A Review on Novel Approach towards the Methods for Detection of Growth Parameters in Plants
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INTRODUCTION

The quantification of chlorophyll in higher plants has been performed using a non-destructive chlorophyll meters such as fluorometry, photo-acoustic spectroscopy, chromatographic techniques, and spectrophotometry[1]. With the advantages of intuitiveness, convenience, and ease, spectrophotometry has been the classic method for chlorophyll determination in the leaves of terrestrial plants and in chlorophyll determination of other photosynthetic organisms. Cadmium and nickel are acting as heavy toxic metal that affect the growth of plants by alternating the normal functioning mechanisms of plants. It also causes the environmental pollution that disturbed the natural ecosystem of environment[2]. It is produced by Industries that are the major source of environmental pollution. Its higher concentration in plants leads to toxicity and death of plant tissues[3] . It also interferes with mineral elements in plants by increasing the activity of enzyme such as superoxide dismutase. Cadmium enter into the root of plant cells by disturbing the overall growth of plants[4].

Inductively coupled plasma-optical emission spectroscopy used to find out the concentration of plant phosphorus and cadmium. It is used in the laboratory testing for detection of chemical elements in plant samples[5]. Inductively coupled plasma-optical emission spectroscopy used to find out the concentration of plant phosphorus and cadmium[6]. Coomassie brilliant blue staining method is used commonly to determine the concentration of soluble proteins in plant samples. Biological activity of enzyme superoxide dismutase is measured by spectrophotometer at absorbance 485 nm. Lipid peroxidation in plant tissues determined by measuring the malondialdehyde formation using the thiobarbituric acid method. This method is based on spectrophotometric principle and red-violet complex is formed by the formation of malondialdehyde. Root activity in plant tissues determined by using the triphenyltetrazolium chloride test. The absorbance is then measured by spectrophotometer at absorbance 485 nm.

Phytoremediation is mostly commonly used method to remove toxic metals from the plants that are affected by heavy metals stress especially cadmium. Lipid peroxidation in plant tissues determined by measuring the malondialdehyde formation using the thiobarbituric acid method[8]. This method is based on spectrophotometric principle and red-violet complex is formed by the formation of malondialdehyde. Root activity in plant tissues determined by using the triphenyltetrazolium chloride test. Coomassie brilliant blue is a dye that bind with specific proteins in plant...
samples through ionic interactions and used for staining the specific proteins[9]. Phytoextraction is a method of phytoremediation that supported the plants for accumulating toxic metals in their special organs such as root system.

Salix is most commonly used for metal phytoremediation in hydroponic as well as field experiments. The common name of salix is wood that helpful for plants for metal decontamination[10]. Phytoextraction of heavy metals especially cadmium can be improved by supplying the optimum concentration of elements such as nitrogen and phosphorus. Enrichment of nitrogen and phosphorus in proper concentration provide advantage to phytoremediation plants for removal of cadmium. This process leads to trap toxic metals and finally degraded them[11].

The sample preparation included that plant sample cuttings transferred to a normal water tank with six to ten small chambers. Hoagland nutrient provided and proper nitrogen and phosphorus concentrations regulated[12]. Then root cuttings cultivated in a greenhouse under a photoperiod of 16h temperature at 25 °C/20 °C in day/night[13]. After 6 weeks, cuttings harvested and root cuttings selected randomly. Root cuttings harvested and deionized water used to wash them. Root cutting further divided into roots, and initial cuttings. Growth measurements such as length, number, dry and fresh weights of shoot and root measured precise[14].When suitable amount of nitrogen and phosphorus added to the growth medium, cadmium accumulation started effectively seen in plants such as Epilobium laxum Royle. Cadmium accumulation increases in when plants grown in hydroponic conditions by supplying the nutrients[15]. Proper supplementation of ammonium ion (NH₄⁺) increases the trapping or accumulation of cadmium in some plants such as sunflower, potato when these plants ideally grown in soils. It has been observed that application of nitrogen fertilizers lead to effective binding to cadmium and decontaminated them seen as in S. plumbizincicola[16].

