Introduction
I. INTRODUCTION

Modernized agriculture with more productive systems has often been accompanied by increased production. Concurrently, the area of traditional crops is showing declining trend. Yet, in many parts of the world, these traditional crops play an important role in maintaining stable and sustainable forms of agriculture. One such traditional group of cereal crops is the minor coarse cereals (small millets). Among these, finger millet is the principal small millet.

The Finger millet, *Eleusine coracana* (L.) Gaertn is originated from tropical Asia and Africa. Finger millet is variously called as Ragi, Birds foot, Nagli and Mandua in different regions.

The genus *Eleusine* belonging to the family *Poaceae* was named after a Greek town Eleusine where ‘Ceres’ was worshipped (Burkil, 1935). However, it is said to have derived its name from the Greek Goddess of cereals ‘Eleusine’ (Chalam and Venkateshwaralu, 1965). The word coracana is derived from ‘Kunukkan’-the Singhali name for this grain. *E. coracana* is the only tetraploid (2n = 4x = 36) member of the genus to attain major agricultural impact both in India and Africa.

The crop has a wide range of seasonal adaptation and is successfully grown in varying soil and temperature conditions. It can be grown throughout the year if moisture is adequate and temperatures are above 15°C. Finger millet has adapted to conditions prevailing at sea level to an altitude of 3000 m. The crop is cultivated in different soil types but is mainly grown on red and laterite soils. Alluvial and black soils are also suitable provided the soils are well drained.

Finger millet, native of Ethiopian region of Africa is an important minor millet crop grown for food and feed. The crop is widely cultivated in Asia and Africa, especially in India, Ceylon, Malaya, China, Japan, and Madagascar and in most parts of Central and East Africa under irrigated and rainfed conditions. In the world, India has the
largest area under finger millet cultivation that is around 1.9 million hectares and it is also
the largest producer, producing around 2.5 million tonnes with the productivity of 1308
kg/ha (Anonymous, 2002). Traditionally, the finger millet growing states are Karnataka,
Tamil Nadu, Orissa, Maharastra, Andhra Pradesh, Uttaranchal, Bihar and Gujarat. Finger
millet is commonly referred to as ‘Ragi’ in Karnataka State. The main Finger millet
growing areas in Karnataka State are the southern districts comprising Bangalore, Kolar,
Tumkur, Mysore, Hassan, Mandya and Chitradurga. Karnataka has an area of 0.99 m ha,
production of 1.54 m t and productivity of 1556 kg/ha. Finger millet with its wide
adaptation, easy to cultivate and drought tolerant has made this crop an indispensable
component of dry farming system.

Finger millet is one of the staple food crops of South India especially the marginal
and economically weaker sections and to a lesser extent for urban community also. Ragi
by virtue of its composition is quite comparable to rice or wheat in its nutritive value. In
protein content, ragi (9 per cent) has edge over rice. As regards to essential amino acids,
ragi protein is unique among cereals to possess very high levels of sulphur amino acids.
It is rich in total minerals, especially in calcium (206-690 mg/100g), iron (2.5 –19.9
mg/100g), and phosphorous (227 – 470 mg/100g) content. Ragi grains contain 1 per cent
fats, 72 per cent carbohydrates, and 4 per cent ash, besides vitamins A and B. The green
straw is suitable for making silage.

Finger millet is known to be one of the hardiest crops relatively free from insect
pests and diseases (Coleman, 1922). With the introduction of high yielding varieties, the
crop is prone to many fungal, viral and bacterial pathogens which causes diseases like
blast, blight, foot rot, smut, mosaic, mottling, etc (Govindu and Shivanandappa, 1967). In
spite of over 45 pathogens recorded to infect ragi, blast caused by the fungus Pyricularia
grisea Sacc. is the only serious disease problem wherever the crop is cultivated
(Anilkumar et al., 2003).

