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Corn and Wheat as Bioindicators for Oil Pollution

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A POT experiment was conducted to determine the effects of various concentration of crude oil on the growth of maize (zea mays) followed by wheat (*Triticum valgura* L).

Germination percentages of wheat and corn seeds were significantly reduced in the oil polluted soils. The germination percentage in oil polluted soil at rate of 4.5 % was 40 – 50 % when seeded at 30 days after contamination (T_{30}), while it was only 20-25 % when seeded at time of contamination (T_0).

Seedings shoot length were severely affected by oil contamination greater than 3 %. Plants exposed to 4.5 % oil contaminated soil has grown to only 17± 0.4 cm tall and 26.5±0.6 cm for corn and 6.5 ± 0.3 and 10 ± 0.4 for wheat at T_0 and T_{30}, respectively. This represents only 30-40 % that of control.

Dry matter (g/pot) of corn and wheat was reduced by increasing oil pollution. The reduction was more in plants cultivated at zero time compared to those cultivated after 30 days of contamination.

The concentrations of Mn and Fe in corn and wheat plants grown in soil polluted by 4.5 % oil was 2 – 2.5 times more than that of control. The concentration of Cd, Pb, Cu and Zn was 3 – 3.5 times more than that of control.

Keywords: Crude oil, Contamination, Plant growth, Wheat, Corn.

A significant accumulation of petroleum hydrocarbons in soils and sediments has been found by various researches (Blumer *et al.*, 1977; Gschwend & Hites, 1981; and Jones *et al.*, 1989). Concentration of polynuclear aromatic hydrocarbons (PAH) in soils of Europe and north America varied between 50-500 μg kg^{-1} (Windsor & Hites, 1979 and Edwards, 1983). Wang *et al.* (1989) reported that beyond 3% concentration oil becomes increasingly deterous to soil biota and plant growth. Baker (1970) Pointed out that investigations on the effects of crude oil on plants have concentrated on the direct toxic effects of oil on plants.

Ghanima Malallah *et al.* (1996) used the *Vicia faba* as a bioindicator of oil pollution in Kuwait. Biomass of the plant grown in polluted soil significantly decreased as compared to the plant grown in control soil. Sugar, phenol, protein, free amino acid contents and protein content of the plants grown in polluted soils showed significantly higher levels as compared to plants grown in control soil.

Very little work studied the effect of hydrocarbon contaminated soil on plants. Therefore, the present study is conducted to examine and achieve proper judgement of the effect of hydrocarbon contaminated soil on plants. For this purpose, green house biotests were carried out to study the effect of concentration of crude oil on plantation. Corn and wheat are used as bioindicators in a pot experiment.

Material and Methods

A pot experiment was conducted on a sandy soil having the following characteristics pH 7.2-7.6, electrical conductivity ($EC_{1:2.5}$) 0.3-1.9 dS/m, organic matter 0.1-0.3 %, $CaCO_3$ 34-48 %, cation exchange capacity (CEC) 2 - 6.4 cmol / kg; sand 60 - 86 %; Silt 8 - 14 %, and clay 4 - 14%. Soil properties were determined according to Jackson (1967); Piper (1950) and Tucker (1971). Total content of Cd, Pb, Fe, Mn, Zn and Cu were extracted by digestion in mixture of nitric, sulfuric and perchloric acids (Hesse, 1971).

Crude oil was obtained from the oil well at about ten kilometer from Ras-Sudr Agricultural Experimental Station of the Desert Research Center, South Sinai, Egypt. Some chemical properties of the used crude oil are presented in Table 1. The heavy metals of the oil were extracted according to the method described by Price (1972), and were measured by atomic absorption spectrophotometer (Perken – Elmer model 2083). Total petroleum hydrocarbons (TPH) were extracted according to the method described by Farag *et al.*, (1990) and were determined using GLS apparatus (GLS pyeunicam pa 4550 capillary chromatography).

TABLE 1. Some chemical properties of the studied crude oil.

