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Research Article

Biosynthesis of Food components in *Abolmosclus esculentus* under environmental stress; Effect of addition of vegetative materials

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Abstract: A study on the respond trends of protein and carbohydrate synthesis in *Abelmoshus esculantus* grown under environmental stress (oil pollution) was carried out at the University of Port Harcourt botanical garden. *A. esculatus* was grown at two level of oil pollution (severe and mild) at 200 and 400ml respectively. Vegetative materials- sawdust and chromolena leaves at the rate of 50g each, were added to both mild and severe pollution treatment and control. Data was collected on two weeks intervals for three months on the protein and carbohydrate accumulation in *A. esculantus* and subjected to statistical analysis. Result showed that stress condition (oil pollution) retards biosynthesis of the food components while vegetative materials enhance its accumulation in *Abelmoschus esculantus* and variations exist among the treatments.
Keywords: *Abelmoshus esculantus*, chromolena leaves, oil pollution

INTRODUCTION

The quest for man's health and satisfaction has led to pressure on the environment through its activities such as generation of pollution or by natural events including occurrence of droughts. Oil pollution dumped harmful chemicals into the soil which caused environmental stress to plants and animals. The growth of plants under such conditions differs compared to when grown in a greenhouse. This implies that pollution arising from oil, pose greatest problem in the world and in Niger Delta area of Nigeria. These effects are increasing everyday causing grave and irreparable damage to plants and animals, leading to concerns on the subject of crude oil pollution especially on arable agricultural land. Moreso, environmental stress condition arising from crude oil pollution destroy vegetations that provide food and shelter thereby disrupting the balance of nature and in extensive cases cause the death of crops and humans [1], hence sustainability and competitiveness has become keywords in synthesis of food components in crops [12].

The fact that environmental stress can lead to retardation to synthesis of major food components, through its ability to reduce aeration, a reduction in the level of available plants nutrient necessary in protein and carbohydrate synthesis [2] and a rise in the toxic levels of certain elements such as Calcium, Copper, Maganese, and Iron [3]. Calls for full scale remediation strategies that are less cost effective and expensive, environmentally friendly and easily affordable and start-up.

The use of biological product consisting of organic materials (plant and animal residues) will be more competitive as it does not only make the soil amenable but also provide necessary environment for biosynthesis of food components in a plant.

From literature, organic materials contribute to plant growth through its effects on the physical, chemical and biological properties of the soil. Its activities under environmental stressed soils have been noted, through its direct and indirect roles on the availability of nutrients needed for microbial degradation and disintegration of hydrocarbon molecules. In addition to serving as a source of nitrogen, phosphorus and sulphur through mineralization by soil micro-organism, organic components influence the supply of nutrient from other sources. (For example it is required as an energy source for nitrogen fixing bacteria).

Organic material also has profound effects on the structure of many soils. Soils adequately supplied with humus from vegetative materials, has less consequences on the structure, reducing its hardness, competitiveness and cloudness. In view of this, aeration, water holding capacity and permeability are all favourably affected. From the works of Offor and Akonye [4], Offor, Iyagba, and Onwugbuta [5], all

these have direct consequences on the biodegradation of hydrocarbon and metal accumulation in soils.

The use of sawdust and chromolaena represent such vegetative components with such characteristics that has been previously tested on cereals and most legumes for soil amendments capable of increasing the synthesis of protein and carbohydrate in plants. Extra tributes such as ability to uphold oil for longer time and blending of soil humus will ensure healthy growth and further biochemical processes in crops.

In the Niger Delta area of Nigeria, Legumes/Arable crops constitute these major crops cultivated and consumed in the area where pollution is prominent. *Abelmoschus esculentus* in this respect represent one of the greatest fruit and vegetable crops consumed by the average inhabitants of the Niger Delta. From literature, [6] *Abelmoschus esculentus* L. moench is believed to have originated from tropical Africa or possible Asia. It is presently widely distributed in tropical Asia (Malaysia, Indonesia, India, Philippines, China etc). Central East, West Africa and the Caribbean (Trinidad and Pueto Rico) and generally in the tropics. It is an annual herb growing up to 2mm in height with heavy stem which is also woody at maturity. The crop is propagated by seeds which are frequently soaked for 24 hours before being sown in deeply cultivated soil or ridges or beds. Seed will only germinate in relatively warm soils. A wide range of soils types has been found suitable well drained fertile soils with an adequate organic material contents and resources of major elements are suitable. The pods may be harvested 60 – 80 days after planting, about 5 – 10 days after flowering depending on the variety/cultivar grown.

