A SEM Study of Genus Badhamia (Myxomycetes)

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Abstract: Capillitial threads, c-granules and other parts in a sporangium of 17 species of Genus Badhamia were investigated in detail using SEM apparatus and a few new findings were obtained. Capillitium is composed by branching and anastomosing threads and plates at an interconnection point. Capillitium of B. affinis and some species are club shaped threads. The one of B. crassipella and some species are thick threads and lump shaped plates. Other species have slender threads, flat plates and other types. Usually capillitium is evenly positioned in a sporangium. Capillitial threads of B. dearnesii and some species are mainly positioned in an upper portion and gradually decreased to a lower portion. Species belong to genus Badhamia have c-granules on a peridial surface and an inside of a capillitial tubular thread. C-granules is globose, smooth on the surface, $0.3-1.9 \mu m$ in diameter. Usually the surface of a capillitial thread has numerous hemispherical processes traced by c-granules inside. Peridium has many small pits on the surface. A pit connects into a capillitial tubular thread extended to the inside of a sporangium. C-granules on a peridum and the one in a capillitial tubular thread are connected with each other through a pit on a peridial surface.

1. Introduction

The Genus *Badhamia* belongs to the family Physaraceae. Species of this genus develop world wide and it has been reported about 40 species up to the present. These species have much calcareous granules (c-granules, it is used in this paper) on the outer surface of the peridium and within the capillitial threads. These c-granules usually are put on the surface as small lumps and it is broken as a dried scab when spores are dispersed from a sporangium. A scattering of c-granules on a peridial surface of c-granules and its dispersing from the peridium are a big differentiation in each species. It has branching and anastomosing capillitial threads in a sporangium. The shape of capillitial thread is differed from

each other species, such as narrow thread, thick club, flat plate and so on. Capillitial thread, peridium, c-granule of 17 species belong to the Genus *Badhamia* collected from USA, Canada and Japan were studied in detail using SEM and some new findings were obtained.

2. Materials and methods

17 species of *Badhamia affinis*, *B. capsulifera*, *B. crassipella*, *B. dearnesii*, *B. foliicola*, *B. gracilis*, *B. iowensis*, *B. lilacina*, *B. nitens*. *B. obovata*, *B. ovispora*, *B. populina*, *B. rhytidosperma*, *B. rugulosa*, *B. spinispora*, *B. utricularis* and *B. versicolor* were provided in this observation. The location of collection is in the USA, Canada and Japan. Materials have been naturally dried and stocked in room temperature. For a SEM (Scanning Electron Microscope) observation, the material were dehydrated in a graded ethyl alcohol series and replaced into xylene. Finally the materials were mounted on specimen stubs with a double faced adhesive tape and sealed with a silver paste. The materials treated in the way mentioned above were coated in about a 40nm layer of gold or gold paradium in a vacuum chamber of an ion coater apparatus, Eiko engineering Co. LTD, IB-3 type, for seven minutes. The ion-coated materials were observed with an SEM, Hitachi Co. LTD, S-4000 type, at 15 Kv accelerating voltage.

Specimens examined:

B. affinis HWK2774 Tarrant Co., Texas, USA, HTN4173 Shimoina-gun, Nagano pref., Japan, B. capsulifera TEB2482 Cumberland Co., Kentucky, USA, B. crassipella DTK5252 Bullte Co., California, USA, B. dearnesii DTK3920 Butte Co., California, USA, B. foliicola DTK5892 Butte Co., California, USA, B. gracilis GWM3247 Johnson Co., Iowa, USA, B. iowensis HWK223 Johnson Co., Iowa, USA, B. lilacina TEB2485 Wastenaw Co., Michigan, USA, B. nitens DTK2634 Butte Co., California, USA, B. obovata HWK2205 Sumter Co., Florida, USA, B. ovispora HWK2172 Boulder Co., Colorado, USA, B. populina JS98 Kapasiwin, Alverta, Canada, B. rhytidosperma HWK2758 Geary Co., Kansas, USA, B. rugulosa Eliasson3719 Washington Co., Arkansas, USA, B. spinispora HWK2768 Cumberland Co., Kentucky, USA, B. utricularis DTK4653 Butte Co., California, USA, B. versicolor TEB3420 Girardeau Co., Missouri, USA. *TEB: T. E. Brooks, HTN: T. Hatano, HWK: H. W. Keller, DTK: D. T. Kowalski, GWM: G. W. Martin, SJ: S. J. Smith