Heavy metals ($\mu g\ g^{-1}$)						Total petroleum Hydrocarbons (TPH) %
Fe	Mn	Zn	Pb	Cu	Cd	
8960	462	95	37	14.3	6.0	82.82

Green house experiment

A pot experiment was conducted during the season (April, 1996-February, 1997) in the green house at the Desert Research Center, using maize (*Zea mays*) followed by wheat (*Triticum valgure* L.) plant. Unpolluted surface soil (0-15 cm) was contaminated with crude oil by spraying and continuous mixing of the soil with the crude oil to reach the contamination level (1.5, 3 and 4.5 %). An uncontaminated control was set up. The soil was then placed into pots measuring 20 cm inside diameter and 25 cm height. seedling was performed at two different times. Time I (immediately after oil incorporation) and time II (30 days after contamination). Three replications were used for each treatment in a complete randomized design (Fig. 1).

Sixteen maize (*Zea mays*) seeds were sown per each pot on the 20^{th} of April 1996. After 10 days from sowing, plants were thinned to 8 plants per pot. Recommended doses of chemical fertilizers (150 kg/fad) superphosphate, 100 kg/fad ammonium nitrate and 50 kg/fed potassium sulphate), were added for each pot. On the 25^{th} of June 1996 (40 days from sowing) plants were harvested.

The same previously used pots were cultivated on the 20^{th} of November 1996 with 20 seeds of wheat (*Tritcum valgure* L.) per pot. Ten days after sowing plants were thinned to 12 seedlings per pot. All pots were fertilized with the recommended doses NPK. Irrigation performed to keep soil moisture content at field capacity. On the 5^{th} of February 1997 (65 days after sowing) plants were harvested. Dry matter at 70° of corn and wheat plants were determined. Soil samples were taken from each pots (after corn and wheat harvesting), air dried and subjected to chemical analysis.

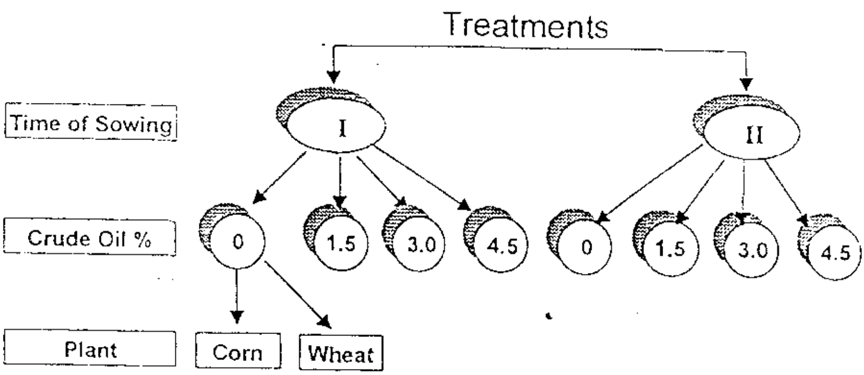

Fig. 1. Sketch diagram of treatments used in the pot experiment.

Results and Discussion

Growth parameters

Germination percentages were determined ten days after sowing. The effect of oil on seed germination has been shown to be inhibitory (Fig. 2). A significant reduction in germination percentage of both wheat and corn was observed in all treatments. The germination of wheat seeds showed rather low percentage than that of corn, which indicate that corn is rather more resistant than wheat regarding hydrocarbons contamination. The germination rate of both seeds was reduced significantly at the greatest application rate of oil (4.5%). Germination percentage was significantly reduced when seeded at the time of contamination (T_0) compared to 30 days after contamination (T_{30}). The germination percentage was 40-50 % at T_{30} while it was only 20-25 % for T_0 in oil polluted soil at the rate of 4.5 %. The inhibitory effect of oil on seed germination was associated with unfavorable soil conditions. Soils contaminated with oil appear waxy and usually hinders water penetration and reduced oxygen contact in the soil. Wet soil becomes water logged, however, upon drying, soils become too hard to allow good germination, (Barolo, 1996).