Blast, locally called as Benki roga or Iluku roga caused by Pyricularia grisea
(Cooke) Sacc. is a serious disease of finger millet. A temperature of 25-30°C, humidity of
around 90 per cent and cloudy days with intermittent rains are congenial for blast development. In India, McRae (1920) first reported the disease from the Tanjore delta of Tamil Nadu. The disease is being prevalent in India in almost all the states where finger millet is cultivated. Outside India, the disease has been observed in Srilanka (Park, 1932), Somalia (Mohammed, 1980) Tanganuika (Wallace and Wallace, 1948) and Uganda (Hansford, 1943). The blast disease in finger millet is one of the most serious diseases in Kharif sown crop and affects the crop at all stages of growth from seedling stage till grain forming and maturity. In India, the extent of estimated mean annual yield loss due of this disease is 28 per cent (Vishwanath and Seetharam, 1989) and in endemic areas it may cause up to 90 per cent loss in yield (Vishwanath et al., 1997).

Considerable work has been done in many crops to identify, clone and characterize the genes responsible for disease resistance and their products. But, ragi has received little attention so far. Earlier studies to understand defense mechanisms of finger millet against blast disease revealed some role of phenols, sugars (Ravikumar et al., 1995), hydrolytic enzymes like esterases (Annapuma, 2001), Phosphatase (Mohan, 1996), chitinase and β-1,3 glucanase (Amruta, 2005) and secondary metabolites like pytoalexins (Mahesh Kumar et al., 2005) and phytoalexins (Sanmathikumar et al., 2006). Ravikumar and Seetharam, (1994) studied genetic variation in yield components in relation to Pyricularia grisea resistance.

Finger millet, being a low value crop, use of chemical fungicides to manage the disease is economically nonviable. No precise information is available about the extent of genetic diversity and relationship among the genotypes of finger millet. It is necessary to exploit defense capacities of the plant, for which a thorough knowledge of the genetic and biochemical basis of disease resistance mechanisms in the plant is needed. Hence, there is a need for molecular and biochemical characterization of blast disease resistance caused by Pyricularia grisea in finger millet.

Based on the factors discussed above, the present investigation was taken up in order to find out biochemical and molecular association with blast disease in finger millet. In
the present investigation, phenolics, tannins, total sugars and protein content before and after infection of different genotypes were studied with a view to know the role of different biochemical parameter in imparting resistance and also to establish the association between biochemical parameters and host resistance against blast pathogen. Role of enzymes i.e. phenylalanine ammonia lyase, polyphenol oxidase and peroxidase in imparting resistance against blast disease has been well established. Among these defenses, the production of pathogenesis related proteins (PR-proteins) demonstrated their involvement in acquired systemic resistance against a pathogen attack.

Different molecular techniques can distinguish finger millet genotypes of which RAPD and AFLP are important. Both RAPD and AFLP techniques can distinguish the finger millet genotypes from each other. AFLP has been found to be more efficient than RAPD in evaluating the genetic diversity and differentiation among finger millet varieties.

The following studies were undertaken in the present investigation on local varieties of finger millet inoculated with blast fungus *Pyricularia grisea* to understand the biochemical and molecular basis of blast disease resistance mechanism at different growth stages of seedlings with the following objectives

1. To determine the levels of proteins, sugars, phenolics, tannins from leaves of ragi plants of susceptible and resistant finger millet genotypes on different days of germination and at different days after blast fungus inoculation.

2. To determine the levels of activity of some of the enzymes from leaves of ragi plants of susceptible and resistant finger millet genotypes on different days after inoculation with blast fungus (PAL, PPO and Peroxidase) and analysis of qualitative and quantitative phenolics upon inoculation.

3. To determine the acidic and basic PR-proteins from leaves of ragi plants of susceptible and resistant finger millet genotypes on different days after inoculation with blast fungus.
4. To screen finger millet varieties for their genetic diversity by Randomly Amplified Polymorphic DNA (RAPD) technique and Amplified Fragment Length Polymorphism (AFLP) techniques.

5. To determine the metabolic profile of blast resistant and susceptible varieties of finger millet by IRMS – $\Delta^{13}$C ratio at different stages of growth upon infection.