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Short communication

Evidence for an osmotic mechanism in the control of powdery mildew disease of wheat by foliar-applied potassium chloride

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Abstract

The mechanism by which foliar application of potassium chloride solution reduces symptoms of powdery mildew disease (*Erysiphe graminis* f.sp. *tritici*) of wheat was investigated. The hypothesis that potassium chloride reduces mildew by an osmotic effect on spore germination was tested in three glasshouse experiments. Either potassium chloride solution or the osmoticum polyethylene glycol 200 was sprayed on wheat at the three- or four-leaf stage. The plants were inoculated with spores and spore germination and leaf area affected by mildew were assessed. Leaf water potential was determined as a measure of the osmotic effect of the treatments. Spore germination and leaf area affected by mildew were related to leaf water potential using regression analysis in groups on the data averaged over the three experiments. Both spore germination on the leaf and leaf area affected by mildew were reduced as leaf water potential decreased. There was no difference between potassium chloride or polyethylene glycol in the relationship between spore germination and leaf water potential, but polyethylene glycol was slightly less effective at reducing mildew symptoms at any given leaf water potential. The results are compatible with the hypothesis that potassium chloride reduces symptoms of powdery mildew by an osmotic effect on spore germination.

There are occasional reports in the literature of reductions of leaf diseases when KCl or NH_4Cl have been applied to crops in liquid form, with the intended target the soil, as fertilizers for supplying the nutrients potassium or nitrogen (Fixen, 1993). The possible use of KCl solution as a spray directly to the foliage for reducing disease was therefore investigated. KCl was chosen because it is the main form of potassium fertilizer used on crops throughout the world (Barber et al., 1988). It is primarily soil-applied, but it can be sprayed as a solution in water on the foliage for more rapid uptake (Eibner, 1988), and is used in some commercial foliar fertilizers. Field experiments have shown that symptoms of glume blotch on wheat (*Septoria nodorum*) (Kettlewell et al., 1990), brown rust on barley (*Puccinia hordei*) (Kettlewell et al., 1992), leaf blotch on wheat (*Septoria tritici*) and powdery mildew on wheat (*Erysiphe graminis* f.sp. *tritici*) (Cook et al., 1993) can all be reduced by foliar sprays of KCl.

The mechanism of the effect of KCl on *E. graminis* both *in vitro* and *in vivo* was studied. Plasmolysis was observed when spores were suspended in KCl solution (J.W. Cook, unpublished). Therefore, it was hypothesised that KCl controls *E. graminis* by a reduction in spore germination through an osmotic mechanism. Three experiments were carried out *in vivo* to test this hypothesis. KCl was compared for effects on spore germination and disease symptoms with a control osmoticum known to be relatively inert to plants, polyethylene glycol (PEG) 200 (Lawlor, 1970). Leaf water potential was used as a measure of the osmotic effect of the applied solutions.

Unvernalised plants (cv. Apollo) were grown singly in pots in a glasshouse and arranged in randomised block designs on a well-watered gravel bed. The glasshouse was maintained at a minimum day temperature of 15 °C and minimum night temperature of 5 °C. Plants were sprayed in the late morning at either the three-leaf stage (Experiment 1) or the four-leaf stage (Experiments 2 and 3) with either distilled water, one of five KCl (AnalaR grade, Merck, Poole, UK) solutions (0.27, 0.54, 0.80, 1.07, 1.34 M) or one of five PEG 200 (Merck) solutions (0.54, 1.08, 1.61, 2.14, 2.68 M). Plants were moved from the glasshouse bench for spraying into an enclosed pot sprayer consisting of a spray head fitted with medium flat fan nozzles (015-F80, Lurmark, Cambridge, UK) travelling on a 1.85 m gantry under pneumatic power. Sprays were applied at 0.3 MPa pressure and plants were returned to the glasshouse bench immediately after spraying; 3 h after spraying, when spray droplets had dried, wheat plants affected by powdery mildew were shaken over the experimental plants to inoculate them with spores of *E. graminis*.

Spore germination was assessed in Experiments 1 and 3 on, respectively, 10 and 9 replicate plants for each treatment. The uppermost fully expanded leaf was detached by cutting at the ligule either 18 (Experiment 3) or 24 h (Experiment 1) after inoculation and cleared with a 3 : 1 mixture of ethanol and acetic acid, followed by rehydration in an ethanol series and staining with trypan blue. All spores on the adaxial surface were counted and classified as germinated if a germ tube was present. Leaf water potential was measured in Experiments 2 and 3 on, respectively, 10 and 9 replicate plants for each treatment. The uppermost fully expanded leaf was severed immediately above the ligule with a scalpel 24 h after spraying when plants were fully turgid. Each leaf was sealed in a pressure bomb (M.J. Cantwell, University of Reading, UK), and the pressure around the leaf raised progressively with nitrogen gas. The leaf water potential was recorded as the pressure at which the first appearance of a droplet of sap was observed. The area affected by mildew was assessed 14 d after inoculation on 25 (Experiment 1), 10 (Experiment 2) and 9 (Experiment 3) replicate plants for each treatment. The adaxial surface of the uppermost fully expanded leaf at the time of spraying was assessed subjectively with the aid of standard area diagrams (Anon, 1976).

Preliminary analysis of the effects of KCl and PEG was by analysis of variance separately for each experiment. Both KCl and PEG significantly reduced spore germination, leaf area affected by mildew and leaf water potential in all experiments in which they were measured (data not presented). Thus the effects of KCl and PEG were repeatable. The data from the three experiments were averaged for further analysis to minimise variability. This was valid since the concentrations of KCl and PEG used were identical, other aspects of the treatments and the measurements were broadly comparable and the overall effects were consistent for the three experiments. Mean values for spore germination, leaf water potential and leaf area affected by mildew were calculated for each treatment by weighting according to the number of replicates in each experiment.