Contamination of soil with petroleum hydrocarbons has a pronounced effect on plant growth. In this study, growth depression was expressed in terms of shoot length. Significant reductions in shoot length were observed for all treatments compared to the control (Fig. 2). After 40 days for corn and 65 days for wheat plants exposed to 4.5% oil contaminated soil have grown to only 17 ± 0.4 cm and 26.5 ± 0.6 for corn and 6.5 ± 0.3 and 10±0.4 cm for wheat at, T_0 and T_{30}, respectively. This represents only 30-40 % that of control. The effect was more pronounced when plants sown at time of contamination T_0. The reduction in shoot length was only 17% at T_{30}, while it was 30% at T_0 when plants grown in soil treated with only 1.5%.

There is a significant decrease in biomas (dry matter) of plants (mg/pot) grown in the polluted soils as compared to control soil. The dry matter yield was reduced in plants cultivated at T0 compared to those cultivated after 30 days of contamination. Crude oil has a caustic or lethal affect only when it comes into direct contact with the tissues of plants. Crude oil indirect effect on soil is confined to a more or less marked reduction in plant growth and biomas, possibly attributable to its effect on soil microbe population (Barolo, 1996). The rather greater C/N ratio tends to favour carbon in oil contaminated soils. This promotes intensive metabolic activity in all micro-organisms which is not inhibited directly by the oil. This causes severe damage to plants by the mobilization of all nitrogen (Antoniewski and Schaefere, 1972). The degradation rate of hydrocarbons is determined by the complexity of their molecules. Linear compounds up to 30C, may disappear in a month where those up to 37C need at least 200 days (Gudin, 1978). This explains the greatest values of plant parameters studied at T_{30} than at T_0 as the chemical composition of crude oil used has more than 90% of the hydrocarbons with molecules of linear compound less than 30C. With increasing the time between contamination and seedlings, the concentration of phytotoxic volatile compounds decreased.

Heavy metals contents in plants

Highly significant differences in heavy metal concentrations in plants were observed between the three levels of petroleum. The concentrations of Mn and Fe in corn and wheat plants grown in soil polluted by 4.5 % oil was about 2-2.5 times compared to that of control. The concentration of Mn and Fe found to be 122 and 465 $\mu g\ g^{-1}$ in corn and 110 and 450 $\mu g\ g^{-1}$ in wheat, respectively, (Fig. 3). The concentrations of Cd, Pb, Cu and Zn in plants grown in soil treated with oil at the rate of 3% were found to be about 3-3.5 times more than that of control.

The concentrations of the six heavy metals were rather greater in corn compared to those of wheat. The concentrations of all studied metals are greater when plants grown at T_{30} (30 days after contamination), as most of the hydrocarbons disappear and more free metals are available.

Heavy metals concentrations in soil

Heavy metals concentrations in the soil showed a tendency to increase with increasing pollution. The largest concentrations are found in the soils treated with 4.5 % crude oil (Fig. 4). The heavy metals concentrations showed slightly decrease in soils after the second cultivation. The total concentrations of the six heavy metals ranged between 1.0-3.8, 7.1-26.5, 6.5-19.3, 28.3-89.4, 215-460 and 4540 - 6850 $\mu g\ g^{-1}$ for Cd, Pb, Cu, Zn, Mn and Fe, respectively.

