected under a dissecting light microscope in a drop of spider Ringer's solution consisting of 160 mM NaCl, 7.5 mM KCl, 4 mM CaCl₂, 1 mM MgCl₂, 4 mM NaHCO₃, 20 mM glucose, pH 7.4 (Groome et al., 1991).

For scanning electron microscopic examination, the whole abdomen which containing a total set of spinnerets were gently removed and fixed in a mixture of 2% paraformaldehyde and 2.5% glutaraldehyde buffered with 0.1 M phosphate buffer at pH 7.4. Postfixation was performed with 1% osmium tetroxide in the same buffer and washed

several times in 0.1 M phosphate buffer following fixation.

The specimens were then dehydrated in ascending concentrations of ethanol from 30 to 100% (1 hour at each concentration, with one repeat at 100% ethanol). The specimens were then transferred to hexamethyldisilazane (HMDS) and allowed to air dry. All samples were coated to a thickness of approximately 20 nm with gold-palladium alloy using a sputter coater and examined on a Hitachi S-4300 (Hitachi Co., Japan) field emission scanning electron microscope (FESEM) operated with accelerating voltage of 5~20 kV.

RESULTS

The silk spigots of the spider, *Dolomedes sulfureus* are located at the surface of the silk spinning apparatus which composed of inner silk glands and outer spinnerets. The spinnerets are composed of three pairs: the anterior, middle, and posterior spinnerets (Fig. 1A, B). The silk glands are associated with a particular type of spigots on each spinnerets, and the silk spigots of both sexes of spiders are connected with a particular type of spinning tubes on each silk glands, namely, ampullate, pyriform and aciniform glands (Table 1).

By scanning electron microscopic visualization, basically two types of spinning tubes could be distinguished: there were a few large sized spigots and numerous small hair-like spools. Both of spigots and spools are composed of two functional segments: the basal and terminal segments. The ampullate glands are connected to the spigot-type spinning tubes, and the pyriform and aciniform glands are connected to the spool-type tubes (Fig. 2A).

The anterior spinnerets comprise 2 pairs of ampullate spigots in both sexes and approximately 62~68 (female) or 45~50 pairs (male) of pyriform spools. Another two pairs of ampullate spigots and 33~40 (female) or 18~25 pairs (male) of aciniform spools in both sexes are connected on the median spinnerets, respectively. In addition, the posterior spinnerets comprise approximately 42~50 pairs (female) or 24~28 pairs (male) of aciniform spools, respectively (Table 1).

The ampullate spigots are located on both the anterior and middle spinnerets. Two pairs of major ampullate glands send secretory ductules to the anterior spinnerets (Figs. 2A, 3A), and another two pairs of minor ampullate glands supplies the middle spinnerets (Figs. 2D, 3D). When viewed from above the spinnerets, two large spinning tubes of