3. A observation results

Following books of Emoto (1977), Keller and Braun (1999), Lister (1925), Martin and Alexopoulos (1969), Thind (1977) and Yamamoto (1998) were used to check species names offered in this study. The morphological features of a capillitia, c-granules, peridia were studied in detail using SEM.

Badhamia affinis Rostaf.

Capillitial thread: Composed of club shaped tubular threads and flat plates at an interconnecting point,

branching and anastomosing but not so conspicuous, contained numerous c-granules inside, scattered with hemispherical processes on the surface traced by c-granules inside. **Position of capillitial thread**: Much threads develop on the upper portion of a sporangium, gradually decreased at a lower portion, usually not developed near the bottom portion on a stalk. **c-granule**: globose, smooth on a surface, 0.4-0.9 μ m in diameter. **Outer surface of peridium**: Closely covered with numerous c-granules, smooth on the surface except parts of c-granules, a lumb of c-granules of 30 μ m in diameter is broken away as a dried scab, when spores disperse from a sporangium (Fig.7). **Inner surface of peridium**: Scattered with numerous fine granules and hemispherical processes traced by c-granules on a outer peridial surface, roughened by fine granules and hemispherical processes, smooth and wavy at parts except fine granules and hemispherical processes.

Badhamia capsulifera (Bull.) Berk.

Capillitial thread: Composed of club shaped tubular threads and flat plates at an interconnection point (Fig. 1), branching and anastomosing, threads are less than other species. Contained numerous c-granules inside, roughened by fine granules and hemispherical processes on the surface traced by c-granules inside, partly wavy. **c-granule**: globose, smooth on the surface, $0.3-0.9\mu m$ in diameter. **Outer surface of peridium**: Covered with numerous c-granules (Fig. 9), roughened, smooth, partly wavy except parts of c-granules. **Inner surface of peridium**: Roughened by fine granules and hemispherical processes on a surface traced by c-granules on an outer peridial surface.

Badhamia crassipella Whiteney & Keller

Capillitial thread: Composed of thick club shaped tubular threads and many angled lump shaped plates at an interconnection point, contained with numerous c-granules inside, covered with hemispherical processes on a surface traced by c-granules inside, roughened (Fig.2), it similar to a capillitial thread of *B. obovata*. **c-granule**: globose, smooth on the surface, 0.6-1.3µm in diameter (Fig. 12). **Outer surface of peridium**: Covered with numerous c-granules at a upper part of a peridium, decreased to the lower portion, it has many fine pits filled with many c-granules on a surface, smooth except parts of fine pits and c-granules (Fig. 8). **Inner surface of peridium**: Roughened, scattered with fine granules and hemispherical processes on a surface traced by c-granules on an outer peridial surface (Fig. 15), hemispherical processes are less at the lower portion of a peridium.

Badhamia dearnesii Hagelst.

Capillitial thread: Composed of slender tubes and acute angled flat plates at an interconnection point, the total amount of threads is less than other species, contained numerous c-granules inside, scattered with numerous hemispherical processes on a surface traced by c-granules inside, roughened. **Position of capillitial thread**: Much threads develop on the upper part of a sporangium, gradually decreased to a

lower portion (Fig. 13). **c-granule**: globose, smooth on the surface, $0.9-1.8\mu m$ in diameter. **Outer surface of peridium**: Covered with numerous fine granules and many c-granules, roughened, c-granules are gradually decreased to the bottom. **Inner surface of peridium**: Roughened by hemispherical processes on a surface traced by c-granules at the upper portion of a outer peridial surface. Gradually become less to the lower portion, almost smooth on a surface.

