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HEAVY METAL TOLERANCE IN *AGROSTIS TENUIS* Sibth. BY ADAPTATION ON AERIAL POLLUTION

by

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Abstract: *Samples of Agrostis tenuis were collected from various sites inside and outside the copper refining factory in SW Lancashire, which showed tolerance to the toxic action of Cu. This can be distinguished as follows:*

1) *Plants from areas which are directly affected for a long time by copper discharged from chimneys, showed a high index of tolerance.*

2) *Populations found at greater distance from the source of pollution and which have been affected for a shorter period of time by the toxic effect of copper, showed a decrease in the index of tolerance.*

3) *Populations outside the area of the factory showed an even lower index of tolerance, which was, however, considerably higher than that of the control population.*

From the above it can be concludded that there is undoubtedly air pollution caused by the heavy metal refining industry, which directly or indirectly affects the organisms living in the surrounding area.

INTRODUCTION

The characteristics of metal tolerance in plant populations growing near mines are well known (*Antonovics, Bradshaw* and *Turner* 1971, a review). Many different species are found growing on mine workings and nearly in all cases the species that are found growing on these areas are also found on ordinary soil (*Bradshaw* 1952, *Gadgil* 1969, *Urquhart* 1970). It has been clearly demonstrated that the populations of species growing on the toxic soil possess the ability to continue rooting in conditions which are so toxic that ordinary species cannot produce roots (*Wilkins* 1957, *Jowett* 1958, *McNeilly* 1968).

However, besides these characteristic toxic sites, many other sources of toxic metal release are known nowadays, which considerably affect the organisms living around them. Therefore, pollution of the environment by various toxic metals has been a subject of considerable interest during the last few years. It is known that many heavy metals are highly toxic for plants even in the small concentrations found in polluted air (*Gehardsson* 1966, *Rühling* and *Tyler* 1968, *Goodman* and *Roberts* 1971). Such an aerial pollution can he severe and then it is rather natural to expect changes both in the distribution of plants as well as in the genetic structure of the populations, which were exposed to such unfavourable influence.

A metal refining industry was established about 1900 at Prescot in SW Lancashire: it now continues as part of British Insulated Callenders Cables Ltd. The main constituent of the dust particles discharged from chimneys is copper. Despite, however, the precautions taken so far, the area of the factory is quite polluted. Soil analyses of various sites in and around the factory showed that some of them contained quite high metal concentrations. The copper content in the surface soil of some sites in the copper refinery area even reached 4.000 ppm copper (*Wu* and *Bradshaw* 1972, *Wu* et al. 1975). In these sites the vegetation has almost disappeared.

The populations of the various areas of the factory which were studied were artificially created by sowing of commercial varieties. These are areas which date since the foundation of the factory, while others are only about 10 years old. In all these sites the majority of the populations are those of *Agrostis stolonifera*, while those of *Agrostis tenuis* are rather scarce. Because of the fact, however, that the area around the factory is covered mainly by *Agrostis tenuis*, a comparison between these sites and some characteristic ones from the area of the factory was considered advantageous.

The purpose of the present study is to find the degree to which aerial pollution has affected the tolerance and adaptation of *Agrostis tenuis* in places directly or indirectly affected by copper.

MATERIAL AND METHODS

a) *Sampling*

The plants of *Agrostis tenuis* which were used in this study were collected from three different polluted areas of a copper processing factory. These areas were selected in such a way as to leave quite a distance between them and to be in the direction of the strongest and commonest prevailing wind. Typical samples from the oldest and most polluted area of the factory were the populations of position A. In contrast, the populations of position B were much newer and the soil toxicity was much lower than that of position A. Finally, two other populations were selected from diametrically opposite positions outside the factory area which were characterized as C.

For a comparison between the above populations, samples of *Agrostis tenuis* were collected from an uncontaminated area (Sefton Park and Garway), which were used as control populations. All the collected populations, having been carefully divided into various genotypes, were then reproduced on normal soil in a glass-house for 2-2,5 months before they underwent a test of their tolerance to copper, zinc and lead.

b) *Copper, Zinc and Lead tolerance test*

The method which was used for the measurement of tolerance was limited only to the test of root production, because this is the most characteristic and the quickest one (*Gregory* and *Bradshaw* 1965).

Plants of *Agrostis tenuis*, having grown for 8-10 weeks on normal soil in a glass-house were collected and put for testing in plastic beakers which contained solutions of heavy metals according to Jowett's method (1958, 1964) as it was modified by *Gregory* and *Bradshaw* (1965) and *McNeilly* and *Bradshaw* (1968). The content of the beaker was changed every other day so that the solution concentration remained unaltered. The same genotypes were put, on the one hand, in a beaker with only $Ca(NO_3)_2$ 0,5 gr/litre, and, on the other hand, in a beaker having $Ca(NO_3)_2$ accompanied by the desired concetrations of various metals.