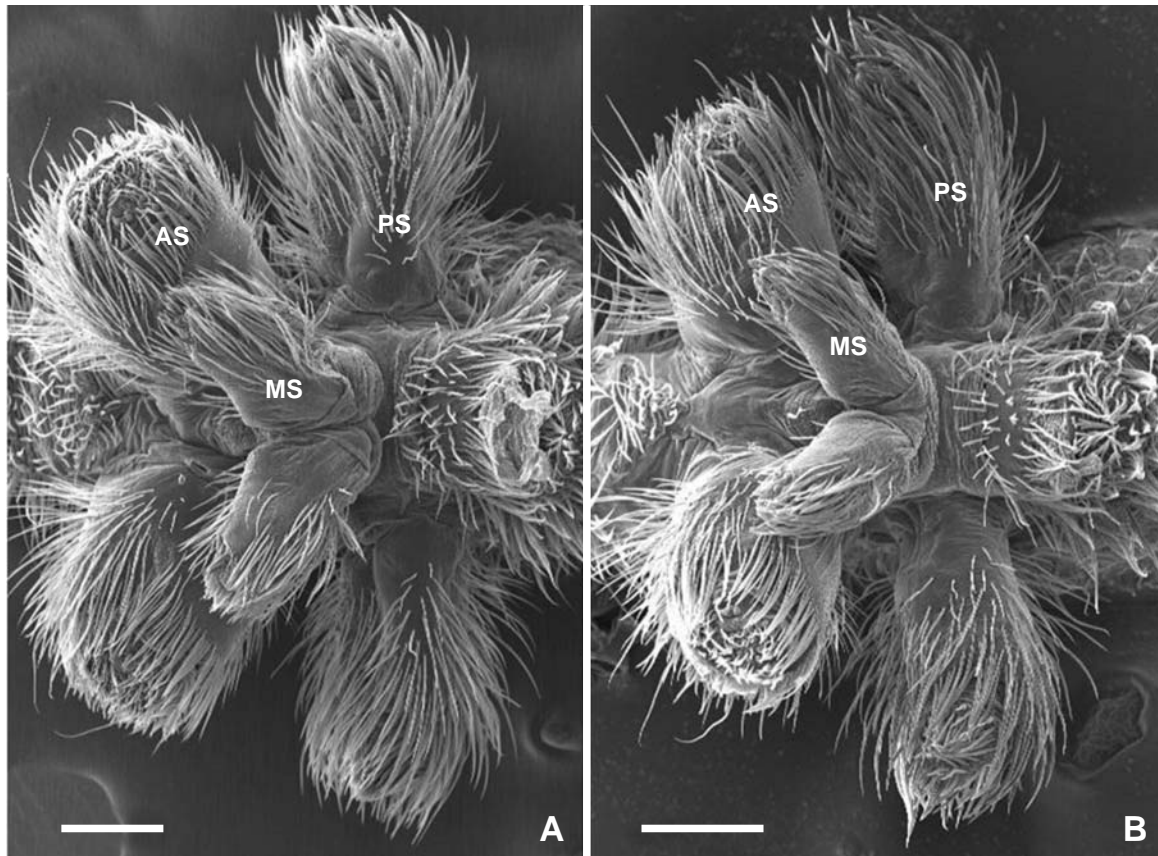


Fig. 1. Scanning electron micrographs of the silk spinning apparatus, which is composed of three pairs of spinnerets in the spider, *Dolomedes sulfureus*. The anterior pairs of spinnerets of both sexes are the most prominent. AS: anterior spinneret, MS: middle spinneret, PS: posterior spinneret. (A): Female spider; (B): Male spider. All scale bars indicate 250 μm .

Table 1. Silk-spinning apparatuses of the spider *Dolomedes sulfureus*

Spinneret	Gland type	No. pair (s)	
		Female	Male
Anterior	Ampullate gland	2	1~2
	Pyriiform gland	62~68	45~50
Middle	Ampullate gland	2	1~2
	Aciniform gland	33~40	18~25
Posterior	Aciniform gland	42~50	24~28

ampullate spigots are the most prominent (Figs. 2B, C, 3B, C).

In addition to the large spinning spigots of the major ampullate glands, numerous small spinning spools of the pyriform glands are also present at the anterior spinneret. These spools of the pyriform glands are quite different from those of the spigots of the ampullate glands with respect to size and morphology. The spinning tubes of the pyriform glands are only found on this anterior spinneret approximately 62~68 (female) or 45~50 (male) pairs. All of these

spools are basically composed of two main segments—more flexible basal segment and slender apical segment (Figs. 2A, 3A).

There are two kinds of spigots on the median pairs of spinnerets—two pairs of ampullates and several pairs of aciniforms. Additional two pairs of minor ampullate spigots are connected on the median spinneret in both sexes. The sizes of these minor ampullate spigots are somewhat smaller than those of the major ampullate spigots on the anterior spinneret. The ampullate spigots are composed of two main segments—more flexible basal segments and elongated terminal segment commonly. Each silk fiber is spun through small opening of the terminal segment (Figs. 2C, 3C).

