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There are a number of obvious errors in thes Itesis. This is because it was written after the candidate left for U.S. ct and no draft copy was submitted for comment. le. V. Fife

28,4.58

A STUDY OF THE CAUSE OF GERMINATION INJURY FOLLOWING CONTACT PLACEMENT OF DRIED BLOOD FERTILIZER WITH SEEDS

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> By Nancy L. Baigent January 1958

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CHAPTER I

INTRODUCTION AND REVIEW OF LITERATURE

The use of organic fertilizers has suffered a decline since the initiation of large scale production of soluble inorganic salts. Nevertheless, a demand for natural organic forms of nitrogen persists, especially in regions of high rainfall and sandy soils, and in connection with the production of crops of high acre value.

In recent years knowledge of the principles and practice of fertilizer application also has expanded rapidly. Localized or contact placement of fertilizer has been found to favour rapid early growth, and to lessen "fixation" of fertilizer nutrients by the soil. Difficulties in the forms of impaired germination and damage to young seedlings have arisen from contact placement of organic fertilizers. Many mechanisms have been put forth to explain the basis of fertilizer toxicity;viz. plasmolysis of root tissues by the high solute concentrations, production of excessive local acidity or alkalinity from fertilizer materials, direct toxic effects on the young plant by free ammonia or cyanide formed by chemical or microbial breakdown of fertilizer materials.

Accordingly, it was decided to evaluate the hypothesis that germination injury to seeds results from the production of injurious quantities of free ammonia during mineralization of dried blood fertilizer placed in contact with the seeds. Since nitrogen is taken up by the plant chiefly as nitrate, or sometimes as ammonia, complex forms of nitrogen present in organic fertilizers must undergo mineralization in the soil before becoming available to the plant. In this process, microorganisms convert protein nitrogen into ammonium salts, and thence to nitrites and nitrates. Excessive water, or the absence of oxygen from any other cause, tend to favour an anaerobic microflora, and may result in stopping the nitrification process, and in the reduction of such nitrates as were already formed under more favourable conditions. That ammonification and nitrification are not necessarily correlated was once more confirmed by Fraps and Sterges (1947). Since ammonification was still significant in all extreme cases, the result was an accumulation of ammonia under the influence of such factors as too high and too low pH, and too high and too low temperature.

Many investigations clearly point to a volatilization of ammonia, not directly from organic substances such as dried blood, but from free ammonia after ammonification. (Screenivasan and Subrahmanyan (1935), Willis and Sturgis (1945), Pochen and Tchan (1947), Pochen et al (1947), to name a few such studies.) Accordingly, a review of the relevant literature on injury to germination through the presence of toxic concentrations of free ammonia will now be presented.

Russell and Petherbridge (1913) found that sand cultures containing 10 parts per millon of nitrogen as free ammonia, retarded the germination of turnip seed, whereas with 100 parts per millon of nitrogen no seed germinated. Since the

sand cultures contained 16.7% of moisture, it appeared that 0.006% of nitrogen as free ammonia was injurious and that 0.06% was fatal.

The work of Willis and Piland (1931) supports the idea that free ammonia in high concentrations acts as an inhibitor of seed germination and plant growth. The fertilizer used was a mixture of C.P. diammonium phosphate, potassium nitrate, and potassium chloride. The free ammonia formed by the hydrolysis of the diammonium phosphate was apparently the most toxic component of the fertilizer mixture for the young cotton seedlings. No injury was observed from the use of ammonium as the sulphate, chloride or nitrate. Nor did the alkalinity of the diammonium phosphate appear to contribute to the injurious effect. Monoammonium phosphate.

The hypotheses were investigated that hydroxyl ions or free ammonia were the toxic factors. This was accomplished through the use of calcium carbonate as a treatment supplemental to applications of diammonium phosphate, and through calcium carbonate and calcium sulphate as supplementary treatments to ammonium hydroxide. There was similarity in the degree of injury from ammonium hydroxide and from diammonium phosphate, although this did not prove that the causes were quantitatively alike. However, the effectiveness of calcium sulphate as a corrective of ammonium hydroxide toxicity gave strong support to that conclusion. The failure of calcium carbonate to correct

the injury from the diammonium phosphate was not thought to controvert the fundamental property of calcium in this respect, because of the possibility, which was suggested, that the calcium carbonate might increase the concentration of free ammonia without correspondingly increasing the concentration of calcium available for antagonism.

The slight but probably significant decrease in the extent of injury from ammonium hydroxide when supplemented by calcium carbonate, eliminated the possibility that a reduction in the concentration of hydroxyl ions was the sole cause of the previously noted remedial effect of the calcium salts. This result with calcium carbonate and ammonium hydroxide strongly indicated that the calcium has a direct physiological effect independent of any chemical reaction within the medium. There was the possibility, however, that the greater efficiency of the sulphate was not due to a more intensive antagonism, but to an additive effect of a chemical reaction, probably the formation of the non-toxic ammonium sulphate.