The immature fruits of *A. esculentus* are eaten as boiled or fried vegetable. They are popular for adding to soups and stews and sometimes are dried, powdered and used as flavoiring. Young shoots and leaves are also edible and the mature seeds contain 20% of edible oil. Any investigation that will lead to improvement in the synthesis of the major food components (protein and carbohydrate) in this crop under stress condition is a welcome development hence this research.

MATERIALS AND METHODS

Seeds of *Abelmoschus esculentus* was procured form the Green River Project of the Nigerian Agip Oil Company, Ebocha. The crude oil used was also supplied by Agip Oil Company, Ebocha base, Port Harcourt, Rivers State. A good garden soil weighing approximately 6600g was obtained at the botanical garden of the University of Port Harcourt and was used to fill black cellophane bags of equal diameter measuring about 50cm and height 45cm leaving a space of 7.00cm from the top end of the polythene bags to make allowance for crude oil or addition of the vegetative materials. Heavy looking seeds previously tested for viability and germination test were soaked in alcohol for 30secs and then soaked in water for 24 hours before being sown deeply in the cellophane bags about 4 – 5 stands per bag in a bag of four (4) per treatment 200 and 400ml of crude oil representing two levels of pollution (3 and 6% mild and severe), were added and thoroughly mixed with the soil using a hand trowel. The vegetative materials – sawdust and chromoleana leaves each measuring 50g were added to the two levels of oil pollution which was sub-divided with three batches viz – pollution with presence of sawdust, pollution with presences of Chromolaena leaves and pollution without vegetative components. The treatments were represented as follows:

- 6% (severe) pollution
- 6% pollution with sawdust
- 6% pollution with chromoleana leaves
- 3% (mild) pollution
- 3% pollution with sawdust
- 3% pollution with chromoleana leaves
- Control
- Control with sawdust
- Control with chromoleana leaves

Food Components

- Determination of Carbohydrate conduct
- Determination of protein contents

Determination of carbohydrate content.

The leaf sample was analyzed for carbohydrate content using the cleq Anthrone method 1. 08 of the samples were dissolved in 13m I of 52% (v/v) perchloric acid. The mixture was allowed to stand for 30 minutes and then diluted to 250mI with distilled water. 10mI of the filtrate of the above solution was pipetted and made up to 100ml with distilled water. 1ml of this was then placed in test tube into which was placed 5ml of 0.1% anthone solution in concentrated· sulphuric acid. The resultant colour was absorbed at 630mml wave length against a standard glucose solution of 1 mg/ml concentration using SP6 pye Unican spectrophotometer.

The concentration of the carbohydrate was calculated by using formula:

$$Percentage\,carbohydrate = \frac{25 \times absorbance\,of\ Samples}{Weight\,of\ Sample \times absorbance\,of\ s\tan dard\ glu\cos e}$$

Determination of total Nitrogen and crude protein

The leaf sample were analysed for total Nitrogen content using kjedahl methods [7]. The digestion reagents comprised of a catalyst blend of 1.09 Cu $S0_4$ $5h_20$ and $15.OgN_{a2}$ $S0_4$ with concentrated Sulphonic acid. Leaf samples were placed in 500ml Kjechahl flask. Then 2. Og of catalyst blend and anti bumping chips were added. 25ml of concentrated sulphuric acid was added to each sample. The mixtures were heated gently (to avoid frothing) in an electro-thermal kjedahl digestion unit. The heating temperature was increased until the charred particles disappeared and the mixture become clear. The digestion was completed after 2 hours, the flasks were left to cool and the digests were transferred into 100 ml volumetric flask and were made up to the 100ml mark with distilled water. By steam distillation in the presence of excess alkali (NaOH) free ammonia was librated from the digestible sample. 25ml of 2% boric acid solution ($H3B0_4$) containing 3 drops of methyl red indicator were added to a 1O0ml beaker. The beaker was placed under the *tip* of-the condenser from a distillation unit ensuring that the outlet tip was below the surface of the boric acid solution. The condenser was then connected to a cold water supply.

An aliquot of the 2.5ml of the digest was added to the kjedahla flask and to this was added 40ml of NaOH solution. The flask was then quietly connected to the distillation unit in order to avoid loss of ammonia. The electrical unit was stopped after collecting thrice the original volume by which the colour of the solution had changed from pink to light green.

