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Phytotoxicity threshold levels of microwave radiation for *Trifolium* and *Medicago* seeds

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Summary

An investigation was conducted on the effects of microwave radiation treatment on *Trifolium* and *Medicago* seeds for the reduction of hard seeds. While the investigation primarily confirmed this action, it was observed that a significant energy threshold was reached, beyond which toxicity rapidly increases. The threshold remained reasonably constant irrespective of the cultivar. The paper reports on this aspect of the investigation.

Résumé

Seuils de phytotoxicité des micro-ondes pour les semences de Trifolium et de Medicago

Une étude a été réalisée des effets d'un traitement par micro-ondes sur *Trifolium* et *Medicago* en vue de la réduction du nombre de graines dures. Cette étude, tout en confirmant en premier lieu cette action, a montré qu'au delà d'un seuil d'énergie déterminé, la toxicité augmentait rapidement. Le seuil reste raisonnablement constant quel que soit le cultivar. Le présent article se rapporte à cet aspect de l'étude.

Zusammenfassung

Die Höhe der Phytotoxiditätsschwelle bei der Bestrahlung mit Mikrowellen für Samen von Trifolium und Medicago

Es wurde eine Untersuchung über die Auswirkungen einer Behandlung mit Mikrowellenbestrahlung an Samen von *Trifolium* und *Medicago* zur Verringerung hartschaliger Samen durchgeführt. Während die Untersuchung in erster Linie diesen Vorgang bestätigte, beobachtete man andererseits, daß eine signifikante Energieschwelle erreicht wurde, jenseits deren die Phytotoxidität sehr rasch zunimmt. Diese Schwelle blieb, unabhängig von der Sorte, ziemlich konstant. Die Arbeit berichtet über diesen Gesichtspunkt der Untersuchung.

Introduction

Materials constituting a dielectric rather than an electrical conductor are rapidly heated when exposed to radio frequency electrical fields. The dielectric heating effect is widely used in the plastics industry for the preheating of moulding powders

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and the welding of plastic sheets. This is usually carried out at a frequency of between 1 and 100 mHz. The development of efficient microwave sources extends this frequency range into the gHz region and microwave ovens for heating and cooking are readily available.

One feature of dielectric heating is the very rapid temperature rise experienced with high dielectric loss materials. Unlike infra-red or radiant heating sources, heat conduction is not primarily involved as energy is absorbed evenly throughout the body if the dielectric has uniform properties. Conversely, areas of high dielectric loss will preferentially heat while other areas remain at a lower temperature.

The use of electromagnetic radiation for the beneficial treatment of seeds has been known for several decades. The work of Riccioni (1942) led to the construction of an industrial installation for seed treatment. More recently, Nelson and Wolf (1964) and Nelson, Nutile and Stetson (1970) have extensively studied the effects on seeds at increasingly higher frequencies. Their reports show various beneficial results including the breaking of dormancy, earlier emergence and the reduction of hard seeds.

There are reasons to believe that these effects are not entirely due to the temperature rise in the seed. Work in Russia by Khvedelidze, Dumbadze, Lamsadze and Datebashvili (1968) shows that a magnetic field influences the germination of seeds. While a constant field was used in their experiments, they suggest that a variable or polarity reversing field would have a similar effect. These conditions would be found in electromagnetic radiation treatment.

Many previous experiments on seeds have been conducted at frequencies well below the microwave region. The amount of power absorbed in the dielectric is directly proportional to the loss factor and the frequency. Small differences in dielectric loss are therefore emphasised with higher treatment frequencies and selective heating will occur within the seed. Dielectric loss is high in areas of large moisture content and energy will be preferentially absorbed in these areas. Jolly and Tate (1971) exploited this difference in their experiments on the killing of chalcid larvae in Douglas fir (*Pseudotsuga menziesii* (Mirb.) Franco) tree seed. Since the grub is largely water the dielectric loss tangent is nearly twice the value for seed flour and a greater proportion of power will be absorbed. They also recorded an increase in germination not entirely due to the disinfestation action.

One advantage claimed for radio frequency treatment is the reduction in the inhibiting effect of a hard seed coat (Nelson and Wolf, 1964). During an investigation of this aspect of the process using microwave radiation, an apparent energy threshold was observed, beyond which toxicity rapidly increases. While research on the primary problem of the reduction of hard seed continues, it is of interest to record the observations made on the toxicity effect.