Badhamia foliicola Lister

Capillitial thread: Composed of slender tubular threads and thin narrow flat plates at an interconnection point, branching and anastomosing, contained a few hemispherical processes on a surface, almost smooth, slightly roughened (Fig. 4). Position of capillitial thread: Evenly developed in a whole part of a sporangium. c-granule: globose, smooth on the surface, $0.6-1.8~\mu$ m in diameter. Outer surface of peridium: Covered with many c-granules on the upper part of a peridium, roughened, almost smooth except parts of c-granules, c-granules are decreased to the lower portion. Inner surface of peridium: Roughened, scattered with numerous fine granules and many hemispherical processes on a surface traced by c-granules on a outer peridial surface, gradually become finely roughened and wavy to a lower portion (Fig. 16).

Badhamia gracilis (T. Macbr.) T. Macbr.

Capillitial thread: Composed of thick club shaped tubular threads and thick lump shaped plates at an interconnection point, irregularly branching and anastomosing, contained with many c-granules inside, scattered with many hemispherical processes on a surface traced by c-granules on a outer peridial surface (Fig. 11). c-granule: globose, smooth on the surface, 0.4-1.5 μ m in diameter. Outer surface of peridium: Covered with numerous fine granules and c-granules on a surface, smooth or wavy on a surface except parts of c-granules. Inner surface of peridium: Roughened, scattered with numerous fine granules and hemispherical processes on the surface traced by c-granules on an outer peridial surface.

Badhamia iowensis T. Machr.

Capillitial thread: Composed of very slender tubular or very thick angled tubular threads and lump shaped plates at an interconnection point, strongly varied in size and shape, filled with numerous c-granules inside, scattered with numerous fine granules and hemispherical processes on a surface traced by c-granules inside (Fig. 10). Position of capillitial thread: Filled with much capillitial threads on a surface of the upper part of a sporangium, threads gradually decreased downward and almost no capillitial threads at a bottom part (Fig. 14). **c-granule**: globose, smooth on the surface, $0.6-1.8\mu m$ in diameter. Outer surface of peridium: Covered with numerous c-granules, many c-granules compose a small lump of $30\mu m$ in diameter, smooth on the surface except parts of c-granules. A lump of c-granules is broken away when spore are dispersed, c-granules become less at the lower portion of a sporangium.

Inner surface of peridium: Scattered with huge fine granules and numerous hemispherical processes on the surface traced by c-granules on an outer peridial surface, hemispherical processes become less at a lower part of an inner peridial surface and increased vertical striations on a surface.

Badhamia lilacina (Fr.) Rostaf.

Capillitial thread: Composed of slender tubular threads and angular flat plates at an interconnection point of threads, usually contained a few c-granules inside, scattered with fine granules and a few hemispherical processes on a surface traced by c-granules inside, smooth except parts of these processes, sometimes remained longitudinal striations. Position of capillitial thread: The number of capillitial threads are decreased from an upper part to a lower part of a sporangium. c-granule: globose, smooth on the surface, $0.9-1.8\mu$ m in diameter. Outer surface of peridium: Covered with numerous c-granules, smooth except parts of c-granules. Inner surface of peridium: Roughened, scattered with fine granules and numerous hemispherical processes on the surface traced by c-granules on the outer peridial surface.

Badhamia nitens Berk.

Capillitial thread: Composed of thick club shaped tubular threads and angler lump shaped plates, sometimes very big angled plates, at an interconnection point, contained with c-granules inside, but not so conspicuous, roughened by fine granules and hemispherical processes on a surface traced by c-granules inside, sometimes with wavy striation. **c-granule**: globose, smooth on the surface, $0.6-1.3\mu$ m in diameter. **Outer surface of peridium**: Covered with many c-granules on a surface, smooth on a surface except parts of c-granules, sometimes slightly striated. **Inner surface of peridium**: Roughened by fine granules and hemispherical processes on a surface traced by c-granules on an outer peridial surface.