After 10 days the longest root of each tiller was measured and the index of tolerance (I. T.) was calculated according to the following ratio:

$$\text{I. T.} = \frac{\text{Mean length of longest root in solution with metal}}{\text{Mean lenght of longest root in solution without metal}} \times 100.$$

RESULTS

Experiment I. In order to determine the most satisfactory separation of tolerant and non-tolerant populations in vitro, the effect on rooting of five different copper concentrations was used. At each copper concentration the corresponding $Ca(NO_3)_2$ concentration always remained unaltered.

Two tolerant genotypes were used, randomly selected from A and

B populations, and these were compared with the control population (Fig. 1). Using this experiment it is possible to find the difference in

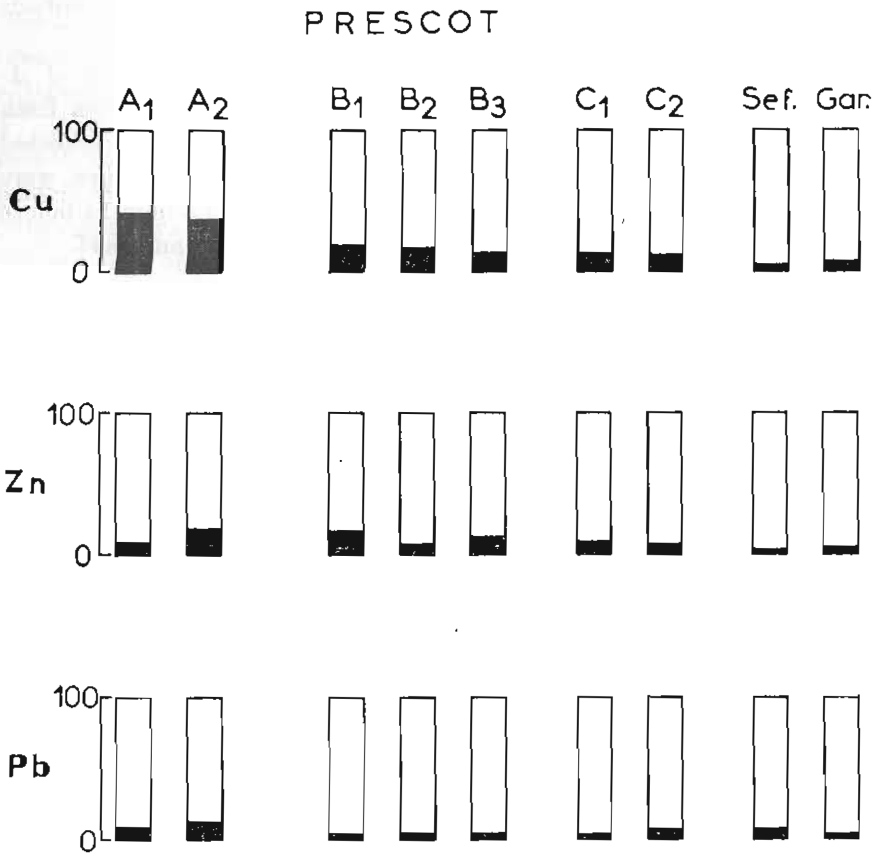


Fig. 1. The root length of populations of Agrostis tenuis in various toxic solutions, expressed as a percentage of their growth.
The tolerant population came from the Prescot refinery area and the two controls from Sefton Park and Garway.

the ability of root production between tolerant and non-tolerant genotypes. Because the control population demonstrated a very small root length when compared to the tolerant populations at the 0.25 ppm copper concentration, this concentration proved the most appropriate one for our measurements.

Experiment II. Nine populations, i. e. two from the more polluted and older area, three from the less polluted and newer area of the factory, two indigenous (outside the factory area) and two pasture populations were tested in three different toxic solutions.

The indices of tolerance of the various popnlations originating from polluted and unpolluted areas are graphically represented in Fig. 1.