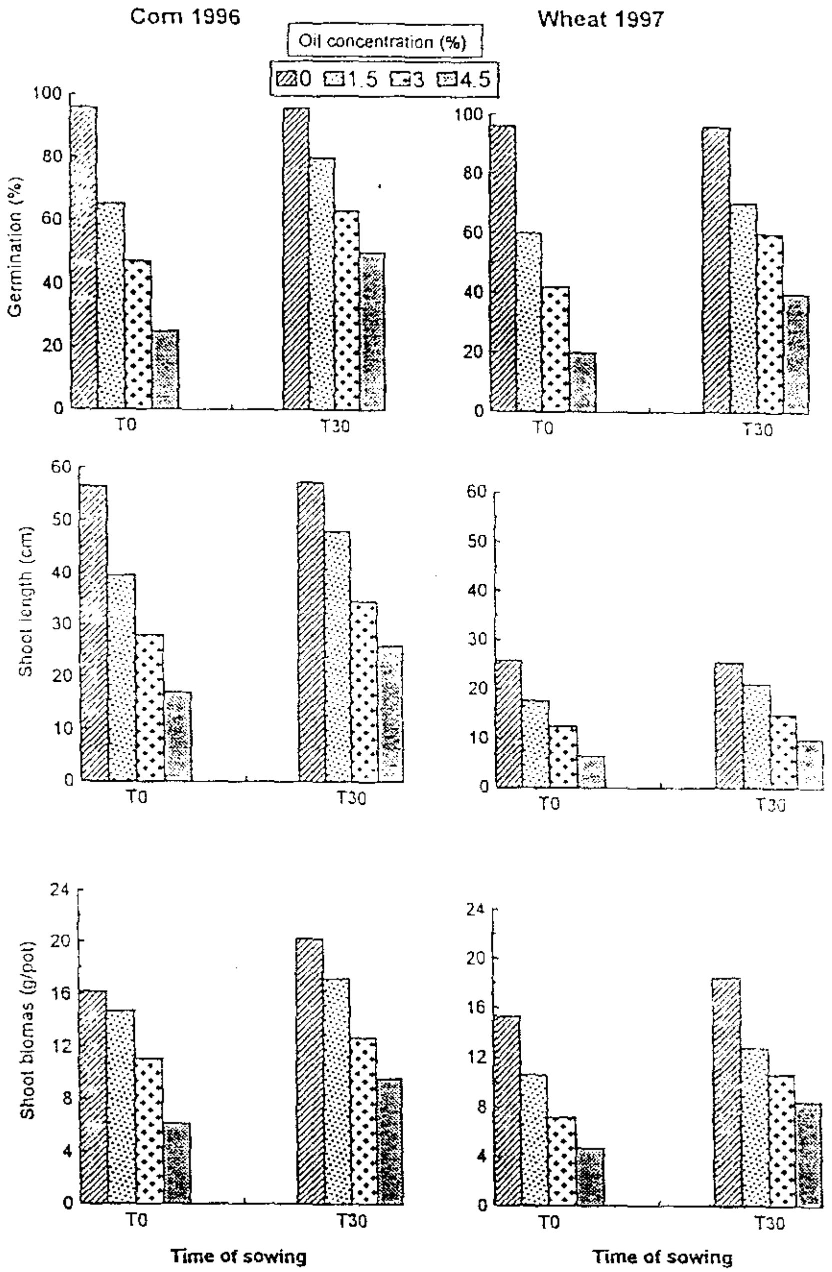

Fig. 2. Growth response of corn and wheat seedlings to crude oil concentration at different times of sowing.