The spinning tubes of the aciniform glands are found on this median (33~40 pairs in females and 18~25 pairs in males) and the posterior spinneret (approximately 42~50 pairs in females and 24~28 pairs in males). The aciniform spigots of the median spinnerets are located in a more anterior position than are the ampullate spigots (Figs. 2D, 3D). While the spinning tubes of the ampullate glands are

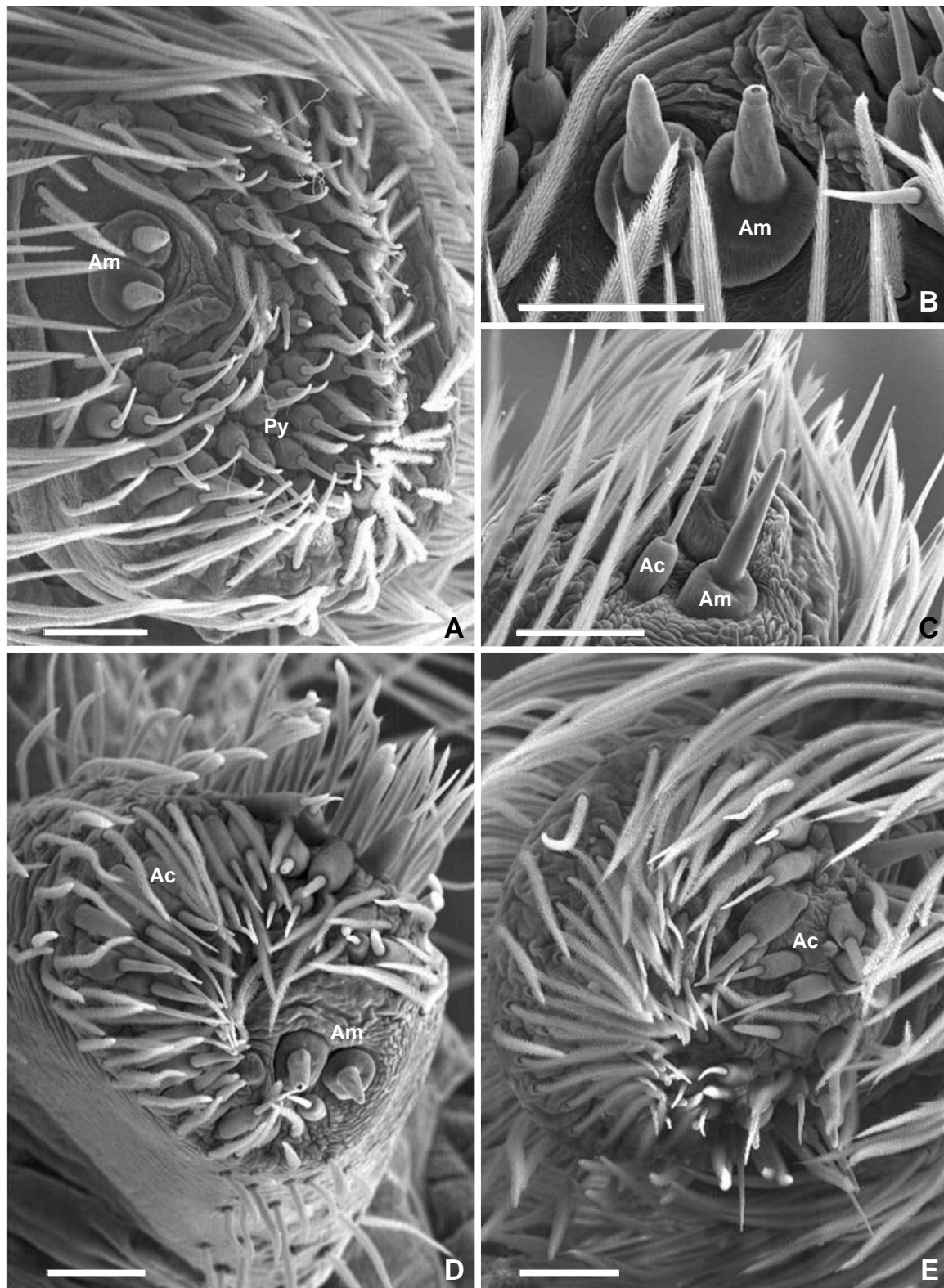


Fig. 2. Scanning electron micrographs of the female spinnerets in *Dolomedes sulfureus*. (A, B) The anterior spinneret has two types of spinning tubes, including two large spigots of the major ampullate gland (Am) and small spinning tubes of pyriform glands (Py). All of these spigots are composed of two main segments—more flexible basal segment and slender apical segment. (C, D) The middle spinneret also has two types of spinning tubes—two pairs of the major ampullate glands and numerous aciniform glands (Ac). (E) Posterior spinneret has only one type of aciniform spigots. All scale bars indicate 50 μ m.

