The inhibition of fatty acid elongation by pebulate can be effectively counteracted by the safener dichlormid
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Abstract
The thiocarbamate herbicide pebulate inhibits fatty acid elongation, which is necessary for surface lipid biosynthesis. As both barley and wild oats are susceptible to pebulate, the safener dichlormid was used to study the reversal of its herbicidal effect. Fatty acid elongation was restored by a dichlormid pretreatment in barley, but not in pebulate-expressed oats.

Introduction
Thiocarbamate herbicides, as their sulphoxide metabolites, inhibit fatty acid elongation, which is necessary for plant surface wax formation [1]. Production or detoxification of these sulphoxides can be altered by safeners. One such compound, dichlormid, can effectively counteract the effects of thiocarbamates within maize and has been utilized in these studies. Its use is limited to grasses where it induces glutathione S-transferases [2], a diverse group of enzymes involved in herbicide detoxification via conjugation with glutathione (GSH).

We used wild oats (Avena ludoviciana) and barley (Hordeum vulgare) to study the thiocarbamate pebulate which, together with its sulphoxide derivative, is effective at selectively inhibiting fatty acid elongation in these crops [3]. Here we report that the herbicidal effect on fatty acid elongases is reversed by dichlormid treatments.

Experimental
Plants were grown in vermiculite at 20 °C with a 16-h light/8-h dark cycle. Shoots were sampled from 7–10-day-old plants of similar height [4].

Safener-treated seeds were incubated with a solution of dichlormid dissolved in a final ethanol concentration of 0.1% (v/v) for 24 h before planting. For herbicide treatments, pebulate was dissolved similarly and applied to the roots of the whole plant 4 h prior to lipid labelling of the shoots with [1-14C]acetate and subsequent analysis [5].

Results and discussion
Treatment of barley and wild oats with 10 μM dichlormid alone had no detrimental effects on growth. While pebulate severely limited growth in both plants, in barley there was a partial safening effect with dichlormid, not seen in wild oats (results not shown). At the lipid level, no significant change in very-long-chain fatty acid (VLCFA) labelling was found with dichlormid treatment compared with controls.

As expected, pebulate treatment caused a decrease in VLCFA labelling in both barley (Figure 1) and in wild oats (Figure 2). The reduction was found for all of the individual VLCFAs analysed, indicating that each of the elongation systems was sensitive. Since total fatty acid labelling was unaffected, there was increased labelling of fatty acids (16:0, 18:0) made de novo.

Dichlormid treatment restored VLCFA labelling to control levels in barley (Figure 1) but not in wild oats (Figure 2). This suggests that the safening compound is able to prevent pebulate toxicity in barley, most probably by accelerating detoxification of pebulate sulphoxide. Therefore, we have begun experiments to examine glutathione S-transferases in barley and wild oats in order to probe the reason(s) for these differences.

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References

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Expression of the wax-specific condensing enzyme CUT1 in Arabidopsis

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Abstract

The Arabidopsis thaliana gene CUT1 encodes a very-long-chain fatty acid-condensing enzyme required for the production of epicuticular wax in bolting stems. We have examined the expression pattern of CUT1 in Arabidopsis at different developmental stages and under different environmental conditions. RNA blot analysis showed that CUT1 was highly expressed in shoots, but not in roots. CUT1 expression was detectable throughout development. Light was required for CUT1 expression, and expression was increased by salt.

Key words: epicuticular wax, VLCFA.
Abbreviations used: GUS, β-glucuronidase; VLCFA, very-long-chain fatty acid; 35S CaMV, 35S cauliflower mosaic virus.

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