Badhamia obovata (Peck) S. J. Smith

Capillitial thread: Composed of thick club shaped tubular threads, usually with no angled flat knots at an interconnection point, branching and anastomosing, especially characterized with strange processes as a coral formation on a surface (Fig. 6), maybe occured by c-granules inside. Position of capillitial thread: Evenly developed in a whole part of a sporangium. **c-granule**: globose, smooth on a surface, sometimes with numerous fine spinules on a surface, $0.8-2.1\mu m$ in diameter. Outer surface of peridium: Covered with many c-granules, smooth on a surface except parts of c-granules. Inner surface of peridium: Roughened, scattered with fine granules and hemispherical processes on a surface traced by c-granules on an outer peridial surface.

Badhamia ovispora Racib.

Capillitial thread: Composed of club shaped tubular threads and flat plates at an interconnection

point, branching and anastomosing, contained numerous c-granules inside, roughened by hemispherical processes on a surface traced by c-granules inside, but not so conspicuous, smooth on a surface except parts of hemispherical processes. Keller *et al* (1975) reported about a portion of capillitium of this species using SEM in a paper of the taxonomic status of B. ovispora. The shape and surface ornamentation of a capillitial thread studied in this paper agreed with the result obtained in their study. **c-granule**: globose, smooth on the surface, 1.2- 2.0μ m in diameter. **Outer surface of peridium**: Covered with many c-granules on a surface, smooth on a surface except parts of c-granules. **Inner surface of peridium**: Roughened, scattered with many hemispherical processes on a surface traced by c-granules on an outer peridial surface.

Badhamia populina A. & G. Lister

Capillitial thread: Composed of very short slender tubular threads and typical angled lump shaped tubular plates, connected with each other plates (Fig. 3), contained numerous c-granules inside, strongly roughened by numerous fine granules and hemispherical processes on a surface traced by c-granules inside. Position of capillitial thread: Evenly developed in a whole part of a sporangium. c-granule: globose, smooth on the surface, 0.7- 1.4μ m in diameter. Outer surface of peridium: Peridum is composed of two layers (Fig. 18). The outer surface of an outer layer has numerous c-granules on a surface, smooth on the surface except parts of c-granules. Inner surface of peridium: Inner surface of an inner layer is roughened by numerous fine granules and hemispherical processes on a surface traced by c-granules on the outer surface of an outer layer.

Badhamia rhytidosperma Keller & Schoknecht

Capillitial thread: Composed of slender, club shaped tubular threads and thick plates at an interconnection point, contained fully numerous c-granules inside, roughened by numerous fine granules and hemispherical processes on a surface traced by c-granules inside of a thread. **c-granule**: globose, smooth on a surface, sometimes slightly roughened, sometimes with a small pit on a surface, 0.4- 1.3μ m in diameter. Outer surface of peridium: Covered with extremely huge numbers of c-granules, sometimes a peridium is separated to two layers (Fig. 17). Inner surface of peridium: Strongly roughened by numerous fine granules and hemispherical processes on a surface traced by c-granules on an outer peridial surface, sometimes with strongly vein-like reticulations.

Badhamia rugulosa Brooks & Keller

Capillitial thread: Composed of club shaped tubular threads and flat plates at an interconnection point, branching and anastomosing, contained fully numerous c-granules inside, scattered with numerous fine granules and hemispherical processes on a surface traced by c-granules inside. c-granule: globose, smooth on the surface, 0.6- 1.2μ m in diameter. Outer surface of peridium: Covered

with many c-granules, smooth or wavy on a surface except parts of c-granules. **Inner surface of peridium**: Roughened by numerous fine granules and hemispherical processes on a surface traced by c-granules on an outer peridial surface.

Badhamia spinispora (Eliass. & Lundq.) Keller & Schokn.