The volume collected was titrated against 0.1 NHCL in a conical flask until the first appearance of permanent pink colour was observed, the total nitrogen present was calculated as follows:

$$\text{Percentages Nitrogen} = \frac{T.V \times 1.4 \times 50 \times 100}{0.1 \times 20 \times 100}$$

Where T.V = Nitrogen volume (Titre)

1.4 = Nitrogen equivalent of the morality of the HCL used in the titrametric analysis.

50 = Total dilution of the sample volume

0.1 = Dry weight of the sample

0.2 - Volume of *the* aliquot used in *the* analysis

100 = conversion factor from gram to milligram.

Total protein

This was calculated by multiplying the value of protein nitrogen by 6.25 (since nitrogen constitute 16% of protein)

Data analysis: Data collected were subjected to analysis of variance and Duncan multiple range text (DMRT) was employed to separate means according to the procedure of statistical analysis system SAS (1991) the standard error bars (SE) is at 5% probability ($P<0.05$)

RESULTS AND DISCUSSION

Fig-1 present Crude protein (Total Nitrogen) content of *Abelmoschus esculentus* as influenced by treatments. At severe pollution (6%), differences in crude protein synthesis and its subsequent accumulation with time was evident from the 4^{th} week in contrast to what was obtained at 6% pollution applied with sawdust. Significant differences in biosynthesis of crude protein and its accumulation in *Abelmoschus esculentus* at 6% polluted soil amended with chromoleana leaves and 3% pollution was not evident. Similarly, crude protein accumulation at 3% pollution and control with vegetative materials (sawdust and chromoleacus leaves) were relatively equal.

The Results in Fig. II on total protein synthesis and accumulation in *Abelmoschus esculentus* showed significant increase in all treatments except at 6% and 3% pollution without vegetative materials. With time, significant differences were noticed from the 4^{th} and 6^{th} week at $p < 0.05$.

In Fig. III, the carbohydrate content synthesis and its subsequent accumulation in *Abelmoschus esculentus* were significant in all treatments with presence of sawdust and chromoleaus leaves from the 4^{th} week at $p < 0.05$. Also significant increase in carbohydrate accumulation was more evident at control with chromoleana leaves. Annova presents a highly significant treatment effects at $P < 0.001$.

From the results obtained in Fig. I and II, the crude protein and total protein were significantly inhibited at 6% pollution and treatments with sawdust at the early stage in comparison to controls. Addition of chromoleana leaves to the same pollution level significantly increased protein content. Comparatively, the respond trend at 3% pollution treatments was similar to that of 6% pollution treatment. The significant increase resulting from the addition of chromolaena leaves may have a direct relationship with its constituents [8,4,9]. Moreso, at mild oil concentration, it is assumed that the concentration of oil is minimal hence promotion of protein synthesis and its subsequent accumulation in *Abelmoschus esculentus* could be possible. The later increase in treatments with sawdust comparable to control can also be attributed to slow degradable activities inherent in this type of vegetative material used, which releases essential nutrient capable of biodegradation and nutrient supply in a slow rate in plants. This is logical as confirmed by Amakiri [10].

The result on carbohydrate synthesis and accumulation in Abelmoschus *esculentus* showed a clear inverse relationship between the vegetative materials used and the level of pollution. These finding are consistent with our previous works [2]. Furthermore, inhibition of carbohydrate has been associated with increase in oil concentration (environmental stress) causing a retardation in photosynthesis and subsequent decrease in carbon dioxide assimilation in plants since carbon-dioxide is a product of photosynthesis.

The control treatments gave higher significant increase in carbohydrate than at 6% and 3% pollution treatments. This is logical and expected since small traces of oil are found within the treatments that may inhibit carbohydrate synthesis and accumulation in *Abelmosochus esculentus.* From this work, application of chromolaena leaves and sawdust increase carbohydrate content than at pollution without amendments, this is supported by previous findings in Lynch[11], Offor, Akonye and Asouzi [2] etc, but the extent of enhancement varies due to its differences in assimilable qualities during their decomposition process as early stated. It is then expected that chromoleana leaves will perform better than sawdust.

This study, a follow-up to our previous study has shown that protein and carbohydrates in fruit and vegetables crops under stress conditions can be improved when vegetative materials are applied to soils.