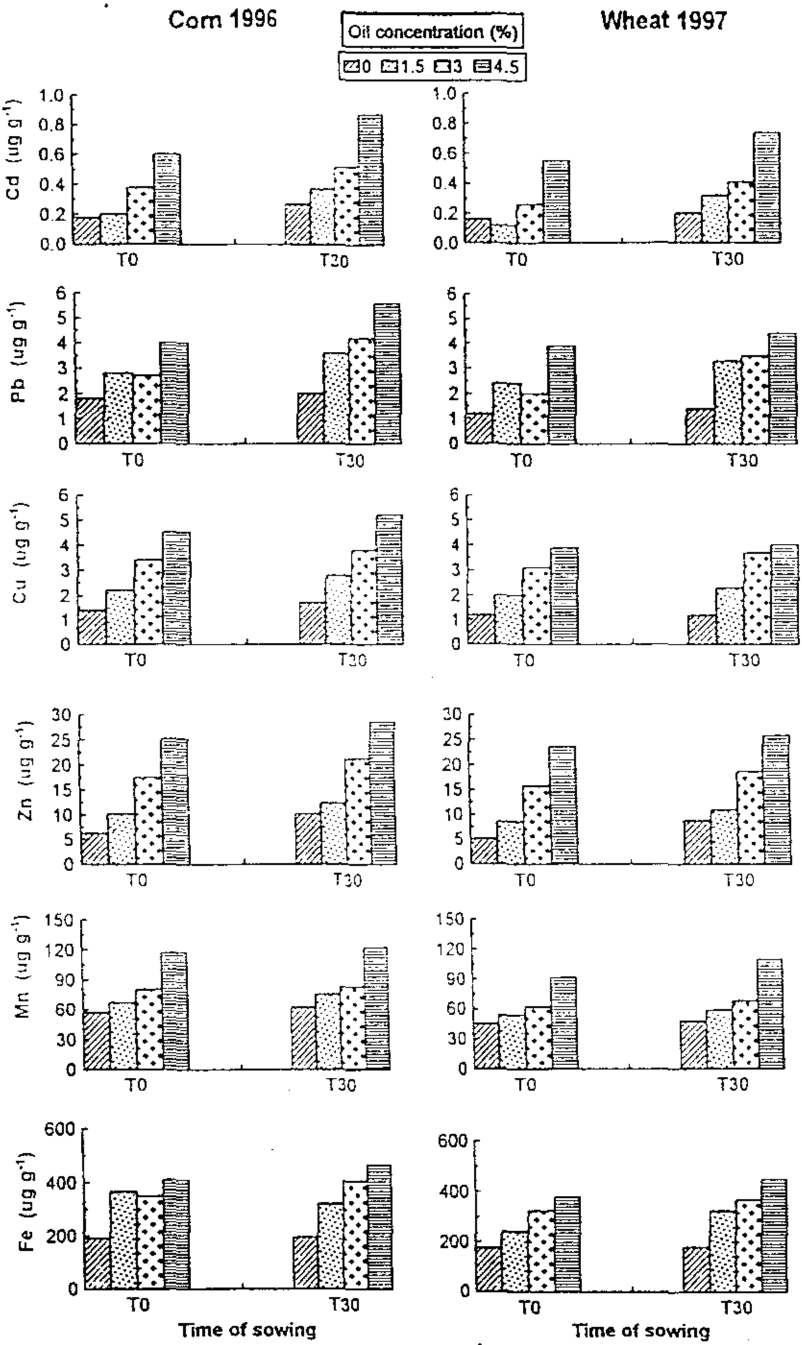

Fig. 3. Concentration of heavy metals (µg g^{-1}) in corn and wheat plants grown in soil polluted by different levels of crude oil at different times of seedling.