Capillitial thread: Composed of club shaped tubular threads, branching and anastomosing, contained fully numerous c- granules inside, scattered with fine granules and hemispherical processes on a surface traced c- granules inside of a tube. **c-granule**: globose, smooth on the surface, 0.5- 1.3μ m in diameter. Outer surface of peridium: Covered with numerous c- granules, smooth on a surface except parts of c-granules, characteristically remained branching and anastomosing vein-like ridges on a surface. **Inner surface of peridium**: Roughened by numerous fine granules and hemispherical processes traced by c-granules on an outer peridial surface, sometimes characteristically with winding ditch-like striations.

Badhamia utricularis (Bull.) Berk.

Capillitial thread: Composed of extreme slender tubular threads and narrow flat plates at an interconnection point, flat plate is not so big, often branching and anastomosing, roughened by granules on a surface of threads and plates, the influence of c- granules is not so strong (Fig. 5). Position of capillitial thread: Evenly developed inside of a sporangium. c-granule: globose, smooth on the surface, $0.5-1.8\mu m$ in diameter. Outer surface of peridium: Covered with c-granules and roughened, smooth on a surface except parts of c- granules. Amount of c-granules is not so much than other species. Inner surface of peridium: Slightly roughened by fine granules and hemispherical processes on a surface traced by c- granules on an outer peridial surface, with wavy striations on a surface.

Badhamia versicolor Lister

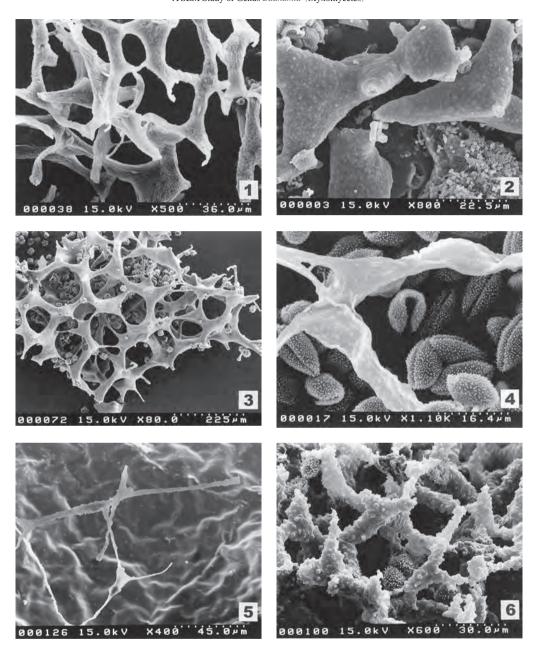
Capillitial thread: Composed of thick club shaped and thick lump shaped plates at an interconnection point, contained fully with c- granules, strongly roughened by numerous hemispherical processes on a surface traced by c- granules on an outer peridial surface. **c-granule**: globose, smooth on the surface, 0.4- 1.2μ m in diameter. **Outer surface of peridium**: Covered with many c- granules, smooth on a surface except parts of c- granules. **Inner surface of peridium**: Roughened by numerous fine granules and hemispherical processes on the surface traced by c- granules on an outer peridial surface, sometimes with wavy winding striations.

4. Discussion

Capillitia are composed by threads and plates at an interconnection point. They are tubular, often branched and anastomosed. Thickness and length of a thread, the shape of a plate are different in each species. Numbers of branching and anastomosing are also different in each species. Generally a

