Chapter-1
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The blue planate Earth is only the reservoir of biodiversity in the solar system, due to luxourial growth of gaint plants and animals to invisible ultra microscopic microorganisms. Among three necessities of the human i.e. bread, cloths and house, bread is most important requirement for zoo kingdom which can be fulfiled by plants – the producers. It is very fascinating to study growth promoting invisible microscopic bacteria which influence the growth of plants like rice crop.

Chhattisgarh is commonly known as bowl of rice due to enormous and diversified varieties of the rice. Since my childhood when ever I pass accrose the rice field the greeneries of field becomes much more attractive due to the growth of man heighted rice crop, which are supplied by bottomly placed microorganism including bacteria cynobacteria, rhizobacteria. So since my childhood I was puzzeld and innocently think why and what is the role of bottomly placed micro organisims. But when I little grown up my school and college teachers told me about the role of these microorganisms which are situated near the roots of plants. At the same time being related to a farmers family I have seen several types of fertilizers were used for enhancing the productivity of rice crop. At that time biomagnifications and important dominant and prominent role of rhizobacteria and there interaction with fertilizer like urea were not come in to my consideration but after styding the role of rhizobacteria suddenly it comes in my mind that what is the impact of urea on plant growth promoting rhizobacteria and ultimetaly on rice crop? Thus present topic has been taken into consideration.

Plant based diet of human is an important guideline to low the risk of disease. Therefore it is necessary to maintain the nutritive value of plants. One of the great
challenges that human population is facing in various poor countries of the world is food security. The food production must be increased by irrigation management; best agricultural practices, production of high yielding variety and drought tolerant crop by genetic engineering and proper use of fertilizers. Further India is an agriculture based country. The main occupation of people of major part of the country is agriculture. In agricultural soils the growth of plant is influenced by many factors which include both abiotic and biotic. Soil plays very important role in availability of nutrients to the plants. Soil also contains diversity of micro and macro organisms. Soil microorganisms mediate various processes like nitrogen fixation, denitrification, decomposition, mineralization and mobilization of nutrients. The thin layer of soil which surrounds plant roots is important and active area for root activity and metabolism. This area is known as rhizosphere. The rhizosphere concept was first introduced by Hiltner. The narrow zone of soil which surrounds the roots where microbial populations are stimulated by root activities is known as rhizosphere.(Hartmann et al., 2008). It is an area of biological and chemical activity which is influenced by root exudates and microorganisms.(Kennedy, 2005). The rhizosphere region is highly favorable habitat for the proliferation and metabolism of numerous types of microorganisms.(Alexender 1977). It is well known that the soil microorganism play an important role in the biogeochemical cycle of many nutrient element of soil. Variation in the number and kind of soil microorganisms depends on the physical and chemical characterization of soil, root system of plants as well as the agricultural practices. Various microorganisms are present in soil. Bacteria are most abundant among them. The rhizosphere bacteria have the beneficial or harmful effect on the development of plant.(Dubey and Maheshwari, 2006)
This is now recognized that the rhizosphere microbial population is important to maintain root health, nutrient uptake, and tolerate environmental stress. Many research in laboratory and greenhouse on inoculation of beneficial bacteria shows increase plant growth but the responses may vary in the field.(Bowen and Rovira, 1999). In the rhizosphere there is a biochemical interaction between soil microorganism, plants and soil microfauna.(Pinton et al., 2001)

The proposed work is focused on the rhizobacteria of rice crop. Rice (Oryza sativa) is a monocot plant belongs to family Gramineae. As a cereal grain it is the most important staple food not only in Chhattisgarh but also in the whole county. The slogan “Rice is Life” is appropriate for India as it plays a vital role in food security. It is a means of livelihood for millions of rural people. It is most important grain with regard to human nutrition and calorie intake. It is normally grown as annual plant having long slender leaves and pendulous inflorescence bearing small wind pollinated flowers. It can grow up to the height from 3.3 to 4.9 feet depending upon the variety and the fertility of soil. Rice contains 365 kcal. energy per 100 g. Although the seed of rice is used as the staple food apart from that the its byproducts such as rice husk, rice bran, broken rice, rice milk, rice flour, rice straw, rice starch are also used for various purpose. It is also used for making beverages. In Chhattisgarh it is also considered as a medicinal plant. (Das et al., 2006).

