

# Effect of Tannery Effluent on Seed Germination, Growth and Development of Cultivable Crops in Ambur, Tamil Nadu

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## Abstract

A study was conducted with two different varieties of paddy (*Oryza Sativa*) ADT-37, ADT-39 and a variety of Red gram (*Cajanus cajan*) LR47 treated with different concentration of chromium (0.05 & 0.1 mM) and effluent (25%, 50% and 100%). The effect of chromium (Cr) and tannery effluent on seed germination, growth and development of crop species cultivable in Ambur Taluk were observed. Germination percentage decreased with rise in tannery effluent concentration. Effluent at 25% concentration induced the growth in root length, shoot length, biomass, photosynthetic pigments, protein and free amino acid. While effluent 50% concentration had reduced the following parameters root length, shoot length, biomass and biochemical attributes as compared to the control and 25% effluent treatment except starch contents. 0.05mM have not shown much effect as compare to control where as growth and biochemical parameter were reduced in 0.1mM Cr concentration, At 100% effluent concentration growth were completely retarded. The results revealed that low concentration of effluent influence the better growth and development of ADT-37, ADT-39 and Red gram (LR-47) at laboratory conditions.

**Keywords:** Tannery effluent, Chromium, ADT37, ADT-39, LR-47.

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## 1. Introduction

The process of tanning consist the transformation of animal skin to leather. The skin is subjected and process to eliminate meat, fat and hide is treated with different chemical such as sodium hypochlorite, sodium hydroxide, enzyme, lime, ammonium salts etc. The semi processed hide is then treated with chromium (Cr<sup>3</sup>) or Cr (VI), tannins, mineral salt are used to obtain leather. The effluent thus generated contains high concentration of sodium sulphates, Fe, Ni, Cr (III) and Cr (VI) and other heavy metals (Mythili and Karthikeyan, 2011). Most of the tannery industries discharge their effluent on to open lands because of high cost of dilution and inadequate treatment facilities. The effective and profitable utilization of the effluent of the industries needs greater attention (Alvarez-Bernal et al., 2006).

Since time immemorial farming is the major source of earning in the semiarid regions like Ambur & Vellore, different crop species are cultivating in Ambur region, among them Paddy and Red gram are highly cultivable crops (major source of carbohydrates and protein). Past few decades, Ambur taluk became hub for tanning industries, one of the major source of income in this region. However farming activities were affected by tannery effluents.

The effect of various industrial effluent and metal elements on seed germination and growth and yield of crop plant have gained the attention of many workers (Rahman, et al., 2002). However studies on seed germination and plant growth and biochemical regulations on paddy and red gram against chromium and tannery effluents are very limited.

This study was aimed to find out the effect of chromium and tannery effluent on seed germination, growth and biochemical characteristic of cultivated crops in Ambur taluk.

## 2. Materials and Methods

### 2.1. Plant Materials

Two varieties of Paddy ADT-37, ADT-39 and red gram (LR-47) variety were used in this study. Seed were collected from Agricultural Extension Centre (Katpadi, Tamil Nadu).

### 2.2. Seed Germination and Treatment

The seeds are washed thoroughly with distilled water, then soaked overnight and next day those seeds were divided into triplicates and placed on the germination paper, raped properly and treated with different dilution of tannery effluent, and two standard concentration of inorganic chromium (Cr 0.1mM, Cr 0.5mM). Each day observations were recorded.

### 2.3. Biochemical Analysis

#### 2.3.1. Chlorophyll Content

Chlorophyll content was estimated by using Arnon method (Arnon, 1949). Shoot of 15 days plants cut and

extracted overnight with 80% acetone, then homogenized and centrifuged next day, absorbance of supernatant were taken at (chl a) 645 nm, (chl b) 663 nm and (chl c) 480 nm respectively.

#### Calculations

$$\text{Chl a (mg/gm fresh weight)} = [12.7(\text{OD}663) - 2.69(\text{OD}645) \times V / 1000 \times W].$$

$$\text{Chl b (mg/gm fresh weight)} = [22.9(\text{OD}645) - 4.68(\text{OD}663) \times V / 1000 \times W].$$

$$\text{Total(a+b)} = [8.2(\text{OD}663) + (20.2 + \text{OD}645) \times V / 1000 \times W].$$

#### 2.3.2. Free Amino Acid Content

Free amino acid was estimated by photometric ninhydrin method as described by Moore and Stein (Moore and Stein, 1948).