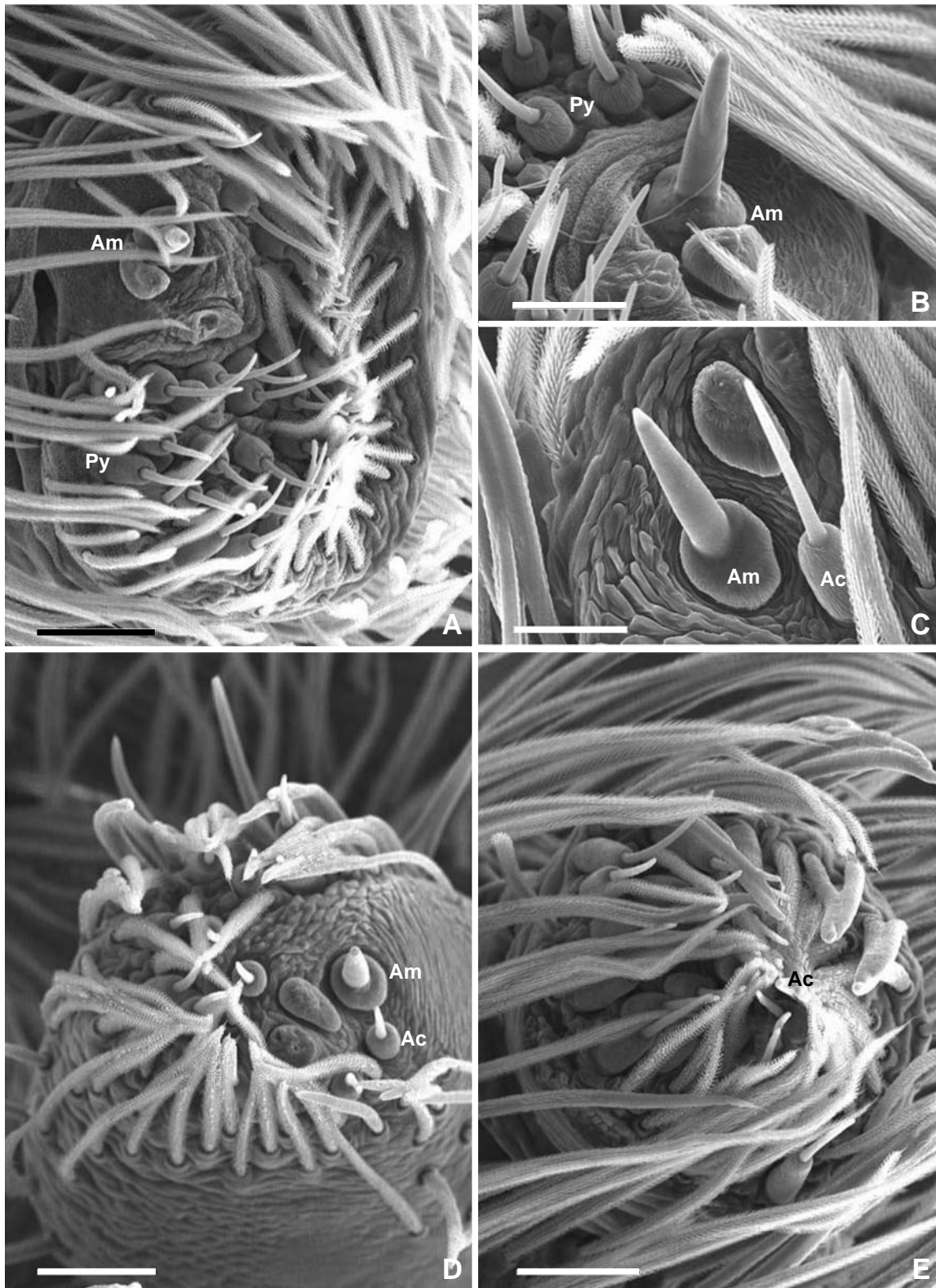


Fig. 3. Scanning electron micrographs of the spinnerets of male spider *Dolomedes sulfureus*. (A, B) Anterior spinneret has two pairs of the major ampullate spigots (Am) and 45~50 pairs of the pyriform spigots (Py). The apical segments of the ampullate spigots are bigger than those of the aciniform spigots. (C, D) Additional 2 pairs of ampullate spigots and 18~25 pairs of aciniform spigots (Ac) are distributed on the middle spinneret. The aciniform spigots are different from those of pyriforms that observed on the anterior spinneret. (E) Posterior spinneret has only one type of aciniform spigots. Scale bars indicate 50 μm (A, D, E) and 25 μm (B, C), respectively.