Rice is the major crop grown in Chhattisgarh. The state has often dubbed as the ‘rice bowl’ of India. Rice can improve the health of millions of people who consume it. Thus the nutritive value of rice needs to be maintained by applying proper nutrients to the rice plant during its cultivation. Rice plants require mineral nutrients in large quantity for their growth, development and grain production.(Saharawat, 2000). Cultivation of rice is affected by temperature and pH of soil. The optimum
temperature required for its cultivation is 18 °C to 32°C and pH is 5-8.5. In Chhattisgarh rice is mainly grown in upland and lowland soil type. (Vittal et al., 2004) Durg district of Chhattisgarh state fall under the lowland soil type. In a particular region the rice cultivation method depends on soil type, irrigation facility and rainfall distribution as well as fertilizer application. Before applying the fertilizer to the soil it is required to test the soil to know the availability of the nitrogen, phosphorus and potassium in the soil. Farmers apply chemical fertilizers to improve the crop production, but they are unaware of the dose applied for soil fertility. Thus the over dose of chemical fertilizer neutralize soil and it is harmful for plant growth, nutritive balance and also for the beneficial microorganism present in soil. The traditional method of rice cultivation is flooding the field with excess amount of water in the seedling stage. The fertilizer dose recommended by Agriculture department of state for rice production is 80, 50 & 30 NPK for high yielding variety and 40, 30 & 20 for local Variety. For better growth and development rice plant require adequate mineral nutrients including nitrogen. Further in production of each tone of rice 16-17 kg Nitrogen is removed. (Saharawat, 2000).

Application of fertilizer in transplanted rice field is quite different from upland rice. A series of reactions physical, chemical and biological take place in transplanted rice fields due to presence of excess water in the field. In the root zone anaerobic environment is formed from aerobic condition due to depletion of oxygen in the soil profile, which is responsible for gaseous loss of nitrogen fertilizer due to de-nitrification process. This anaerobic environment also affects the behavior of phosphorus and micro-nutrients especially iron and manganese.

Generally, urea is applied as the nitrogen source for rice production in different stage of plant growth because of its high nitrogen concentration. (Vittal et al.,
Urea is an organic compound with the chemical formula CO(NH$_2$)$_2$. Many soil bacteria possess the enzyme urease which catalyze the conversion of urea molecule to ammonia and carbon dioxide. Urea is highly soluble in water and thus used for fertilizers. The urease enzyme carrying bacteria such as species of *Nitosomonas* which are present in soil were able to assimilate the carbon dioxide and harvest energy by oxidizing ammonia to nitrite. This process is termed as nitrification. Another soil bacterium *Nitrobacter* oxidizes the nitrite to nitrate. This nitrate is extremely mobile in soil causes water pollution from agriculture. Ammonia and nitrates are absorbed by plants which is a source of nitrogen for the growth of plants. (Wikipedia urea). Thus a sustainable amount of urea nitrogen is lost by different mechanism which causes environmental pollution. (Choudhary and Kennedy, 2005)

The urea used for long term reduces organic matter present in soil. Thus there is a need to apply the alternate source of nitrogen which will help minimizing these problems. (Jeyabal, *et al.*, 2001). The rhizosphere microorganisms have either beneficial or harmful effect on the growth and development of plant. (Dubey *et al.*, 2006). The group of rhizobacteria that increase plant growth and yield are termed as plant growth promoting rhizobacteria (PGPR). (Bashan *et al.*, 2004). International concern for food and environment quality is increasing and thus the use of PGPR to reduce chemical fertilizer in the agriculture becomes an important issue. PGPR have been commercialized and applied as biofertilizer to various crop to enhance growth and yield. (Dey *et al.*, 2004). This PGPR lives naturally and enhance the growth of plants by producing growth hormones, making the essential nutrients available for plants by solubilizing them, protect plants against pathogen and they are also involved in cleaning up the environment. (Marschener *et al.*, 1994; Trivedi *et al.*, 2003). During last few decades number of rhizobacteria belonging to genus *Pseudomonas,*
Azotobacter, Azospirillum, Klebsilla, Enterobacter, Alcaligens, Bacillus, Burkholderia and Serratia have been reported to increase plant growth. (Glick, 1995; Kloeper et al., 1989). Thus utilizing biological nitrogen fixation technology by applying the PGPR can reduce the use of urea nitrogen and prevent the depletion of soil organic matter. (Choudhary and Kennedy, 2004). With the increase food crises public recognition and attention towards fertilizers seems to be the solution of food security in the world. In many countries there was inadequate and unbalanced fertilization to the crops decreases the crop yield. (Roberts, 2009). The main factor affecting the yield of rice productivity are soil texture, pH, moisture, erratic rainfall pattern, weed management system, low fertilizer use etc. (Tripathi, 2002). Also illiteracy, poverty, lack of source and supply of good quality of seed, lack of knowledge of quality and doses of fertilizers affect the crop yield. Thus it can be said that the farmer’s socio economic status is poor. They were unable to adopt improved production technology although facing low rice production as compared to other rice growing regions. (Sanger, 2002). Agriculture practice includes the use of chemical and inorganic fertilizers. Although the use of chemical fertilizers increase the crop yield and productivity but if used excessive and regularly it adversely affects soil fertility i.e. make the soil infertile and also pollute soil environment which contributes to the human and animal health problem as well.