The growth of roots in the 0.25 ppm copper solution was much higher for the populations A_1 and A_2 than for the populations B and C, their index of tolerance reaching levels above 35%. However, the populations B and C showed quite a low index of tolerance ranging from almost below half the corresponding index of A. Although the populations at C were indigenous and collected at quite a distance

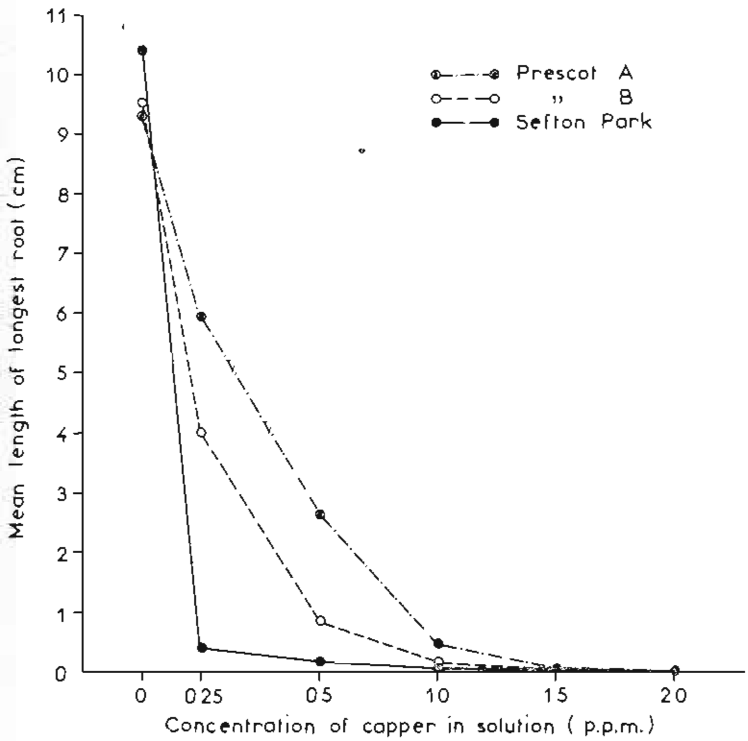


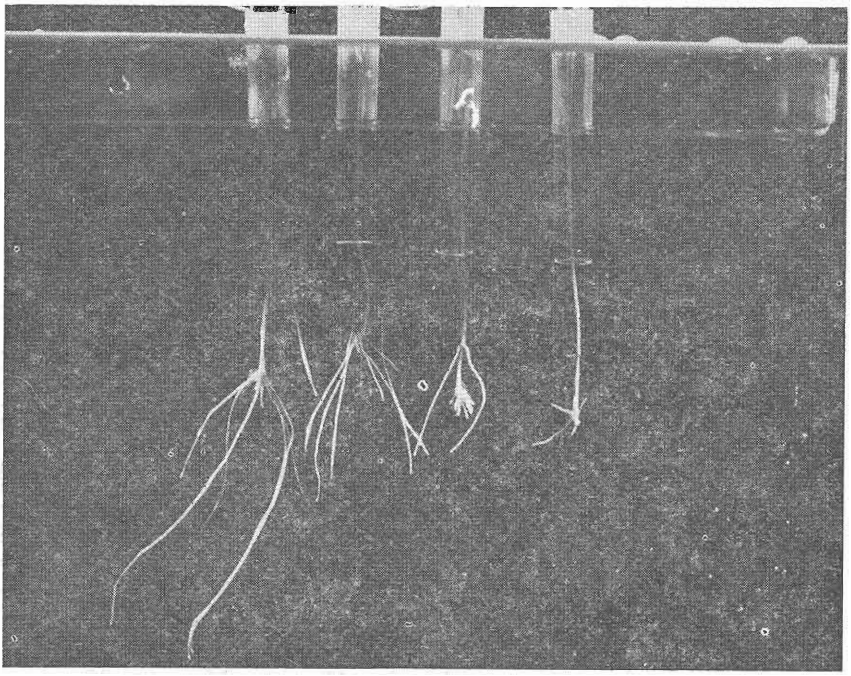
Fig. 2. The effect of copper concentrations on the length of the longest root from two different tolerant genotypes of Agrostis tenuis compared with a non-tolerant genotype.

from the factory, their indices of tolerance were little Iower than those of position B (Fig. 3).

In contrast to Cu, the effect of the toxic action of Zn and Pb solutions demonstrated characteristically low levels of tolerance. However, by comparing the indices of tolerance to Zn and Pb, we find that those of Zn are relatively higher than those of Pb.

DISCUSSION

The ability of some *Agrostis tenuis* populations to survive in areas polluted by various metals is due to certain features which are genetically determined (*Gregory* and *Bradshaw* 1955, *McNeilly* and *Bradshaw* 1968, *Tiku* and *Snaydon* 1971, *Wu* and *Bradshaw* 1972, *Walley* et al. 1974, *Gartside* and *McNeilly* 1974, *Wu* et al. 1975).



Fif. 3. Root growth of Agrostis tenuis genotypes collected from three different polluted areas of the copper factory and control (right), grown in 0.25 ppm copper solution.