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Materials and methods

The study was restricted in these early stages to four cultivars of small seeded legumes, the choice of seed being dictated by the prevalence in three species of a high proportion of impermeable seed coats.

Irradiation was carried out using a 1.2 kW domestic type microwave oven operating at a frequency of 2450 mHz. The seeds were contained in a glass petri dish positioned centrally in the microwave cavity. Due to technical problems in accurate power measurement no attempt was made at this stage to determine the amount of energy received by each batch. However, using a dummy water load of 100 cm³ volume and calorimetric measurements, the power setting employed in all the experiments corresponded to 690 W. Sample quantities of seeds were treated at each time from storage normalised bulk supplies and two 2 g batches were treated at each time setting with a constant power level input. After treatment, the seeds were tested for germination using the standard growing procedure established by the International Seed Testing Association (1966).

Two samples of 100 seeds were counted from each 2 g batch, giving four evaluations for individual time settings. The germination test period was 10 days and the results were recorded in four categories.

- (a) Vigorous sprouting seed defined as germinable.
- (b) Completely unaltered seed, as hard seed.
- (c) Imbibing and swelling but no emergence, as fresh seed.
- (d) Remainder as dead seed.

A control batch of 4 \times 100 seeds was also grown.

Results

The results are shown graphically, the time plot corresponding to treatment times of 5, 10, 15, 20, 30 and 45 s. The yield of germinable and hard seeds in each batch of the same cultivar showed an unusual consistency of result, the standard error never exceeding six and being generally of the order of two. The yield of hard seed is also plotted. This tends to indicate that for treatment times up to the point of a rapid increase in mortality the gain in germination is obtained from seeds originally possessing a hard coat. The number of fresh seeds in each batch was not more than 2% and showed no significant change after treatment.

In three, white clover (*Trifolium repens* L.) 'Kersey', white clover 'Kent' and red clover (*Trifolium pratense* L.) 'Hungaropoly' there was a marked increase in germination with a corresponding decrease in hard seed content for exposure times of between 5 and 15 s. The lucerne (*Medicago sativa* L.) cultivar 'Eynsford' showed only a slight increase in germinability with no effect on hard seed yield. In all species a rapid decrease in germination occurred at 20 s exposure, while a treatment time of more than 30 s produced almost complete mortality.



White clover 'Kersey'. Percentage germinable and hard coat seed in terms of treatment time.



White clover 'Kent' approved. Percentage germinable and hard coat seed in terms of treatment time. 674

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Red clover 'Hungaropoly'. Percentage germinable and hard coat seed in terms of treatment time.



Lucerne 'Eynsford'. Percentage germinable and hard coat seed in terms of treatment time.

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Discussion

The selective toxicity of UHF electromagnetic radiation has previously been reported without mention of a catastrophic mortality. Attempts have been made to propose the mechanism involved and this has included a study of the seed parameters and their influence (Davis, Weyland and Merkle, 1973). High frequency radiation is known to produce both thermal, (van Overbeek, Brantley and Potapenko, 1939) and nonthermal effects (Heller and Teixeira-Pinto, 1959; Teixeira-Pinto, Nejelski, Cutler and Heller, 1960) in biological material. The thermal action is primarily a function of dielectric loss and depends mainly on the amount of water present in discrete areas of the seed structure. With a stabilised seed batch a linear function would be expected with an absence of any sharply defined mortality threshold. The very short time interval during which the seed is exposed to high temperature is incompatible with the observed threshold. The rapid increase in mortality for all species at the same exposure time suggests that other biological factors are involved, such as molecular breakdown of the enzyme structure by resonance. As the resonance effect can be catastrophic a sharp peak of mortality would result. Further work is required to investigate this possibility, one approach being to vary the frequency and thus alter the lattice dimensions for which stimulated resonance would occur.

Acknowledgments

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Dr. Zory R. Glaser National Institute for Occupational Safety and Health Room 8A-30 5600 Fishers Lane Rockville, Md. 20857 U. S. A.

14th December 1978

Dear Dr. Glaser:

Through the Bioelectromagnetics Society Newsletter I note that you are asking for details of papers on Microwave effects.

I enclose a copy of my recent paper covering effects on seeds for your files.

Yours sincerely,

A. E. brouf

A.E. Crawford, Director.

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