Possible causes of rarity of sexual reproduction in *Pellia endiviifolia* (Dicks.) Dumort.

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Abstract: Reproduction in bryophytes is by asexual as well as sexual means. Asexual reproduction is a significant adaptation to special environments and occurs by a large variety of methods. During the last few decades, many of the bryophytes, particularly the dioecious ones are reported to have registered a decline in their fertile populations. There is, therefore, a need to understand the reasons for their disappearance.

1. Introduction:
During the last few decades, “Reproductive biology” has emerged as one of the most important areas of research in Plant Sciences (Baker, 1959; Queenborough *et al.*, 2007). While sufficient literature is available on reproductive biology of flowering plants (Jhonston and Schoen, 1996; Husband and Schemske, 1996; Kaul *et al.*, 2002; Godwille *et al.*, 2005; Kudo, 2006), very little attention has so far been paid to unravel the reproductive strategies adopted by their non-flowering counterparts including bryophytes.

Bryophytes, an important group of non-vascular land plants inhabit a wide range of habitats i.e. epilithic (rock / stone surface), non-epilithic (soil surface), epiphytic (on other plants) and aquatic (inside water). This enables these plants to establish themselves over wide distributional range and subsequently ensure their survival even in event of loss/destruction of some of their habitats.

Bryophytes are believed to have originated from algae and migrated from aquatic to terrestrial habitat somewhere during Mid Ordovician to early Silurian periods; 476 – 432 million years ago (Groth-Malonck *et al.*, 2004). In order to ensure successful existence on land, they acquired a number of morpho-anatomical and reproductive characters. The group shows a marked advance over their algal progenitors in the mode of sexual reproduction. Without exception, it is oogamous in the whole group. Furthermore, jacketed sex organs, embryo and sporophyte made their appearance in the group for the first time during the evolution of plant kingdom.

Bryophyte taxa may be monoecious or dioecious. Approximately 68% liverwort species are reported to be dioecious. Of these, 44% belong to order Marchantiales and 64-72% to Jungermanniales (Crum, 2001).
It has been suggested that a higher proportion of dioecious liverwort species fails to produce sporophyte (Lindberg et al., 2000). Possible cause of this rarity suggested is, the spatial distribution of plants of two sexes. Studies on the reproductive biology of dioecious hepatics, therefore, assume special importance. Pellia Raddi., a genus of the order Metzgeriales of class Hepaticopsida has robust thallus with broad, elongated lobes. It is worldwide in distribution. Out of the six species reported world over (P.neesiana, P.epiphylla, P.columbiana, P.megaspora, P.borealis and P.endiviifolia), three (P.neesiana, P.epiphylla and P.endiviifolia) have been reported from India (Kashyap, 1929; Srivastava, 1998 and Bapna and Kachroo, 2000). Two of these species (P.endiviifolia and P.epiphylla) have been recorded from J&K state (Kashyap, 1929; Kachroo et al., 1977; Srivastava, 1979; Langer et al., 2003; Tanwir, 2005 and Tanwir et al., 2008). While P.endiviifolia is dioecious, P.epiphylla is monoecious. Sexual reproduction in liverworts has recently been reported to be related to microhabitat (McLetchie et al., 2002). In view of the paucity of literature available in this direction and to understand this phenomenon further, a dioecious liverwort species, P.endiviifolia was selected for the present investigation.

2.Material and Methods:

Present communication is based on the studies on reproductive details of six populations of P.endiviifolia collected from Nagbani (Jammu district), Jib and T-Morh (Udhampur district) in Jammu region of J&K state. Nagbani, a small village lies 14 km North West of Jammu city at an altitude of 350 m. A small canal (Fig.1) flows through Nagbani area whose banks are inhabited by a number of hepatic and moss taxa growing on brick wall (KA NB1; Fig.2), cemented wall (KA NB2; Fig.3) and moist soil (KA NB3; Fig.4). Some of the patches of KA NB3 collected from moist soil were partially submerged in canal water (Fig.5).The other three populations were collected from Udhampur; two from Jib inhabiting brick wall (KA JB1; Fig.6) and rock surface (KA JB2; Fig.7) and one from T-Morh inhabiting a cave (KA TM; Fig.8 ). Collections were made fortnightly between January 2009 and February 2010. Populations were photographed under the natural habitats and a small portion of patch was removed from the substratum and preserved in 70% ethyl alcohol. Data on reproductive phenology (initiation, maturation and persistence of male and female gametangia and sporophytes) were recorded in the field, whereas anatomical features of gametangia and sporophyte were studied in laboratory under microscope. For determining spore-elater output, a capsule was crushed on a glass slide with the help of a needle and mounted in a drop of glycerine. For counting them, a coverslip was divided into quadrates with the help of a marker. Number of spores and elaters were counted individually for each quadrate. Spore/elater output per capsule was calculated by adding the number of spores and elaters. Spore-elater ratio was calculated by using the formula:

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\text{Spore/ elater ratio} = \frac{\text{number of spore per capsule}}{\text{number of elaters per capsule}}
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3.Results:

Thalli of all the six populations of P.endiviifolia investigated reproduced vegetatively. Vegetative reproduction occurred exclusively through adventitious branching (Figs.9 and 10). Female thalli were collected from three populations, all growing at Nagbani; female gametangia (archegonia) embedded within dorsal, green, tubular involucres (Fig.11) which appeared during first week of February and persisted till May. V.S. passing through involucres revealed the presence of 1-2 (Fig.12),
5-6 (Fig.13) and 7-10 (Fig.14) archegonia in populations KA NB1, KA NB2 and KA NB3 respectively. Male plants were recorded in only one population (KA NB3). Antheridia appeared during April and were seen as light green circular spots scattered irregularly on the thallus (Fig.15) and were embedded in the thallus in antheridial chambers (Fig.16). The population which produced male plants (KA NB3) grew at a distance of about 20 m from KA NB1 and 5 m from KA NB2. No reproductive plants were collected from any of the three populations of Udhampur.

Sporophytes were collected in May only from the thalli of KA NB3 and that too from the patch submerged in water. Sporophyte consisted of foot, seta and capsule (Fig.17). Seta was transparent and attained a length of 1.5 cm (Figs.18 and 19); capsule globular with a bistratose wall, produced numerous spores and elaters (Fig.20), a tuft of elaters attached to the capsule base forming an elaterophore (Figs.20 and 21); spores yellowish brown with smooth surface, oval or spherical, large, 75.5-98.9 μm in diameter (Figs.22 and 23); elaters light brown, slender, spindle shaped, bispiral to tetraspiral, unbranched, 44-202 μm (Fig.24); spore and elater output per capsule was 800-1650 and 300-600 respectively and the spore elater ratio came to be 2.45:1.
4. Discussion:
Reproduction is one of the most essential features of life, common to all living organisms. They reproduce
either by asexual or sexual means. Asexual reproduction is a significant adaptation to specific environments
and biological conditions where the cost of sexual reproduction to a species is disadvantageous. It is very
prevalent among bryophytes. In this group as a whole, asexual reproduction occurs by a large variety of
modes such as persistant apices, fragmentation, adventitious branches, tuber formation, gemmae, stolons,
cladia etc. The exclusive occurrence of asexual reproduction in the gametophytic phase in the life cycle of
bryophytes is unique among terrestrial plants and it has been estimated that 40% of leafy hepatics

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Figs.9 and 10: Thalli showing adventitious branching; Fig.11: Patch of female thalli bearing involucres; Figs. 12-14: V.S. of thalli passing through involucres surrounding 1-2, 5-6 and 7-10 archegonia; Fig.15: Patch of thalli bearing antheridia; Fig.16: v.S. of thalli passing through an antheridium.
reproduce asexually (Wyatt, 1982 and 1994). Vegetative regeneration is known to be an effective means of maintenance of local populations (Longton and Schuster, 1983; Mishler, 1988) among bryophytes. *Pellia* reproduces vegetatively by death and decay of older parts and adventitious branching. All the six populations of *P. endiviifolia* presently investigated, produced adventitious branches. Occurrence of such branches has earlier been reported by several workers (M’Ardle, 1895; Cavers, 1903; Schuster, 1984) in this species.

