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International Journal For Research in  
Applied Science and Engineering Technology



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# INTERNATIONAL JOURNAL FOR RESEARCH

IN APPLIED SCIENCE & ENGINEERING TECHNOLOGY

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**Volume: 4      Issue: VI      Month of publication: June 2016**

**DOI:**

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# **Effect of coal mine waste on protein activity of *Vigna mungo* (L.)**

Rohini Kumari Senapati<sup>1</sup>, Bandita Deo<sup>2</sup>

<sup>1,2</sup> *Plant Physiology and Biochemistry Division, Regional Plant Resource Centre, Nayapalli, Bhubaneswar 751015, Odisha, India.*

**Abstract-***The present study was carried out at Regional Plant Resource Centre, Bhubaneswar, Odisha, India to study the effect of coal mine waste on the protein activity of black gram (*Vigna mungo* L.). The experiment was carried out as a pot culture experiment using different proportion of coal mine waste and garden soil in the ratio of 0:4 (T), 1:3 (T1), 2:2 (T2), 3:1 (T3), 4:0 (T4). The protein contents were between 6 mg/ml to 8 mg/ml in all the plants after 15 days of planting which increased till 45<sup>th</sup> day but drastically dropped to 2.48 mg/ml (T4) and 4.81 mg/ml (T) after two months.*

**Keyword:** *Coal mine waste, *Vigna mungo* L., protein.*

## **I. INTRODUCTION**

The environmental impact of the coal industry includes issues such as land use, waste management, water and air pollution, caused by the coal mining, processing and the use of its products. In addition to atmospheric pollution, coal burning produces hundreds of millions of tons of solid waste products annually, including fly ash, bottom ash, and flue-gas desulfurization sludge, that contain mercury, uranium, thorium, arsenic, and other heavy metals. Heavy metals are essential for the normal growth and metabolism of plants and play very important role in the biosynthesis of some enzymes and growth hormones [4] but the growth and metabolism of many plant species were reported to be affected adversely by excess supply of heavy metals [3]. Emission of particulate matters and noxious gases cause environmental hazards because the ash comprises of SiO<sub>2</sub>, Al<sub>2</sub>O<sub>3</sub> and oxides of iron and other toxic metals. These oxides change the chemical and biochemical composition of the plant grown on fly ash [6]. Heavy metal pollution is one of the most serious environmental problems which have been a subject of extensive research in recent years [2]. Experiments on the toxicity of mixtures of pollutants may reflect the actual toxicity to ecosystem in a more realistic way than experiments in which toxicants are tested individually [7], and several studies have investigated the combined effects of heavy metals on certain plant species. Presence of Cd, Pb and Zn in polluted mining soils and their uptake by plants were also studied [5]. Natural processes of ecosystem developing including accumulation of organic matter and nutrient in these habitats is a slow process. These waste dumps still have high metal concentrations and continued to be a source of metal pollution long after extraction activities were ceased [11]. Black gram (*Vigna mungo* L.) is an important short duration pulse crop grown in many parts of India, cultivating both in Kharif and Rabi season. As an excellent source of plant protein it is cultivated extensively in the tropics and subtropics. The crop has special importance in intensive cropping system for its short growing duration. The crop is potentially useful in improving cropping system as a catch crop due to its rapid growth and development. Black Gram is a perfect combination of all nutrients which include 20 to 25% proteins, 40 to 47% starch, ash, fats, carbohydrates and essential vitamins.

## **II. MATERIAL AND METHODS**

### *A. Collection of seeds*

The seeds of black gram (*Vigna mungo* L.) used in the experiment were collected from Odisha University of Agriculture and Technology (OUAT), Bhubaneswar, Odisha.

### *B. Collection of mine spoil and garden soil*

Soil samples were collected from South Bolanda coal mine site and transported to the experimental site at Regional Plant Resource Centre, Bhubaneswar. Control or garden soil was also collected from the experimental site, Bhubaneswar, Odisha. Here control (garden) soil and coal mine soil were used as the growing medium in the proportion of 0:4 (T), 1:3 (T1), 2:2 (T2), 3:1 (T3), 4:0 (T4) respectively.