#### 2.3.3. Protein Content

Protein contents were measured by using Bradford's method (Bradford, 1976).

#### 2.3.4. Carbohydrate Content

Soluble sugars and starch were estimated by using Mc Cready, Method (Mc Cready et al., 1950).

#### 2.4. Growth Analysis

After germination maintained up to 15 days plant were harvested. Length of shoot, Length of root, weight of fresh shoot, weight of fresh root, and dry weight of both the shoot and root were analysed at the completion of experiment.

#### 2.5. Statistical Analysis

Each experiment was performed in triplicates. Mean average, standard deviation and standard error were derived.

### 3. Results and Discussion

#### 3.1. Chlorophyll Content

The result of present study reveals that higher chromium concentrations adversely influence the chlorophyll content shown in table 1. The varieties ADT-39, ADT-37 and LR-47 treated with 0.1 mM chromium concentration gives less chlorophyll content as compare to control and 0.05 mM chromium treatment. In case of effluent treatment more the concentration of effluent lesser the chlorophyll content, however chlorophyll was absent after 100% effluent treatment. The studies indicate that heavy metals and metalloids have effects on chlorophyll content in plants. Heavy metals are known to hinder with chlorophyll synthesis either through direct inhibition of an enzymatic step or by inducing deficiency of an essential nutrient (Van and Clijsters, 1983).

Table 1. Effect of tannery effluent on chlorophylls content

Effluent dilution	Chlorophyll a and b ( mg/gm)								
	ADT-39			ADT-37			LR-47		
	Chla	Chlb	Total	Chl a	Chl b	Total	Chl a	Chl b	Total
Control	0.438	0.297	0.735	0.279	0.534	0.813	0.460	0.406	0.866
0.05mM Cr	0.161	0.283	0.443	0.18	0.237	0.423	0.392	0.190	0.582
0.1mM Cr	0.130	0.030	0.160	0.131	0.033	0.164	0	0	-
25%	0.644	0.090	0.734	0.215	0.368	0.583	0.486	0.548	1.034
50%	0.237	0.074	0.311	0.202	0.143	0.345	0.266	0.306	0.572
100%	-	-	-	-	-	-	-	-	-

#### 3.2. Free Amino Acid Content

The greatest effect on amino acid content was observed in the varieties treated at 0.1 mM chromium and 100% effluent concentrations (Fig 1). Higher chromium concentration led to drastic decrease in amino acid

content from control to 0.1 mM. If goes through tannery effluent treatment it was observe that the chlorophyll content drastically decreases from 25% to 100% concentrated effluent treatment, at 100%

effluent the chlorophyll content was totally vanished. The highly affected variety was LR-47 at concentration of 0.1mM chromium and 100% effluent concentration.

