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Novel attempt: To study anastomosis groping in *Rhizoctonia bataticola* causing dry root rot in chickpea

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Abstract

Anastomosis behavior of different isolates of *R. bataticola* with each other collected from different locations of Chhattisgarh was investigated in all possible combinations. All isolates exhibited self-Compatibility that is fusion of wall and membrane and showed homologous anastomosis (called reaction type 3).

Keywords: Novel attempt, anastomosis, *Rhizoctonia bataticola*, Chickpea

Introduction

Chickpea belongs to Leguminaceae family usually grown after rainy season on conserved soil moisture during winter in the tropics; in spring in the temperate and Mediterranean regions. It is the important grain legume grown for protein rich seeds for human consumption and to maintain the soil fertility by its nitrogen fixing capability.

Anastomosis is one of the phenomenon to know variability among the fungi. Similar fungi leads to fusion between branches of the same or different hyphae. Hence the bifurcating fungal hyphae can form true reticulating networks. By sharing materials in the form of dissolved ions, hormones, and nucleotides, the fungus maintains bidirectional communication with itself. Two different haploid mating types - if compatible - merge. Somatically, they form a morphologically similar mycelial wave front that continues to grow and explore.

The genus concept of *Rhizoctonia* spp. was established by de Candolle (1815) [4]. However, the lack of specific characters led to the classification of a mixture of unrelated fungi as *Rhizoctonia* spp. (Parmeter and Whitney, 1970 [8], Moore, 1987). Ogoshi (1975) [5]. Enhanced the specificity of the genus concept for *Rhizoctonia* by elevating the characteristics of *R. solani* to the genus level.

Anastomosis reactions between hyphae of paired isolates of *R. solani* consist of several types; such as perfect fusion, imperfect fusion, contact fusion and no reaction (Matsumoto *et al.* 1932) [7]. At present, four categories of anastomosis (C3 to C0) defined by Carling *et al.* (2002) [2]. have been accepted by many researchers.

These are useful for a better understanding of the genetic diversity of *R. solani* populations, because of the background genetically supported by vegetative or somatic compatibility (VC or SC) of confronted isolates (Mac Nish *et al.* 1997) [6].

Anastomosis behaviour of different isolates with each other from different crops were investigated in different combinations and it was found that the type 2 reaction was observed in pairings between rice and maize with moongbean and groundnut isolates. Pairing of moongbean, soybean and groundnut isolates also resulted in similar type of reaction. When the soybean, rice and maize isolates were paired, they resulted in hyphal contact and apparent contact of wall but no membrane to membrane contact was observed and the mycelium grew in close contact with each other (reaction type 1). Pairings between moongbean, soybean and groundnut isolates resulted in type 2 reaction. It was found that all the isolates were self-compatible and also showed self-compatibility in the isolates from the same hosts. Anastomosis is one of the phenomenon to know variability among the fungi. Similar fungi leads to fusion between branches of the same or different hyphae. Hence the bifurcating fungal hyphae can form true reticulating networks. By sharing materials in the form of dissolved ions, hormones, and nucleotides, the fungus maintains bidirectional communication with itself. Two different haploid mating types - if compatible - merge.

Somatically, they form a morphologically similar mycelial wave front that continues to grow and explore

Materials and Methods

The present investigations were carried out in the Department of Plant Pathology, IGKV, Raipur Symptomatic dry root rot infected plants were collected from seventeen chickpea growing areas of Chhattisgarh state (Ambikapur, Dhamtari, Bemetara, Bhatapara, Balod, Durg, Dhamdha, Gandai, Kanker, Kawardha, Narayanpur, Patan, Raigarh, Raipur, Rajnandgaon, Saragaon and Sitapur) for isolation and purification of the causal fungus was brought in the laboratory. Among these locations *R. bataticola* was isolated from nine locations (Kawardha, Rajnandgaon, Kanker, Raigarh, Balod, Dhamtari, Durg, Raipur and Saragaon). So, at the end there were 9 isolates.