The study remains unclear about methods measuring the growth parameters in Salix species. The aims of this review most included the advanced methods that are used recently for measuring the growth parameters in Salix species. Accumulation of nitrogen and phosphorus is crucial and important for investigating the Salix spp cuttings. Mechanism remains unclear how phytoremediation plants show physical responses by nutritional supplementation of nitrogen and phosphorus under hydroponic conditions. Changes the concentration of nitrogen and phosphorus for trapping of cadmium using salix plants not completely understood.

Detection of Phosphorus and cadmium Concentrations

ICP-OES basically used to find the concentration of elements epically heavy metals in a particular sample[17]. It is used in the laboratory testing for detection of chemical elements in plant samples. Inductively coupled plasma-optical emission spectroscopy based of principle of emission spectroscopy in which plasma is produced by excitation of the atoms. Inductively coupled plasma-optical emission spectroscopy used to find out the concentration of plant phosphorus and cadmium. Plants samples such a root, shoot and initial cutting grinded[18]. Nitrogen concentration detected by using the laboratory analyzer such as inductively coupled plasma-optical emission spectroscopy. The most commonly gas used is the argon gas in Inductively coupled plasma-optical emission spectroscopy[19].

Detection of Soluble Proteins

Coomassie brilliant blue staining method is used commonly to determine the concentration of soluble proteins in plant samples. Coomassie brilliant blue is a dye that bind with specific proteins in plant samples through ionic interactions and used for staining the specific proteins. The bands appear as when a dye coomassie brilliant blue gently apply on protein sample[20]. The proteins that contains the slices in the form of gel that incubated with enzyme such as trypsin.
Biological activity of enzyme superoxide dismutase measured by spectrophotometer at absorbance 560 nm. Superoxide dismutase has function against free radicals formation in plants by preventing the long chain that causes free radicals formation. Peroxidase enzyme activity also determined with spectrophotometer at absorbance 560 nm. Peroxidase enzyme has also functions in regulating the oxygen reactive species in plant tissues and thus helps in plant defense mechanism[21].

Fig-2: Coomassie brilliant blue staining method for determination of soluble proteins.

**Measurement of Lipid Peroxidation**

Lipid peroxidation in plant tissues determined by measuring the malondialdehyde formation using the thiobarbituric acid method. This method is based on spectrophotometric principle and red-violet complex is formed by the formation of malondialdehyde[22]. The plants samples collected for testing the lipid contents in specific tissues. TBA reagent bound to the plant sample. As a result of this reaction, dark red colour appears that indicated that reaction is positive. The sample then putted on micro plates and absorbance is noted by spectrophotometer at 532nm[23].

Fig-3: Thiobarbituric acid kit assay for determination of lipid peroxidation

**Detection of Root activity**

Root activity in plant tissues determined by using the triphenyltetrazolium chloride test. During this test, triphenyltetrazolium chloride detected or be appear as a red colour and enzyme dehydrogenase shows biological activity in the root of mitochondrial respiratory chain[24]. Root sample collected from plants, grinded into fine powder to make a thick paste for testing the root activity in plants[25]. The root activity in plants measured within twenty four to forty eight hours. The absorbance is then measured by spectrophotometer at absorbance 485 nm. The most important parameter in this test is that keep avoiding the root samples with fungal infection. If no couour appear in the final sample, then there is no respiration and there is no staining because dehydrogenase appears only in red colour sample[26].

**CONCLUSION**

As accumulation of cadmium is crucial and important for investigating the Salix spp cuttings. This study dectedbe helpful for discovering the mechanism that under hydroponic conditions, phytoremediation plants show physiological and biochemical responses by nutritional supplementation of nitrogen and phosphorus in proper concentrations. This study dectedbe aid in discovering the biochemical basis of using the different concentrations of nitrogen and phosphorus for trapping of cadmium using salix plants.
REFERENCES