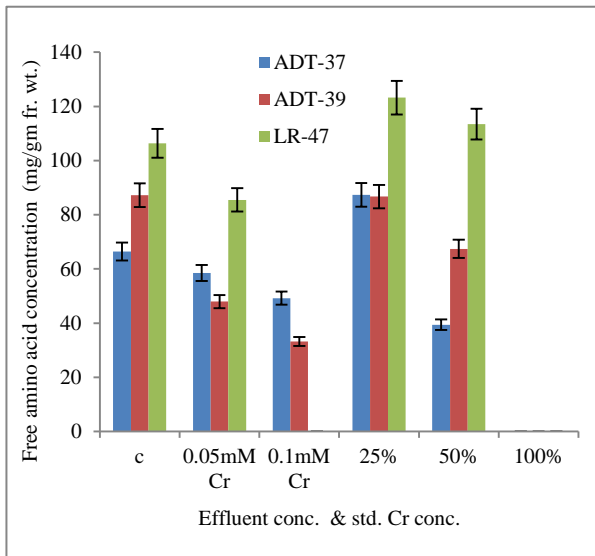


Fig. 1. Free amino acid content in shoot

### 3.3. Protein Content

The effect of the chromium and effluent treatment to ADT-37, ADT-39, and LR-47 by the Bradford's method on protein content was investigated (Fig 2 and Fig 3). As shown in figure 2 the protein content in shoot of plant varieties was decreased as chromium concentration and effluent concentration increased. For plant treated with chromium 0.1mM Cr, protein content were less as compare to 0.05 mM Cr treatment and untreated control samples. After comparing the varieties treated with effluent, there was drastic variation was observed in all varieties at 25% effluent treatment the graph shows no change in protein content when compare to control, however 100% effluent treated varieties shows absent of protein content within plants. The result of protein analysis from the root reveals that the protein content was higher for control and 25% effluent treated LR-47 variety (Fig 3). While treating with 0.1mM chromium the protein content were vanished within the same variety. The protein content pattern remains same for ADT-37 and ADT-39 varieties. The protein gets vanished due presence of heavy metals and metalloids in tannery effluent.

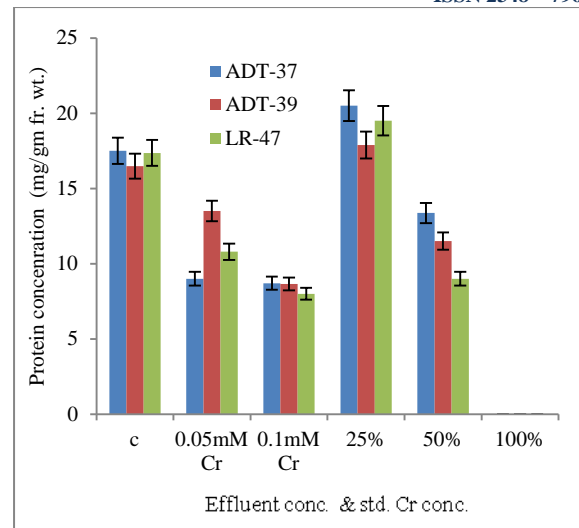


Fig. 2. Protein content in shoot

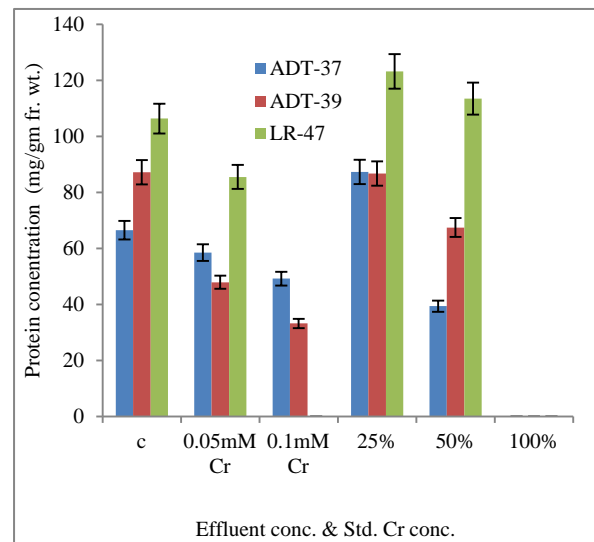


Fig. 3. Protein content in root

### 3.4. Carbohydrate Content

The finding of experiment are shown in fig 4 and 5. The result in fig 4 show the starch content are not significantly affected at control, 0.05mM, 0.1mM treatment and 25%, 50% effluent treatment. At 100% effluent treatment the starch content was affected negatively. ADT-39 paddy variety lessly affect by the chromium and effluent content. Fig no 6 summarize that the starch gets vanished due pesence of chomium and other metal component in efflent. The results were indicate that 0.1mM chromium treatment significantly reduce the starch content of LR-47 to zero, however plant varities treated with 100% effluent reduce starch content to zero. In case soluble sugars there was no significant difference when compare the shoot with root, however the soluble sugar content were affected within the varities LR-47. After treatment of 100% concentrated effluent soluble sugar got deminished from all plant varieteis.