To study anastomosis, disposable plastic Petri dishes were used. Glass slides (3"×1") were cleaned with soap and were rinsed with distilled water. They were sterilized in hot air oven for 2-3 hours, by placing slides in each well cleaned and air dried plastic plates sterilized in laminar air flow by giving UV rays. Slides were then coated with a thin layer of sterilized 2% water agar. Discs from actively growing cultures of *R. bataticola* were cut out with the help of corn borer from the edge and were placed 1 cm. apart on the coated slides in two replication (Windels and Nabben, 1989) and were then incubated for 36-48 hours at 27±2°C in BOD incubator, until advancing hyphae made contact and slightly overlapped (Mikhail *et al.* 2010) All possible combination were made (9×9=81) in two replication. The area of hyphal contact was then examined under the microscope. The branching habit of the isolate and the direction of hyphal growth were traced back to their origin in order to ensure that anastomosis has occurred between mycelium of two isolates. The grouping of the isolate was done on the basis of reaction types (Singh 1987) [10]. following microscopic observation.

Result

Anastomosis behavior of different isolates of *R. bataticola*

with each other collected from different location of Chhattisgarh was investigated in different combination. All isolates exhibited self-compatibility that is fusion of wall and membrane and showed homologous anastomosis (reaction type 3).

Discussion

In accordance to above finding self- anastomosis reported to occurs commonly in culture of *R. solani* and it is often assumed that cross anastomosis between cultures also occurs commonly. Schultz (1936) [11], paired a large number of cultures and, on the assumption that successful anastomosis indicated a close genetic relation, they attempted to group their cultures according to their success in anastomosing. Whitney and Parmeter (1963) [12]. Investigated hetero karyon formation following anastomosis between single- spore cultures derived from one parent. They found that two distinctive single spores culture, when paired regularly gave to a culture type different from that of the single spore culture but similar to the original parent.

Fusion of wall and membrane and was designated as compatible reaction (reaction type 3). As fungi grow, fusions between hyphae, or mycelium, that makes up the fungal individual. Networked hyphae are presumably important in intrahyphal communication and homeostasis in an individual colony during growth and reproduction. Hyphal fusions are a way to increase protoplasmic flow restricted by septa, which may be important in influencing hyphal pattern

Formation in the mycelium (Buller, 1933 [1]. Davidson [3]. *et al.* 1996; Rayner, 1996) [3]. Different fungal individuals are capable of undergoing hyphal fusion with each other to form a heterokaryon, whereby genetically different nuclei may coexist in a common cytoplasm. Heterokaryon formation has potential benefits of functional diploidy and mitotic genetic exchange (par asexual cycle) Pontecorvo (1956) [9]. or may increase biomass for cooperation in physiological efforts such as resource exploitation or asexual/sexual reproduction (Buller, 1933 [1]. and Gregory, 1984).

Table 1: Following microscopic observation was as follows

Reaction Type	Descriptions
0	No anastomosis or repulsion. Neutral response. Occasional hyphen contact/ parallel growth but only for a short distance.
1	Hyphen contact and apparent contact of wall but no membrane to membrane contact. Both mycelium must grow in close contact with each other.
2	Wall to wall connection, pore formation and incompatible fusions of cytoplasm leading to death of the fusing and adjoining cells.
3	Fusion of wall and membrane. Compatible reaction.

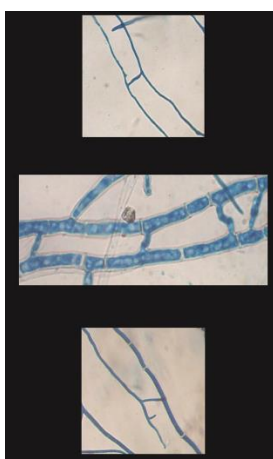


Fig 1: Anastomosis among *R. bataticola* isolates exhibiting self-compatibility (reaction type 3)

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