Sexual reproduction is an important event in the life cycle as it is a means of bringing about genetic variation. The male and female organs are remarkably uniform in fundamental structure throughout.

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bryophytes. The antheridium is a delicate sac enclosing antherozoid mother cells and ultimately, mature male gametes and the archegonium is flask shaped, differentiated into rounded venter and long neck. In order Metzgeriales to which *Pellia* belongs, the sex organs are never borne on receptacles typical of Marchantiales.

Population sex ratios in dioecious bryophytes are known to be influenced by microhabitat specialization. Maintenance of sexually dimorphic traits and biased sex ratios can lead to a lack of sexual reproduction (McLetchie et al., 2002). In *Lophozia silvicola*, for instance, Lindberg (2000) recorded highest frequency of fertile colonies on wood. Data obtained presently also seem to support a positive role of habitat in sexuality as out of the six populations investigated, five were epilithic (KA NB1, KA NB2, KA JB1, KA JB2 and KA TM) and one non-epilithic (KA NB3). Quite surprisingly, sporophyte formation was recorded only in the sole non-epilithic population. Furthermore, it would be worthwhile to mention here that the sporophytes were recorded only in thalli of *P.endiviifolia* submerged in water. This clearly indicates that in the taxon under investigation, both substratum (non-epilithic) as well as availability of water are major factors responsible for sexual reproduction and sporophyte formation.

Another cause of failure of sporophyte production among dioecious bryophytes is, the limited range of fertilization. Among the bryophytes, it is well known that many dioecious taxa never produce capsules (e.g. *Sphagnum*; Cronberg, 1991), presumably due to absence of the opposite sex or to inability of the sperm to reach the female plant and its reproductive structures successfully. Burr (1939) also observed that in *Cyathophorum bulbosum*, sporophytes were developed only in the patches that grew on moist soil, where male plants were located nearly a meter above the females, but on a nearby bank the entirely female population was completely barren. Rydgen et al. (2006) found that 85% of the female shoots with sporophytes were situated within a distance of 5cm from the nearest male plants and the maximum distance travelled by antherozoids was, 11.6 cm. For dioecious species, fertilization is believed to be inversely proportional to the distance between male and female plants. However, fertilization ranges reported for majority of taxa were generally much shorter (Vanderpoorten and Goffinet, 2009). Present observations on three populations studied at Nagbani also seem to support this view. Sporophytes were recorded only in one population in which thalli of two sexes grew intermingled with each other. Remaining two populations with female thalli which grew 5 cm and 20 cm apart from male thalli did not form any sporophytes. It can be inferred from their observations that in *Pellia endiviifolia*, range of fertilization is less than 5 cm.

Effect of microhabitat on sporophyte production has also been observed in case of *Marchantia nepalensis* (Arora, 2011) and *M. palmata* (Arora and Langer, 2011). Out of a total of 14 populations studied, only three, one of *M.nepalensis* and two of *M.palmata* reproduced sexually while others did not produce any receptacles/ sporophytes, or both. In this case, sporophytes were produced only by the epilithic populations indicating that a correlation exists between habitat and sexuality in these species as well, although the trends obtained are entirely different for *Marchantia* and *Pellia*. Observations recorded for another species of *Marchantia* (*M. inflexa*) were, however, totally opposite (Fuselier and Lecthie, 2004). In this species, both male and female plants were found to inhabit epilithic as well as non-epilithic habitats but male plants were collected from an additional habitat (epiphytic). On the basis of the data collected presently for *P.endiviifolia*, it can be concluded that:

(i) there exists a correlation between sexual reproduction and microhabitat and
(ii) failure of sporophyte formation in two populations (KA NB1 and KA NB2) bearing female plants is due to limited range of fertilization.
In view of different trends exhibited by Pellia and Marchantia, it is suggested that many more populations of these two and other liverwort taxa will have to be subjected to such studies in order to obtain a clear picture.

5. References:

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