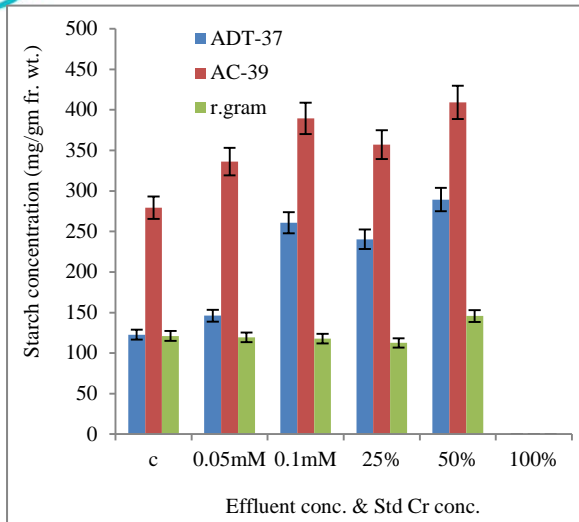


Fig. 4. Starch content in shoot

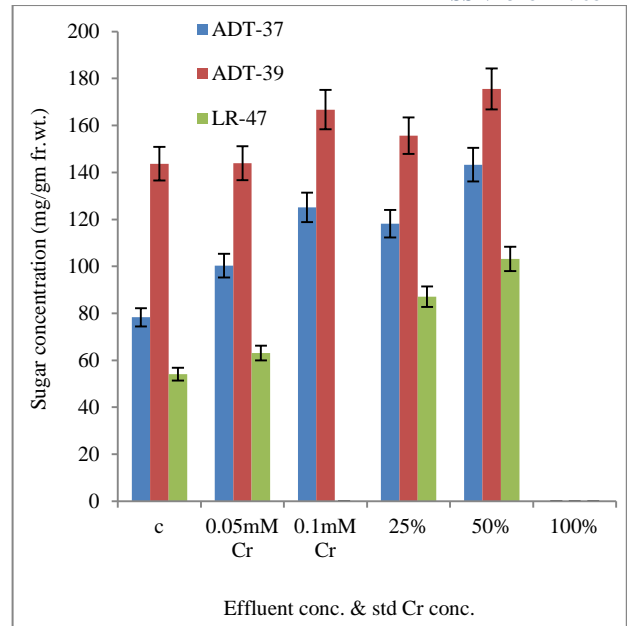


Fig. 7. Sugar content in root

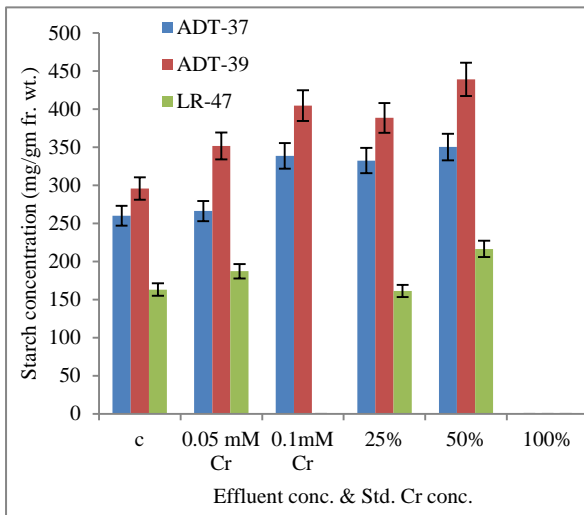


Fig. 5. Starch content of root

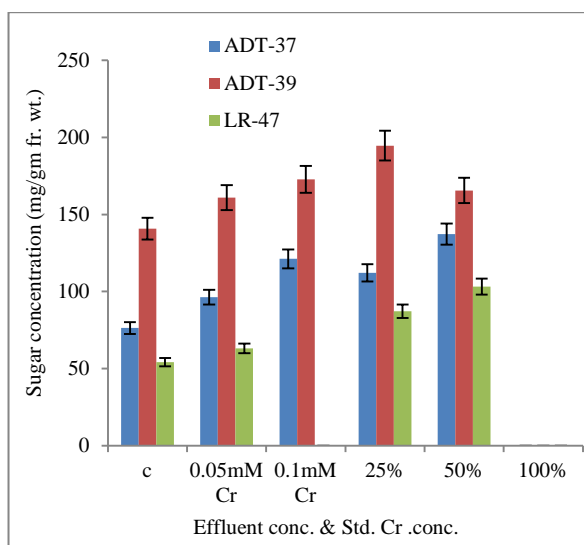


Fig. 6. Sugar content in shoot

## 4. Growth Analysis

### 4.1. Effect of tannery effluent on length of shoot and root

The seed germination was noted for 12 days and 15 days. The results after exposure of chromium and tannery effluent to ADT-37, ADT-39, and LR-47, shows considerable reduction in shoot and root elongation. The shoot length of ADT-39 and LR-17 adversely influence due to chromium and effluent treatment at 12 and 15 days elongation (Table 2). The average shoot length of ADT-37 was not significantly affected at 12 and 15 days time period. Growth of plant varieties were inhibit after 100% effluent treatment. While goes through table 3, it was found that for 15 days growth, root length was significantly effluence.