connected to the bullet-type spigots, the aciniform glands are connected to the cannon-type spools (Figs. 2C, 3C). The aciniform spools also comprise basal segments and elongated terminal segment that have fine terminal tips (Figs. 2D, 3D).

The posterior spinnerets have only one type of spinning tubes on their spinning surfaces. Thus, approximately 42 ~ 50 (females) or 24 ~ 28 (males) pairs of aciniform spools are counted only on the posterior spinnerets, respectively. They are distributed on the whole spinning surfaces of the posterior spinnerets, and are spread concentrically toward its long axis of spinneret (Figs. 2E, 3E)

DISCUSSION

It has been known that the functional specialization of the silk-spinning apparatus involves precise modifications of the spinnerets, the number and morphology of spigots, and anatomical characteristics of the silk glands (Peters & Kovoov, 1991; Foelix, 1996). Although, the spinning apparatus often undergoes adaptative variations, some basic characteristics usually remain unchanged at the familial level (Peters, 1987; Shear, 1994). Therefore, spiders can be classified by their shape and number of components of their silk-spinning apparatus.

Even though, spiders produce various kinds of silks which are used for the remarkably diverse silk constructs (Denny, 1976; Coddington, 1986), the main function of the spider silk is prey-catching (Nentwig & Heimer, 1987). However, the *Dolomedes* spiders make no web to catch prey. They use their silk as a dragline and a nursery web for protecting their eggs (Foelix, 1996). Because of their behavioral characteristics, the *Dolomedes* spiders are generally classified as one of typical hunting spiders somewhat similar to those of wolf spiders or lynx spiders (Coddington & Levi, 1991; Moon, 2006). Nevertheless, they also possess various silk spigots even though the exact functions are still not fully defined.

It has been revealed that the silk-spinning apparatuses of this *Dolomedes* spider correspond to the patterns found in those of wandering lynx spiders (Moon, 2006) or crab spiders (Moon & An, 2005, 2006). However, the *Dolomedes* spiders do not correspond to those of wandering wolf spiders (Moon, 1998a) or other wandering Salticid spiders (Kovoov, 1987). Three basic types of silk spigots-the ampullates, the pyriforms and the aciniforms-were identified in *Dolomedes sulfureus*. The two pairs of ampullates and

the pyriforms were observed on the anterior spinnerets; another two pairs of the ampullates and the aciniforms were observed on the middle spinnerets, and the rest of aciniforms were identified on the posterior spinnerets.

On the basis of fine structural analysis using scanning electron microscope, It has been revealed that even the female spiders possess none of the tubuliform glands for cocoon production and the so-called "triad" spigots for capture thread production. Recently, this type of simple spinning apparatus has been reported in two related species of the crab spider *Misumenops tricuspidatus* (Moon & An, 2005), *Oxytate striatipes* (Moon & An, 2006) and a lynx spider *Oxyopes licenti* (Moon, 2006).

Previous work has shown that most web-building spiders produce a gluey material from a complex structure of a flagelliform and two aggregate glands (Kavanagh & Tillinghast, 1979; Moon & Kim, 1989; Moon, 1998b). This functional unit known as the triad spigots (Peters, 1987) are connected to posterior spinneret and are used for spinning the sticky spirals of capture thread in a web (Peters, 1987; Peters & Kovoov, 1991; Moon & Kim, 2005). However, both sexes of *Dolomedes sulfureus* spiders lack the triad spigots similarly to those of other wandering spiders such as the wolf spider, *Pardosa astrigera* (Moon, 1998a), the funnel-web spider, *Agelena limbata* (Park & Moon, 2002), the crab spiders *Misumenops tricuspidatus* (Moon & An, 2005), the green crab spider *Oxytate striatipes* (Moon & An, 2006) and the lynx spider *Oxyopes licenti* (Moon, 2006).