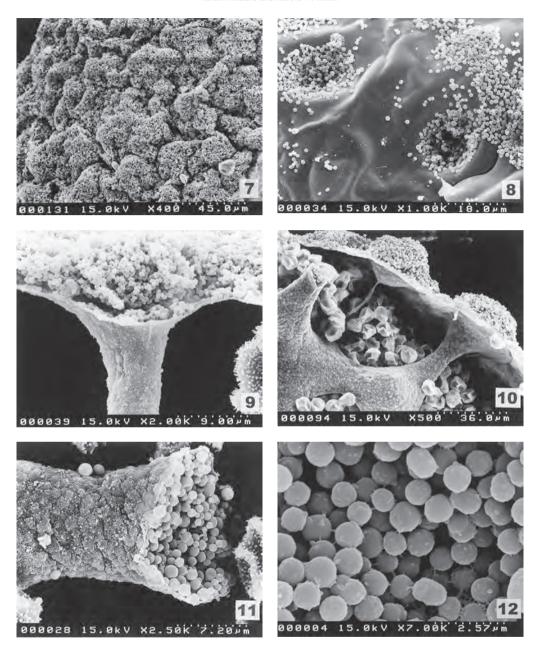
capillitium is composed of club shaped threads and plates, they are slightly thickened angular plates. Species of B. affinis, B. capsulifera, B. ovispora and B. rugulosa have club shaped threads mentioned above (Fig. 1). This shape is a normal type in a genus Badhamia. Species of B. crassipella, B. gracilis, B. nitens and B. versicolor have thick threads and lump shaped plates (Fig. 2). Species of B. iowensis and B. populina have slender threads and lump shaped plates (Fig. 3). Species of B. foliicola and B. utricularis have long slender thread, usually thick plates are not developed in these species (Figs. 4, 5). B. lilacina and B. dearnesii has slender threads and flat plates. B. obovata has special shaped threads. It looks like coral formation (Fig. 6). Usually it has not flat knots. The surface of capillitial thread of species belongs to genus Badhamia generally has fine granules and hemispherical processes on a surface traced by c-granules in a tube thread and strongly roughened. Especially, species of B. crassipella, B. gracilis, B. iowensis, B. nitens, B. populina, B. spinispora, and B. versicolor have thick capillitial threads that are fully filled with c-granules in a tube thread (Fig. 11). The surface of its thread has typical hemispherical processes and strongly roughened. Species of B. foliicola and B. utricularis have slender threads and narrow plates (Fig. 4). These are not contained so much c- granules inside and the surface of thread is smooth or hemispherical processes are less than a thick capillitial thread. The condition of development of capillitial thread in a sporangium is different in each species. The threads of B. foliicola, B. populina and B. utricularis are evenly develop in a sporangium. Threads of B. affinis, B. dearnesii and B. iowensis have many capillitial threads at the upper part of a sporangium and gradually become less to the lower portion (Fig. 13). Especially, B. iowensis has no capillitial threads at a bottom near a stalk (Fig. 14).

Species belongs to Badhamia has numerous c-granules. A c-granul is globose in whole species examined. Usually smooth on a surface (Fig. 12), but c-granules of B. obovata has numerous fine spinules on the surface. A small number of granules of B. rhytidosperma have very fine pits on the surface of a granule. The size is usually 0.3- 1.9μ m in diameter. The outer surface of a peridium has numerous c-granules. The surface except parts dispersed c-granules is almost smooth (Fig. 8). Sometimes the surface of a peridium has a great number of c-granules and make a thick layer or double layer (Fig. 17). The inner surface of a peridium is usually roughened by hemispherical processes traced by c-granules on the outer peridial surface. It also has numerous fine granules on a surface (Fig. 15). Usually the inner surface of a peridium is same as an outer surface of a capillitial thread. Usually the peridial membrane is one layer but sometimes, such as B. populina, has double layers (Fig. 18). The surface of a peridium has numerous small pits (Fig. 8). These pits connected with the inside of a capillitial thread (Fig. 9). Species belongs to Badhamia has numerous c-granules on a peridial surface and the inside of a capillitial thread. These c-granules have same origin and continuously connected with each other between a peridial surface and a capillitial thread (Fig. 10).