The chromium treatment shows drastic reduction in root length for ADT-39 and LR-47, at 0.1mM chromium concentration there was no growth of LR-47. After treatment of 100% concentrated effluent, growth of all plant varieties were retarded from the first day of plantation. Several author reported that the inhibition of root length caused by heavy metals may be due to metal interference with cell division, together with inducement of chromosomal aberrations and irregular mitosis (Liu et al., 2003), which can be effected on seedling growth (Samantaray et al., 1999).

Table 2. Effect of tannery effluent on shoot length

Effluent Dilution	Length of shoots (cm)					
	12 <sup>th</sup> day			15 <sup>th</sup> day		
	ADT-37	ADT-39	LR-47	ADT-37	ADT-39	LR-47
Control	5	6.5	17	9	8	20.5
0.05mM Cr	5	5	16	7	6.6	19
0.1mM Cr	4.25	4.5	15	5	5.7	0
25%	9	5	9	9.8	5.5	19
50%	3	3.5	9	4.5	4	12
100%	-	-	-	-	-	-

Table 3. Effect of tannery effluent on root length

Effluent Dilution	Length of Root at 15 days (mm)		
	ADT-37	ADT-39	LR-47
Control	7	6	20
0.05mM Cr	6.5	7	21
0.1mM Cr	6.25	6	0
25%	9.5	6	12
50%	6	5	10.5
100%	-	-	-

#### 4.2. Effect of tannery effluent on fresh and dry weight of shoot and root

The root and shoot fresh weight of ADT-37, ADT-39 and LR-47 were severely affected due to increase chromium and tannery effluent treatment (Table 4 and Table 5). The result predicts that chromium and tannery effluent treatment after 15 days was significantly affecting the fresh and dry weight of both shoot and root. Table 4 and Table 5 demonstrate that, the fresh shoot weight of ADT-37, ADT-39, and LR-47 significantly decreased with increased in chromium concentration. At 0.1mM chromium treatment the fresh weight of shoot and Root of LR-47 reduced to zero. The tannery effluent treatment gradually reduces fresh weight of both shoot and root and at 100% effluent concentration it becomes zero. Dry weight of shoot and root for all varieties obey the same pattern as fresh weight except values. In a study conducted by Vajpayee, it was found that dry matter production affected by chromium concentration above 2.5 m/L Ag in nutrient medium (Vajpayee et al., 2001).

## 5. Conclusion

The results of this study clearly demonstrate that germination was significantly reduced in higher effluent treatment. The adverse effects may be due to the higher concentrations of suspended solids together with different types of dissolved chemicals, as reported by Nesmann (Nesmann et al., 1980). Tannery effluents reduced the photosynthetic material in the plants during their growth and development. The affected plants seem to be pale yellowish in colour as compared to the control plants. Effluents application showed adverse effect on chlorophyll, protein and carbohydrate contents.

The seeds treated with 100% effluent had shown no growth compared with control and 25% diluted effluent (Sangeetha et al., 2012). This indicates that heavy metals which are present in the effluent are responsible for variations in the germinations. The results obtained from the atomic absorption spectrometry of untreated effluent sample, the chromium concentration level was 0.225mg/lit of effluent. So this result indicates chromium concentration is responsible for the variations in germination growth and development of paddy and red gram (ADT-37, ADT-39 and LR-47). Due to the presence of inorganic nutrients in the diluted effluent, the seeds germination, growth and development were maximum in 25% as compared to control. Growth was less in 0.1mM chromium as compared to control, 25% effluent and 0.05mM chromium solutions Eco-friendly bio sorption methods are required to eliminate the contaminants which may helpful in the growth and development of crop species. Among the bio sorption methods, bio-sorption by spirulina (blue green algae) is widely accepted method.