Like the other web-building spiders (Peters & Kovoov, 1991; Park & Moon, 2002; Moon & Tillinghast, 2004; Moon & An, 2006), the ampullate glands were the largest one in this *Dolomedes* spider. Most of araneid spiders use silk from the ampullate glands for making draglines, frame threads and web building (Coddington, 1986). Previous reports have shown that there are two pairs of ampullate glands for the genus *Araneus* (Tillinghast & Townley, 1986; Townley et al., 1991; Moon & Tillinghast, 2004), genus *Nephila* (Tillinghast & Christenson, 1984) and the other araneid spiders (Peters & Kovoov, 1991). However, there are four pairs in *D. sulfureus* similarly to those of wandering salticid (Kovoov, 1987), lycosid (Moon, 1998a), agelelid (Park & Moon, 2002), and thomisid (Moon & An, 2005, 2006) spiders.

As the functional specialization of silk producing apparatus involves precise modifications of the shape of the spinnerets, the number and morphology of spigots, and anatomical characteristics of the silk glands (Peters, 1987; Shear, 1994; Moon & Tillinghast, 2004), the variations in

the composition of the spinning apparatus of araneid spiders are regarded as important characteristics of the modern spiders. Accordingly, the silk-spinning apparatus of this spider can be considered as the simplest system among the araneid spiders with the exception of some exclusively primitive species (Kovoor, 1987; Peters & Kovoor, 1991).