In order to test the hypothesis that the effect of KCl on spores and disease was by an osmotic effect on spores, it was thought to be more pertinent to analyse the data in relation to the osmotic effect of the applied solutions rather than in relation to concentration. The osmotic effect of a given concentration sprayed on the leaf will vary between plants and experiments depending on many factors, including environment and leaf surface characteristics, and the leaf water potential provides the most appropriate measure of the osmotic effect of the applied solutions. The treatment means were therefore analysed further by linear regression of spore germination and leaf area affected by mildew against leaf water potential. Linear regression in groups was used to test the overall effect of KCl compared with PEG. Spore germination and leaf area affected by mildew were the dependent variables, leaf water potential the independent variable and KCl or PEG were the groups to be compared. The data from plants treated with water were excluded from the analysis (but presented in the figures) because data from this single treatment cannot logically be assigned to either the KCl or the PEG group in the analysis.

The regression in groups showed that spore germination on the leaf was significantly ($P < 0.001$) reduced as the leaf water potential decreased, with no significant ($P = 0.170$) difference between the two chemicals. Therefore, these data are best represented by one single regression line (Figure 1(a)). Leaf water potential accounted for 91% of the variation in spore germination. The leaf area affected by symptoms of mildew was also significantly ($P < 0.001$) reduced with decreasing leaf water potential. The regression in groups analysis revealed that, in contrast to spore germination, KCl gave a significantly ($P = 0.006$) greater reduction in mildew than PEG at any given leaf water potential. The analysis also showed that there was no evidence of different slopes of the regression lines ($P = 0.165$), and therefore the data are best represented by two parallel lines (Figure 1(b)). Leaf water potential together with the difference between KCl and PEG accounted for 92% of the variation in leaf area affected by mildew.

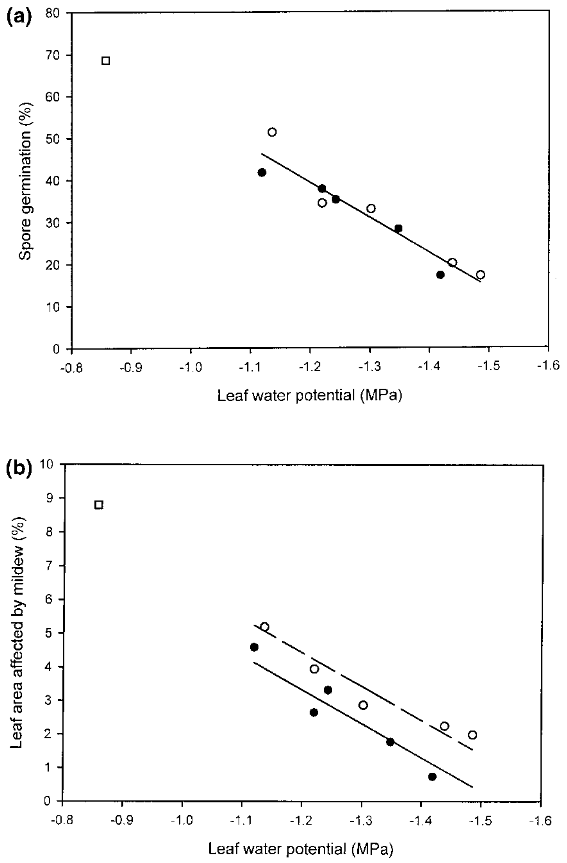

Figure 1. Relationship of (a) germination of *Erysiphe graminis* conidia and (b) leaf area affected by powdery mildew to water potential of wheat leaves sprayed previously with various concentrations of potassium chloride (filled circles, solid line in (b) or PEG 200 (open circles, broken line in (b). Data from plants sprayed with distilled water (open squares) were excluded from the regression in groups analysis.

The close relationships between the germination of spores and leaf area affected by mildew with the osmotic effect of KCl, as assessed by leaf water potential, indicates that the control of powdery mildew by KCl may be through an osmotic mechanism. The lack of difference between the response of spore germination to the salt KCl and to the relatively inert polymer PEG lends support to the hypothesis that control of mildew by KCl is through an osmotic mechanism on spore germination. Because KCl had a significantly greater effect on mildew symptoms than PEG at any given water potential, the mode of action of KCl on disease symptoms may also involve a second, smaller effect additional to the osmotic effect on spore germination. Alternatively, some other chemical or physical property of KCl different to PEG, e.g., pH, might enhance the effect of KCl on mildew symptoms compared with PEG.

The mechanism of the osmotic effect on spore germination is presumed to be by spore dehydration, but it cannot be discounted that the effects of KCl and PEG may be mediated through other mechanisms of damage to the spores, e.g., to enzyme activity, membrane integrity or cell wall configuration. The ability of the leaf to support germination may also be reduced when stressed by osmotic effects. Leaves remained fully turgid throughout the period of the experiment, however, and no visual symptoms of scorch or other damage from the KCl were observed.

Since the main effect of KCl in reducing mildew appears to be through osmosis, any other fertiliser salt should have similar effects. A practical advantage of KCl, however, compared to some fertiliser salts, e.g., nitrates, is the low risk of damage to the plant from leaf scorch (Gray, 1977). We propose that, given further research, there may be a role for crop protection agents based on an osmotic mode of action, and that the total expenditure on crop inputs might be reduced by transferring a proportion of soil-applied KCl fertiliser to foliar application.

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