Willis and Piland (1931) concluded that, in practice, fertilization with diammonium phosphate, or other materials productive of free ammonia, might not be injurious on highly absorptive soils, or when the fertilizer was applied long enough in advance of planting to provide for complete absorption. Under other conditions the use of gypsum as a supplement might constitute an effective means of control. Ground limestone would probably not serve the same purpose. They considered that this type of fertilizer injury constitutes a problem only with germinating

and seedling plants, and it is probable that the ultimate effect on the crop would depend on the nature of the rooting systems of the plants fertilized. Taprooted plants would naturally be most subject to damage, but this might be corrected by the later development of the lateral roots.

That injury to germinating seeds by free ammonia occurs was also demonstrated by Barton (1940). She investigated the effects on various organisms of different concentrations of gases applied for several periods of time. Germination of watersoaked seeds of radish exposed for as long as 240 minutes to 1000 parts per million of ammonia gas was not only delayed, but reduced. Extension to 960 minutes killed off all the seeds. In no case did exposure of dry seeds of radish to ammonia gas cause reduction in germination percentage. Rye seeds were more sensitive than those of radish. Exposure of soaked rye seeds to 1000 parts per million of ammonia gas for 240 minutes resulted in 100% kill, while those exposed to 250 parts per million for 960 minutes had a germination capacity of only 48%.

Duisberg and Buehrer (1954) studied the extent of the inhibitory effect on seed germination of applications of ammonia by irrigation or by injection. Ammonia applied by surface irrigation in an amount to give a concentration of 230 parts per million of nitrogen on the dry soil basis, prevented the germination of barley, whereas when applied by injection, a concentration of 270 parts per million of ammonium nitrogen not only permitted germination, but resulted in more vigorous plants than in the controls.

To determine the limit of ammonia concentration at which germination is entirely prevented, pots of soil uniformly injected with ammonia at a series of concentrations were planted to barley. Germination was complete at all ammonia concentrations up to 450 parts per million during the first 10 days after planting. Increase in concentration of ammonium nitrogen tended to delay germination.

Independent germination tests were made to determine whether inhibition is due to the initially high hydroxyl concentration, or to ammonium ion concentration. This was done by surface-irrigating the soil with calcium hydroxide, ammonium chloride, and ammonium sulphate solutions prior to planting. Germination was not inhibited by hydroxyl ions at an initially high concentration as long as the ammonium ion was absent. Ammonium nitrogen at concentrations above 450 parts per million prevented germination completely. At lower concentrations of ammonium ion germination occurred, but the plants were feeble and stunted. The plants growing in the ammonium sulphate-treated soil were, in general, more vigorous than those growing in the ammonium chloride-treated soil, which pointed to a possible toxic effect of the chloride ion.

The literature on the subject of causes of germination injury through contact placement of dried blood with the seed, is limited. Sayre and Clarke (1935) found that many organic fertilizers are injurious to seeds and roots of plants, when first applied to the soil. Their experimental results with peas, beans, and corn in the glasshouse were confirmed by field trials.

They found that the lower rate of solution of organic fertilizers, as compared with inorganic fertilizers, indicated that the cause of injury to roots resided in some factor or factors other than excessive concentration of soluble salts resulting in plasmolysis of root tissue. They considered that injury from organic fertilizers was caused by various soluble organic substances such as amino-acids and soluble peptides. In addition it was found that organic fertilizers greatly stimulated mould growth, with the logical conclusion that they might also stimulate more rapid growth of parasitic organisms that attack roots and seeds in the soil, and thus indirectly cause injury.

The period of injurious concentration was found to vary with the rate of application, the soil moisture content, and the soil texture. In their experiment with dried blood, it required from 9-14 days in the soil before this fertilizer was no longer toxic to roots. The detrimental effects of dried blood were definitely restricted to an area in close proximity to the fertilizer band, as shown by the fact that seeds sown $\frac{1}{2}$ ⁿ to the side of the fertilizer band germinated satisfactorily.

Sherwin (1923), working with cotton and corn seeds, found that organic fertilizer reduced the germination count by causing the death of the seedling before its appearance at the soil surface. He noted absence of roothdirs and considerable root decay. The growth of fungi was also stimulated, especially those fungi which are injurious to root systems.

The mineralization studies and germination injury experiments reviewed in the foregoing section suggest that the most likely source of germination injury through contact with dried blood is the production of free ammonia during the mineralization process. Realizing the practical difficulties of demonstrating directly the role of free ammonia production in a system in which water is present, it was decided to approach an answer by the indirect procedure of showing (1) the production of ammonium-nitrogen in quantities sufficient to constitute a source of germination injury and (2) both qualitative and quantitative similarity in germination injury caused by free ammonia from inorganic sources, and that resulting from application of the dried blood fertilizer.