Table 4. Effect of tannery effluent on fresh and dry weight of shoot

Effluent Dilution	Fresh and dry weight of shoot (gm)					
	ADT-37		ADT-39		LR-47	
	Fresh wt	Drt wt	Fresh wt	Drt wt	Fresh wt	Dry wt
Control	0.041	0.008	0.040	0.012	0.205	0.025
0.05mM Cr	0.015	0.007	0.032	0.010	0.177	0.021
0.1mM Cr	0.011	0.004	0.020	0.009	-	-
25%	0.030	0.008	0.038	0.011	0.177	0.020
50%	0.018	0.006	0.029	0.08	0.084	0.016
100%	-	-	-	-	-	-

Table 5. Effect of tannery effluent on fresh and dry weight of root

Effluent Dilution	Fresh and dry weight of root (gm)					
	ADT-37		ADT-39		LR-47	
	Fresh wt	Dry wt	Fresh wt	Dry wt	Fresh wt	Dry wt
Control	0.018	0.005	0.015	0.010	0.038	0.012
0.05mM Cr	0.011	0.003	0.010	0.006	0.030	0.010
0.1mM Cr	0.008	0.002	0.008	0.005	-	-
25%	0.015	0.004	0.012	0.005	0.035	0.007
50%	0.009	0.003	0.08	0.003	0.012	0.004
100%	-	-	-	-	-	-

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### References

- [1] Alvarez-Bernal D., Olalde-Portugal V., Frias-Hernandez J. T., Dendooven L. (2006). Effects of tanneries waste water on chemical and biological soil characteristics. *Applied Soil Ecol.* 33, 269–277.
- [2] Arnon D. I. (1949). Copper enzyme in isolated chloroplast polyphenoloxidase in *Betavulgaris L.* *Plant physiol.* 24, 1-15.
- [3] Bradford M. M. (1976). Rapid and Sensitive Method for the quantization of Microgram Quantities of Protein Utilizing the Principle of Protein-Dye Binding. *Anal. biochem.* 72, 248-254.
- [4] Liu D., Jiang W., Gao X. (2003). Effect of Cadmium on Root Growth, Cell Division and Nucleoli in Root Tip Cells of Garlic. *Plant Biol.* 47, 1, 79-83.
- [5] Mc Cready R. M., Guggolz J., Silviera V., Owens M. S. (1950). Determination of starch and amylose in vegetables. *Anal. Chem.* 22, 1156-1158.
- [6] Moore S. and Stein W. H. (1948). Photometric ninhydrin method for use in the chromatography of amino acids. *J. Biol. Chem.*, 176, 367-388.
- [7] Mythili K. and Karthikeyan B. (2011). Bioremediation of tannery effluent and its impact on seed germination (blackgram and sunflower). *Current Bot.* 2, 8, 40-45.
- [8] Nesmann E. R., Lee E. G. H., Metula T. I., Duglas G. R., Muller J. C. (1980). Mutagenicity of constituents identified in pulp and paper mill effluents using the Salmonella/mammalian microsome assay. *Mutat. Res.* 79, 203-212.
- [9] Rahman, K. S. M., Banat, I. M., Rahman, T. J., Thayumanavan, T., Lakshmanaperumalsamy, P. (2002). Bioremediation of gasoline contaminated soil by a bacterial consortium amended with poultry litter, coir pith and rhamnolipid biosurfactant. *Biores. Technol.*, 81, 2532.

- [10] Samantaray S., Rout G. R., Das P. (1999). Studies on Differential Tolerance of Mung Bean Cultivars to Metalliferous Minewastes. *Agribiol. Res.* 52, 3-4, 193-201.
- [11] Sangeetha, R., Kamalahasan, B., Karthi, N. (2012). Use of tannery effluent for irrigation: an evaluative study on the response of antioxidant defences in maize (*Zea mays*). *Int. Food Res J.* 19, 2, 607-610.
- [12] Vajpayee P., Rai U. N., Ali M. B., Tripathi R. D., Yadav V., Sinha S. (2001) Chromium Induced Physiological changes in *Vallisneria spiralis* L. and its role in phytore-Mediation of Tannery Effluent. *Bulletin of Envir. Contam. and Toxicol.* 67, 2, 246-256.
- [13] Van Assche F., and Clijsters H. (1983). Multiple Effects of Heavy Metals on Photosynthesis. *The Hague*, 371-382.