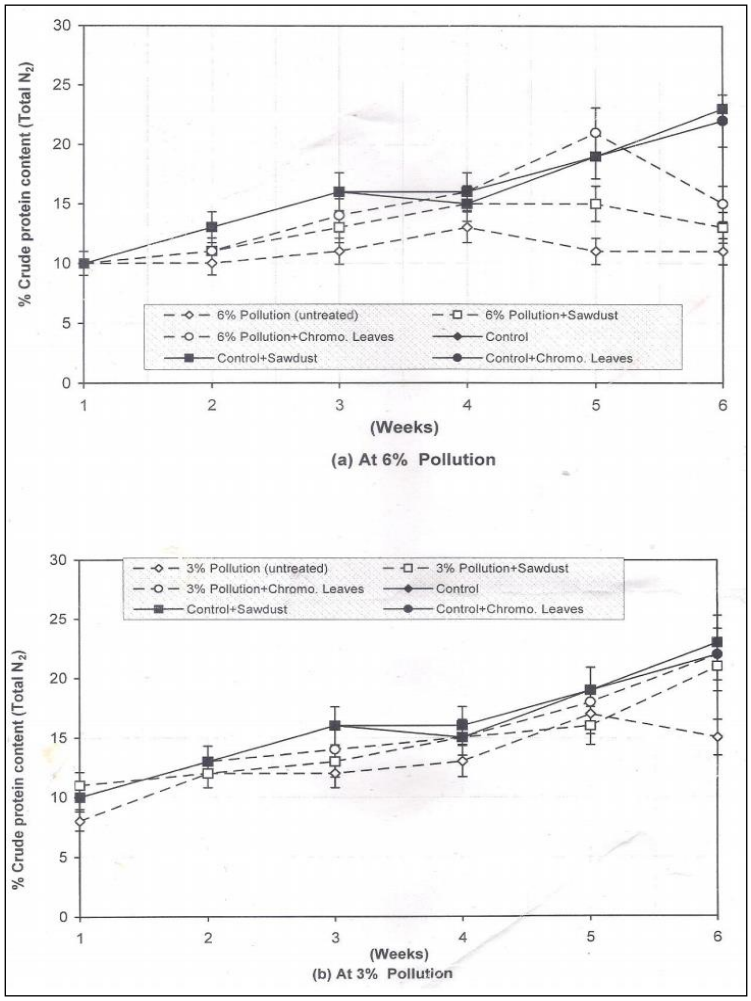

Fig-I: Influence of treatments on the percentage crude protein (Total N_2) content of *Abeimoschus esculentus* (L) Moench