Due to the continuous cropping without proper supply of organic manure there occurs deficiency of essential nutrients in the agriculture lands. Also the nutrients present in soil are in unavailable form. Plants cannot utilize these nutrients in this unavailable form. Therefore the application of nutrients in the form of chemical fertilizer is necessary to get optimum yield of crop. In chemical fertilizers pure synthetic nutrients are present which facilitate growth of plants in initial
application. (Steiner et al., 2007). The chemical pesticide and fungicide are also used for crop protection from various pest and fungus pathogen. In past four decades the use of agrochemicals as fertilizers, pesticides and fungicides doubled the grain production but it also results in unsustainable agriculture. Most of the agrochemicals are not absorbed by plants they lost by surface runoff or reaches to ground water. (Renck et al., 2004).

Therefore the recent research is attentive towards the use of beneficial rhizobacteria in agriculture which increases the crop yield, protect environment and minimize the chemical fertilizer input. In this context the present work was undertaken to isolate the potent plant growth promoting rhizobacteria of rice crop. The work also focused the effect of chemical fertilizer urea on the growth of potential PGPR.

The present study is divided into six chapters. Chapter 1 focused on general introduction of plant growth promoting rhizobacteria, rice plant and its nutritive value, urea and its effect. Objective of the present work was also presented in this chapter. Chapter 2 presents the complete review of Plant growth promoting rhizobacteria, rice plant and application of urea. Chapter 3 presents the methodology for isolation, characterization identification and screening of growth promoting properties of plant growth promoting rhizobacteria and effect of urea on its growth. Chapter 4 presents the results with discussion of the methodology performed. This chapter includes the tables and graphs. Chapter 5 focuses the summary of the work and Chapter 6 gives the conclusion and future prospects.
Study Area

The study area selected for present study is District Durg of Chhattisgarh State, India. It is one of the densely populated districts situated in the northern part of Chhattisgarh plains. Total area covered by district is 2238.36 sq km. Geographically the Durg district lies between 20º54’ and 21º32’ North latitude and 81º10’ and 81 º 36’ East longitude. It is 317 meters above mean sea level. The district is bounded by Bemetara district in north, Rajnandgaon district in west, Balod district in south and Raipur and Dhamtari in east. Climate of the district is tropical type. Summer is a little bit hotter. Rise in temperature begins from the month of March to May. May is the hottest among others. Annual average rainfall of the district is 1052 mm. During the year most rainfall occurs during monsoon month i.e. June to September.
Objective

The present studies will be undertaken to isolate and screen the PGPR strain that are compatible with rice for its various plant growth promoting properties in Durg district of Chhattisgarh and also to investigate the effect of different doses of urea on it.

Following are the objectives of the proposed work

- To isolate the bacteria from rice rhizospheric soil of selected agricultural field and identify plant growth promoting rhizobacteria from them.

- Characterization of the PGPR isolates.
  
  (a) Morphological Characterization.
  
  (b) Characterization for motility.
  
  (c) Biochemical characterization

- To investigate the growth behavioral pattern of PGPR isolates under different temperature, pH, light and dark condition.

- Screening of PGPR isolates for auxin production, phosphate solubilization and siderophore production.

- Application of different doses of urea on PGPR isolates to investigate LD<sub>50</sub>.

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