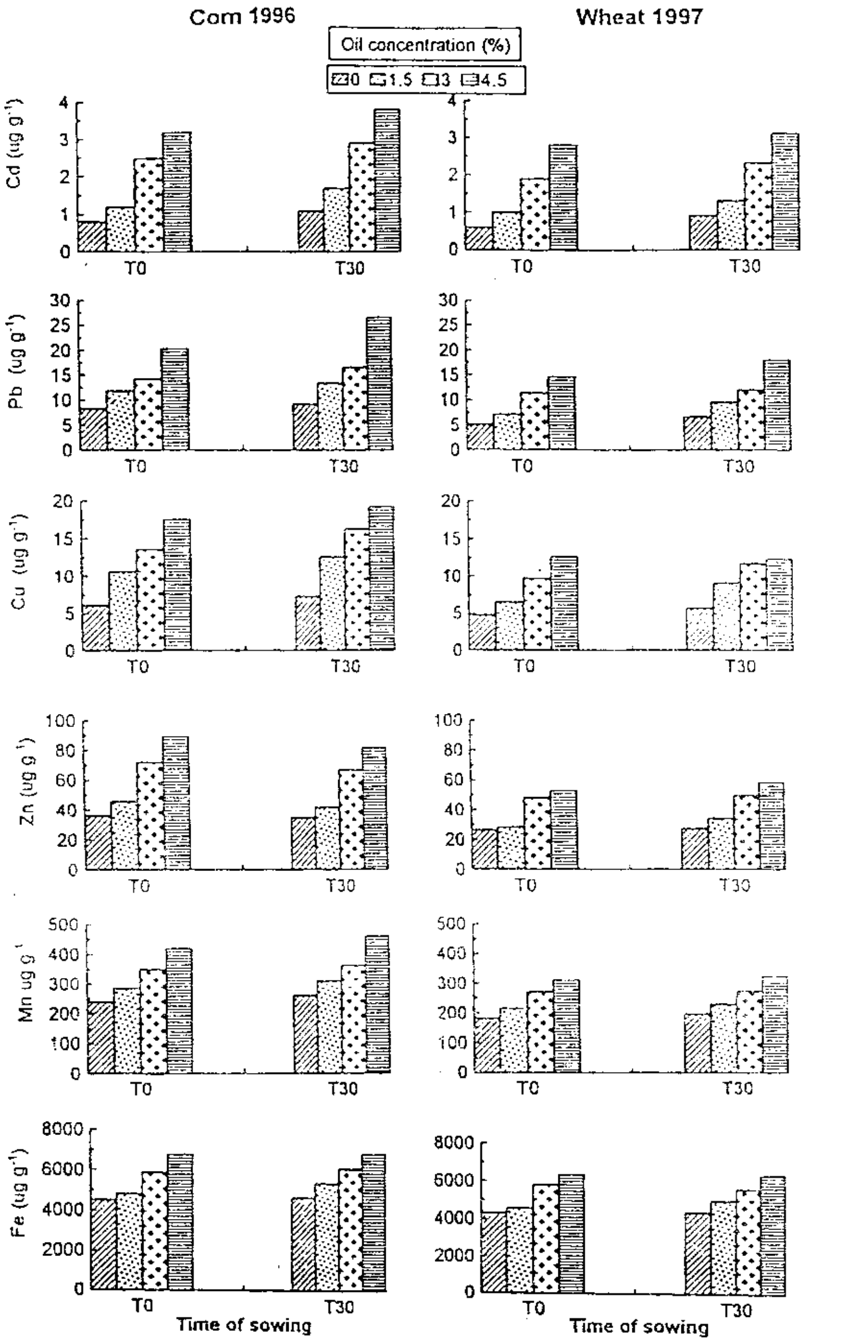

Fig. 4 Total heavy metals ($\mu g\ g^{-1}$) in the soil polluted by different levels of crud oil.

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الذرة والقمح كدليل بيولوجى للتلوث بالبترول

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أجريت تجربة أصص لدراسة تأثير التركيزات المختلفة من خام البترول على نمو نبــات الذرة (*Zea mays*) ونبات القمح (*Triticum valgura*).

ولقد بينت الدراسة أن نسبة الإنبات لكل من الذرة والقمح نقصت بدرجة كبيرة فــى الأراضى الملوثة بالبترول. حيث بينت الدراسة أنه عند تلوث التربة بــالبترول بتركيــز ٤,٥% وصل النقص فى الإنبات إلى ٤٠ – ٥٠ ٪ عند الزراعة بعد ٣٠ يوم من التلوث بالبترول بينما كانت ٢٠ -- ٢٥ ٪ عند الزراعة بعد التلوث مباشرة.

كما أوضحت الدراسة التأثير الشديد للتلوث على أطوال النباتات بزيادة التلــوث عن ٣ ٪ . حيث وصلت أطــوال النباتــات الناميــة فى أراضى ذات تلــوث ٤,٥ ٪ إلـــى ١٧± ٠,٤سم ، ٢٦ ± ٠,٦سم للذرة وكانت ٦,٥ ± ٠,٣سم ، ١٠± ٠,٤سم للقمح عنــد الزراعة بعد التلوث مباشرة وبعد ٣٠ يوم من التلوث على الترتيب. وتمثل هذه الأطــوال ٣٠–٤٠٪ فقط من الأطوال فى حالة الأراضى الغير ملوثة.

وبينت النتائج نقص المادة الجافة لكل من القمح والذرة بزيادة التلوث. وكان الــنقص أكبر فى حالة الزراعة بعد التلوث مباشرة عنه عند الزراعة بعد ٣٠ يوم من التلوث.

وأدى التلوث إلى زيادة قيم Mn, Fe فى نباتات القمح والذرة النامية فى الأراضــى الملوثة بتركيز ٤,٥% وكانت الزيادة ٢ – ٢,٥ مرة أكثر من الأراضى الغيــر ملوثــة. وكانت الزيادة فى عناصر Cd, Pb, Cu, Zn ٣ – ٣,٥ مرة أكثر من الأراضى الغيــر ملوثة.