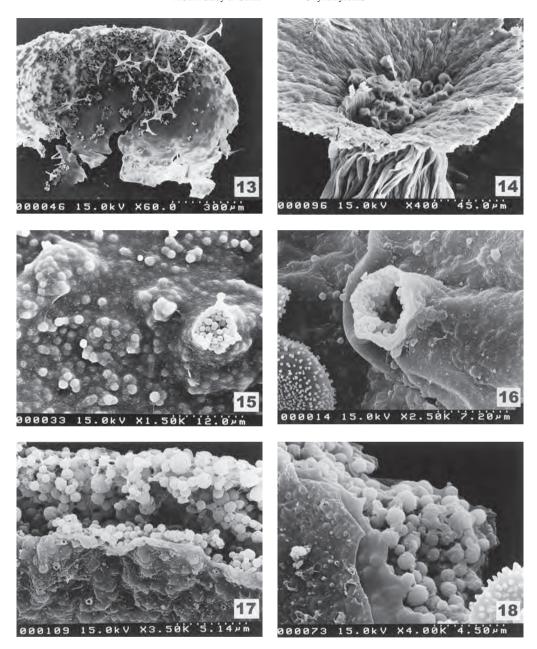
Figs. 1-6. Capillitium of selected Badhamia

1. Capillitial threads of *B. capsulifera*. It is composed of club shaped threads and flat plates. 2. Capillitial threads of *B. crassipella*. It is composed of thick club shaped threads and angled lump shaped plates. The surface is roughened by hemispherical processes. 3. Capillitial threads of *B. populina*. It is composed of very short slender threads and angled lump shaped plates. 4. Capillitial threads of *B. folicola*. It indicates flat plates at an interconnection point of a capillitium. Surface is slightly roughened. 5. Capillitial threads of *B. utricularis*. It is composed of extreme slender threads and narrow flat plates and slightly roughened on a surface. 6. Capillitial threads of *B. obovata*. It is characterized with strange processes as a coral formation on a surface.



Figs. 7-12. C-granules and capillitium of selected Badhamia

7. Scabs of c-granules on a peridial surface of B. affinis. A scab is about $30\mu m$ in diameter, broken fragmentarily away, when spores disperse. 8. An outer surface of peridium of B. crassipella after roughly taking off c-granules on a surface. Peridial surface except c-granules is smooth or slightly wavy, it also indicates two small pits on a surface connected into capillitial threads and many c-granules are recognized in a pit. 9. The crossing broken face of a sporangial surface at a fine pit and a capillitial thread of B. capsulifera. Upper is outside of a sporangium, below is inside of a sporangium. It indicates a pit connects into a capillitial thread. It also indicates numerous c-granules are on a pit continuously connected with in a capillitial thread. 10. The crossing broken face of an upper part of sporangium of B. iowensis. It indicates a connection between small pit on a peridial surface and a capillitial thread. C-granules on a peridial surface comes into a capillitial thread from a pit. 11. The crossing broken face of a capillitial thread of B. gracilis. It fully filled with c-granules inside. The surface of a thread is strongly roughened. 12. Typical c-granules on a peridial surface of B. crassipella.



Figs. 13-18. 13-14: Position of capillitium thread of selected *Badhamia*13. Capillitial threads of *B. dearnessii* are unevenly positioned, much threads are on an upper part of a sporangium, decreased to a lower portion. 14. It is indiated no capillitial threads at a bottom part of a sporangium of *B. iowensis*, 15-18: Peridium and c-granules of selected *Badhamia* 15. An inner surface of a peridium of *B. crassipella*. Strongly roughened by hemispherical processes traced by c-granules on an outer surface of a peridium. Right is a hole after a capillitial thread was broken away, it appears many c-granules on an outer surface. 16. An inner surface of a peridium at a lower portion of *B. foliicola*, finely roughened and wavy. Center is a hole of capillitial thread. This hole leads to a small pit on a peridial surface. 17. *B. rhytidosperma* has much c-granules on a peridial surface and sometimes a layer of c-granules is separated to two layers. 18. Peridium of *B. populina* is composed of two layers. It is indicated an inner and an outer layer of a peridium and c-granules on the outer surface of a peridium.

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