REFERENCES

- Coddington J: The monophyletic origin of the orb web. In: Shear WA, ed, Spiders: Webs, Behavior, and Evolution, pp. 319-363, Stanford University Press, Stanford, CA, USA, 1986.
- Coddington JA, Levi HW: Systematics and evolution of spiders (Araneae). *Ann Rev Ecol Syst* 22 : 565-592, 1991.
- Denny M: The physical properties of spider's silk and their role in the design of orb-webs. *J Exp Biol* 65 : 483-506, 1976.
- Foelix RF: *Biology of Spiders* (2nd ed). Oxford University Press, London, 1996.
- Gertsch WJ: *American Spiders* (2nd ed). Van Nostrand Reinhold Co., New York, 1979.
- Gorb SN, Barth FG: Locomotor behavior during prey-capture of a fishing spider, *Dolomedes plantarius* (Araneae: Araneidae): Galloping and stopping. *J Arachnol* 22 : 89-93, 1994.
- Groome JR, Townley MA, de Tschaschell M, Tillinghast EK: Detection and isolation of proctolin-like immunoreactivity in Arachnids: Possible cardioregulatory role for proctolin in the orb-weaving spiders *Argiope* and *Araneus*. *J Insect Physiol* 37 : 9-19, 1991.
- Kavanagh EJ, Tillinghast EK: Fibrous and adhesive components of the orb webs of *Araneus trifolium* and *Argiope trifasciata*. *J Morphol* 160 : 17-32, 1979.
- Kovoor J: Comparative structure and histochemistry of silk-producing organs in Arachnids. In: Nentwig W, ed, *Ecophysiology of Spiders*, pp. 159-186, Springer-Verlag, Berlin, 1987.
- Moon MJ: Fine structural analysis of the silk producing apparatus in wolf spider, *Pardosa astrigera* (Araneae: Lycosidae). *Korean J Entomol* 28 : 201-211, 1998a.
- Moon MJ: Fine structure of the silk producing apparatus in the garden spider, *Argiope bruennichi*. *Korean J Entomol* 28 : 345-354, 1998b.
- Moon MJ: Microstructure of the silk spinning nozzles in the lynx spider, *Oxyopes licenti* (Araneae: Oxyopidae). *Integrative Biosciences* 10 : 85-91, 2006.
- Moon MJ, An JS: Spinneret microstructure of silk spinning apparatus in the crab spider, *Misumenops tricuspidatus* (Araneae: Thomisidae). *Entomol Res* 35 : 67-74, 2005.
- Moon MJ, An JS: Microstructure of silk spigot of the green crab spider, *Oxytate striatipes* (Araneae: Thomisidae). *Entomol Res* 36 : 133-138, 2006.
- Moon MJ, Kim TH: Microstructural analysis of the capture thread spinning apparatus in orb web spiders. *Entomol Res* 35 : 133-140, 2005.
- Moon MJ, Kim WK: Fine structural study on the capture threads-producing organs in *Nephila clavata* L. Koch (Araneae: Araneidae). (I) Aggregate glands. *Kor J Zool* 32 : 211-220, 1989.
- Moon MJ, Tillinghast EK: Silk production after mechanical pulling stimulation in the ampullate silk glands of the barn spider, *Araneus cavaticus*. *Entomol Res* 34 : 123-130, 2004.
- Nentwig W, Heimer S: Ecological aspects of spider webs. In: Nentwig W, ed, *Ecophysiology of Spiders*, pp. 211-225, Springer-Verlag, Berlin, 1987.
- Park JG, Moon MJ: Fine structure of the spinning apparatus in the funnel-web spider, *Agelena limbata*. *Korean J Entomol* 32 : 223-232, 2002.
- Peters HM: Fine structure and function of capture threads. In: Nentwig W, ed, *Ecophysiology of Spiders*, pp. 187-202, Springer-Verlag, Berlin, 1987.
- Peters HM, Kovoor J: The silk-producing system of *Linyphia triangularis* (Araneae: Linyphiidae) and some comparisons with Araneidae: Structure, histochemistry and function. *Zoomorphology* 111 : 1-17, 1991.
- Shear WA: Untangling the evolution of the web. *Amer Sci* 82 : 256-266, 1994.
- Suter RB, Gruenwald J: Predator avoidance on the water surface? Kinematics and efficacy of vertical jumping by *Dolomedes* (Araneae, Pisauridae). *J Arachnol* 28 : 201-210, 2000.
- Suter RB, Rosenberg O, Loeb S, Wildman H, Long JH Jr: Locomotion on the water surface: Propulsive mechanisms of the fisher spider *Dolomedes triton*. *J Exp Biol* 200 : 2523-2538, 1997.
- Tillinghast EK, Christenson T: Observations on the chemical composition of the web of *Nephila clavipes* (Araneae: Araneidae). *J Arachnol* 12 : 69-74, 1984.
- Tillinghast EK, Townley MA: The independent regulation of protein synthesis in the major ampullate glands of *Araneus cavaticus* Keyserling. *J Insect Physiol* 32 : 117-123, 1986.
- Townley MA, Horner NV, Cherim NA, Tugmon CR, Tillinghast EK: Selected aspects of spinning apparatus development in *Araneus cavaticus* (Araneae: Araneidae). *J Morphol* 208 : 175-191, 1991.

< 국문 초록 >

배회성거미류의 일종으로 주로 물가에 서식하는 닳거미과 (Pisauridae)의 황닷거미 (*Dolomedes sulfureus*)를 실험재료로 실험 크 분비장치의 미세구조를 주사전자현미경 (FESEM)으로 관찰하고, 토사관의 미세구조적 특성을 분석하였다. 황닷거미는 먹이를 포획하기 위한 거미줄을 만들지 않지만, 유충을 보호하기 위한

그물을 만드는 종류로 병상선 (ampullate glands), 이상선 (pyriform glands), 포도상선 (aciniform glands) 등, 세 종류의 견사선과 이와 연결된 실젯 표면의 독특한 토사관을 가지고 있었다. 두 쌍의 대 병상선은 앞실젯을 통해, 그리고 또 다른 두 쌍 (수컷의 경우 1~2쌍)의 소병상선은 가운데실젯을 통해 분비관이 연결되어 있었

다. 이상선은 앞실젯 표면의 토사관을 통해 (♀: 62~68쌍, ♂: 45~50쌍) 분비되었고, 포도상선은 가운데실젯 (♀: 33~40쌍, ♂: 18~25쌍)과 뒷실젯 (♀: 42~50쌍, ♂: 24~28쌍)의 토사관을 통해 분비됨이 확인되었다. 이들 중, 병상선과 연결된 토사관이 암수 모두에서 가장 현저한 분비 특성을 가진 것으로 관찰되었다.