The results of the tolerance study of the different sites are graphically illustrated in Fig. 1 and 2, in which it is demonstrated that populations from polluted areas show a characteristically high level of tolerance to Cu, while those from unpolluted areas show a very low one. Also, by comparing the populations A, B, C to the pasture populations, we find a gradual reduction in the index of tolerance, depending rather on the position and the time of the effect of pollution.

Indeed, the populations collected from position A, because of their immediate vicinity to the factory chimneys and the long-term effect of the discharged particles from the chimneys, were chosen so that the ones already existing would be already adapting to this environment. Also the fact that, except for *Agrostis stolonifera* and some *Agrostis tenuis* populations, no other appears, allows us to assume that these populations represent a series of individuals which evolve from the adaptation point of view. During the elapsed period of time all the individuals of non-tolerant genotypes have disappeared, while the tolerant ones tend to occupy the uncovered areas.

In contrast to position A, the area where the populations B were collected shows a low index of tolerance and more uncovered areas. This can be explained if we take into consideration that: 1) This area is chronologically newer, and consequently, insufficient adaptation was attained during this short period of time. 2) The effect of the dust from the chimneys is rather high, though not as high as in position A.

Taking into consideration the above two factors in relation to the fact that in the newer positions there appear more uncovered areas, we can explain the appearance of a low index of tolerance in these positions, accepting that their populations are in the process of adaptation.

In relation to the older areas, the populations from outside the factory would be expected to behave like the pasture populations. However, the indices of tolerance differ considerably from the corresponding pasture populations and very little from the populations of position B. This unexpected behaviour of the populations of C around the factory is most probably due to the following factors:

a) To the toxic effect of the Cu particles which are discharged from the factory chimneys.

b) To the gene flow and

c) To a combination of the above.

Because of the above prevailing conditions it is possible to find tolerant genotypes in positions quite a distance from the factory.

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ΠΕΡΙΛΗΨΙΣ

ΑΝΘΕΚΤΙΚΟΤΗΣ ΤΟΥ AGROSTIS TENUIS Sibth. ΕΝΑΝΤΙ ΒΑΡΕΩΝ ΜΕΤΑΛΛΩΝ ΔΙΑ ΠΡΟΣΑΡΜΟΓΗΣ ΕΙΣ ΤΗΝ ΡΥΠΑΝΣΙΝ ΤΟΥ ΑΕΡΟΣ

Ὑπὸ

Σ. Σ. ΚΑΡΑΤΑΓΛΗ

Δείγματα *Agrostis tenuis* συνελέγησαν ἐκ διαφόρων θέσεων ἐντὸς καὶ ἐκτὸς τοῦ Ἐργοστασίου ἐπεξεργασίας χαλκοῦ τοῦ SW Lancashire, τὰ ὁποῖα ἐνεφάνισαν ἀνθεκτικότητα ἔναντι τῆς τοξικῆς δράσεως τοῦ Cu. Δύναται δὲ αὕτη νὰ διακριθῇ ὡς ἀκολούθως:

1) Φυτὰ περιοχῶν ὑφιστάμενα τὴν ἄμεσον καὶ ἐπὶ μακρὸν χρονικὸν διάστημα ἐπίδρασιν τοῦ ἀποβαλλομένου ἐκ τῶν καπνοδόχων χαλκοῦ, ἐνεφάνισαν ὑψηλὸν δείκτην ἀνθεκτικότητος.

2) Πληθυσμοὶ εὑρισκόμενοι εἰς μεγαλυτέραν ἀπόστασιν ἐκ τῆς ἑστίας μολύνσεως καὶ οἱ ὁποῖοι ὑπέστησαν ἐπὶ μικρότερον σχετικῶς χρόνον τὴν τοξικὴν ἐπίδρασιν τοῦ χαλκοῦ, παρουσίασαν ἐλάττωσιν τοῦ δείκτου ἀνθεκτικότητος.

3) Πληθυσμοὶ ἐκτὸς τοῦ Ἐργοστασίου ἔδειξαν ἀκόμη μικρότερον δείκτην ἀνθεκτικότητος, ὁ ὁποῖος ὅμως ἦτο σημαντικὰ ὑψηλότερος τοῦ Control population.

Ἐκ τῶν ἐκτεθέντων συνάγεται ὅτι ἀναμφιβόλως προκαλεῖται ρύπανσις τῆς ἀτμοσφαίρας προερχομένη ἀπὸ τὴν βιομηχανίαν ἐπεξεργασίας βαρέων μετάλλων, ἡ ὁποία ἐπηρεάζει ἄμεσα ἢ ἔμμεσα τοὺς πέριξ διαβιοῦντας ὀργανισμούς.