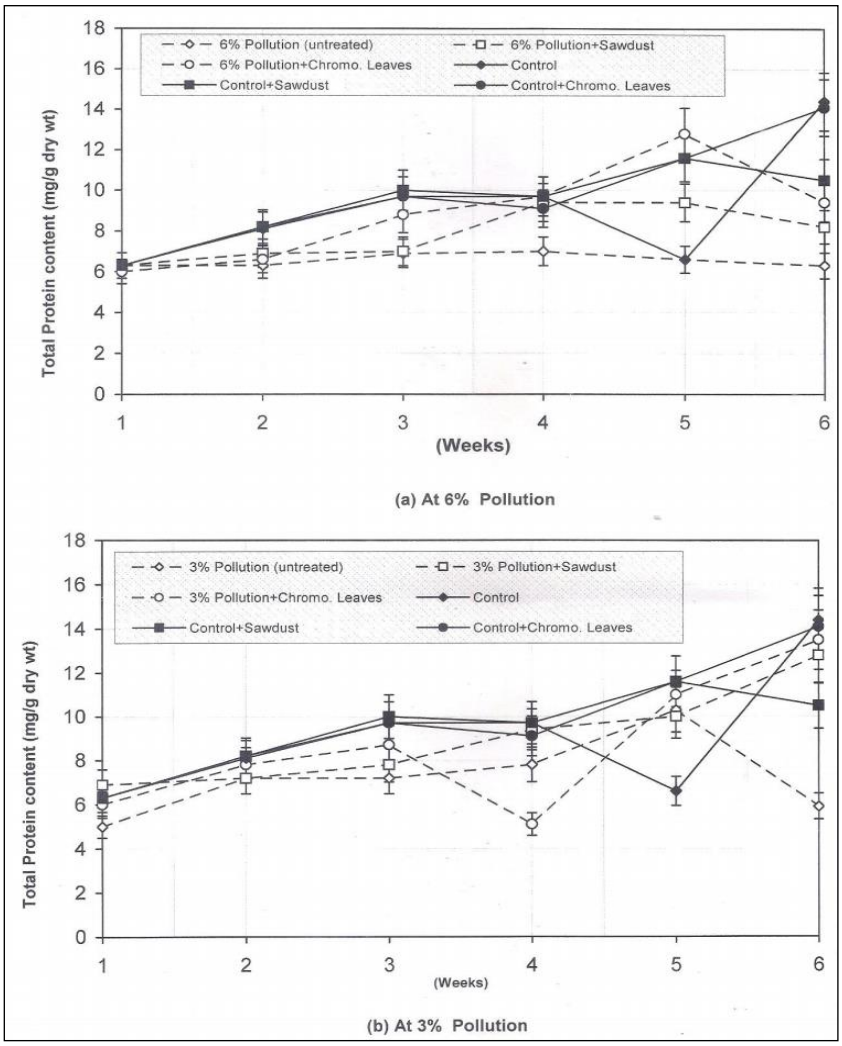

Fig-II: Influence of treatments on the Total protein content of *Abeimoschus esculentus* (L) Moench

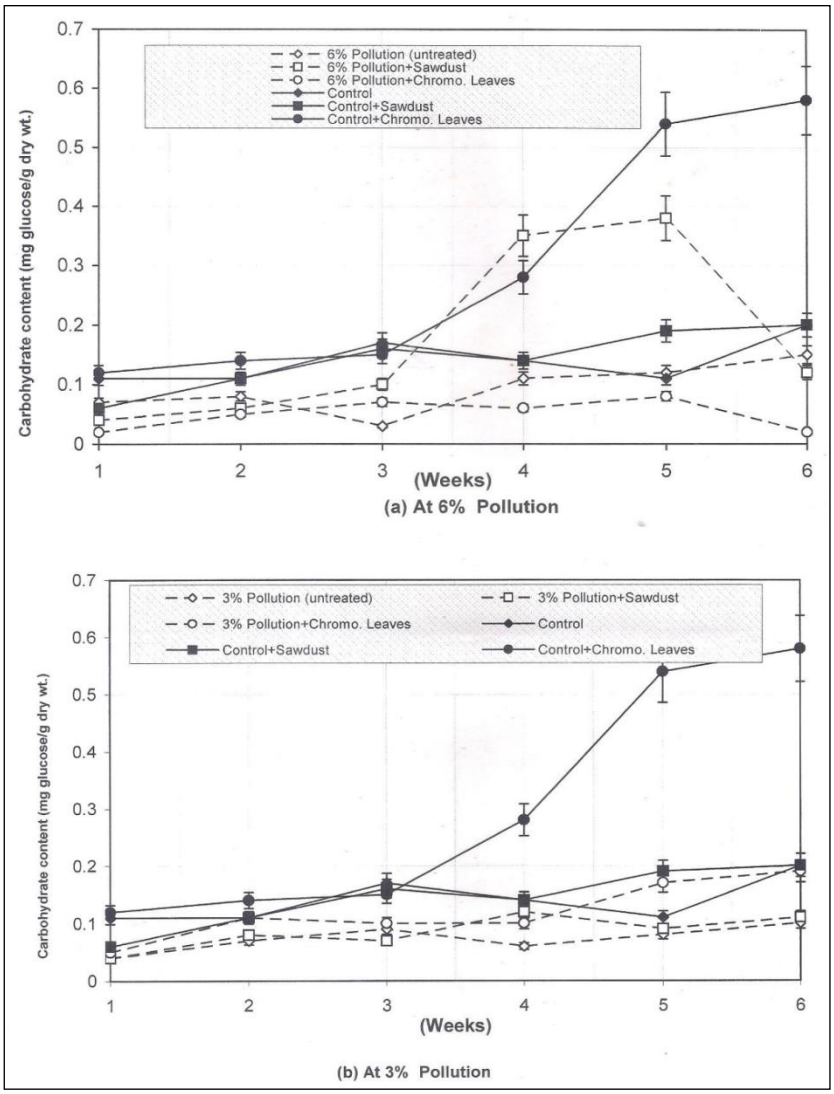

Fig-III: Influence of treatments on the Carohydrate content of *Abeimoschus esculentus* (L) Moench

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