### *C. Pot culture experiment*

The experiment was carried out for 3 months at RPRC, Bhubaneswar, India. 10 inches earthen pots used in the experiment were cleaned thoroughly and filled up with different doses of coal mine waste and garden soil as mentioned above. 20 Seeds in each pot were sown directly in the pots at about 1-2 cm depth. The pots were kept in a light controlled green house with 12 hour photoperiod

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and day / night temperature of 30/18<sup>0</sup>C. Twenty seeds were sown per (each pot) treatment and the treatments were replicated three times. Sampling was done at random at 15 days interval from the treated and untreated pots for study. The experiments were repeated three times with 5 replicates per treatment. Plants were watered to field capacity daily. Plants were thinned to a maximum of five per pot. The data for various morphological parameters such as, root and shoot length, number of nodules, dry weight of root and shoot were also recorded.



Fig. 1- Black gram planted in pot.

### D. Estimation of protein

The protein estimation of blank gram (*Vigna mungo* L.) was done by following Bradford reagent method [1].

#### Isolation and extraction of protein-

500 mg of leaf sample were taken in pre-chilled mortar and pestle from all the five treatments including control and different concentration of coal mine waste. Then 30 ml of 2 % pvp were added to it. Then they were grinded with 0.5 ml of cold extraction buffer (25 mM tris HCl, pH 8). Then it was transferred to a 1.75 ml pre-chilled eppendorf tube. The mortar was washed with 1 ml of extraction buffer to the tube. Then the homogenate were centrifuged at 10000 rpm for 20 minutes at 4<sup>0</sup> c. then the supernatant was transferred to another eppendorf tube stored at 4<sup>0</sup>c. Then they were added with 5 ml of Bradford reagent and absorbency was read at 595 nm in a uv-vis spectrophotometer. Then a curve was drawn with the resulted data values.

### III. RESULT

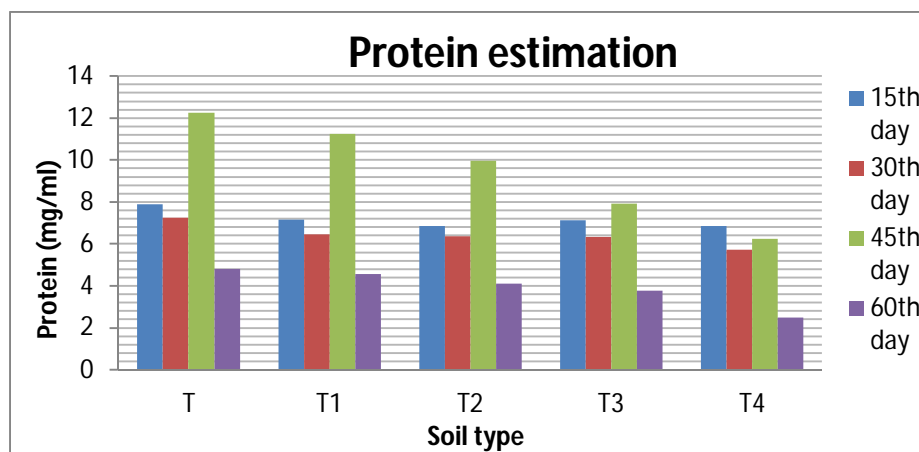


Fig. 2- Protein estimation of *Vigna mungo* L. (in mg/ml).

From the above data it was observed that the protein content of the plants grown in garden soil (T) was high on each observation than the plants grown in mixed mine soil (T1, T2, T3, T4). In all treatments protein content increased till the 45<sup>th</sup> day. After two months the adverse effect of coal mine waste was marked in plants grown on T4 had 2.48 mg/ml protein, which was much less in compared to plants grown in control garden soil, T (4.81 mg/ml) (Figure 2). Protein content of control soil is high as compared to Mine waste. Likewise, Koziol and Cowling, (1980) [10] reported that protein content was low in polluted area. The protein content of varies from species to species in same genus. The amount of protein varied even in the same species according to phytochemical

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conditions. These mine spoils are not suitable for plant growth because of low organic matter content, unfavorable pH, and drought arising from coarse texture or oxygen deficiency due to compaction [8]. The other limiting factors for revegetation of mine spoil may be salinity, acidity, poor water holding capacity and inadequate supply of plant nutrients [9].

### IV. CONCLUSION

From the above study it was concluded that the coal mine waste helps in the growth of black gram plant to a certain extent and then gradually the growth and metabolism of all the plants reduced. The protein contents were between 6 mg/ml to 8 mg/ml in all the plants after 15 days of planting which increased till 45<sup>th</sup> day but drastically dropped to 2.48 mg/ml (T4) and 4.81 mg/ml (T) after two months.

### V. ACKNOWLEDGEMENT

The authors are thankful to Regional Plant Resource Centre, Dept. of Forest and Environment, Govt. of Odisha, India for providing facility to carry out the research work.

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