Effects of hydroxamic acids on the resistance of wheat to the aphid *Sitobion avenae*

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Summary

Resistance to the grain aphid, Sitobion avenae was assessed in relation to levels of hydroxamic acids, in particular DIMBOA (2,4-dihydroxy-7-methoxy-1,4-denzoxazin-3-one) in cultivars and species of Triticum and Aegilops. Antibiosis (measured as intrinsic rate of increase (r_m), and its components) was recorded. Strong correlations were demonstrated between r_m and acid levels, with correlation coefficients up to 0.96. Genetic patterns of acid levels within the wheat genome have been detected, which increases the potential for a plant breeding programme aimed at improving levels of aphid resistance in modern wheats.

1.1 Introduction

Sitobion avenae F. is a sporadically damaging pest of wheat in temperate climates (5, 10, 3). Wheat extracts contain hydroxamic acids (Hx) (11) which have been shown to be important in resistance against insects in several Gramineae (6, 7, 8, 1). The most abundant of these acids in wheat is 2,4-dihydroxy-7-methyoxy-1,4-benzoxazine-3-one (DIMBOA). This compound has also been shown to be involved in the resistance of several wheat cultivars to the aphid species Metopolophium dirhodum (Wlk.), Schizaphis graminum (Rond.) and Rhopalosiphum maidis (Fitch) (4), but S. avenae has not been investigated in this context.

The main objective of this investigation was to assess a range of Triticum species and cultivars in order to measure and rank them for antibiotic resistance to S. avenae, and for hydroxamic acid levels at the seedling stage. Any correlations between aphid performance and acid levels would have implications for future screening of wheat material against this important European pest and would usefully reinforce the relationship demonstrated by (2).

1.2 Methods

Seed of Triticum material was germinated in a culture room based on the design of (9). The temperature was 20°C with a 2°C range. Light intensity was 75 $\mu Em^{-2}s^{-1}$. Plants were harvested at the two-leaf stage(G.S.12) (12). A portion 1.5 g, of plant tissue (10-15 plants)was then macerated with a mortar and pestel in water (6 ml total volume). filtered through cheesecloth and left for 15 min at room temperature. The extract was adjusted to pH 3 with 1 M HCl and centrifuged at 10 000 g for 10 min. The supernatant was extracted three times into equal volumes of ethyl ether and the organic

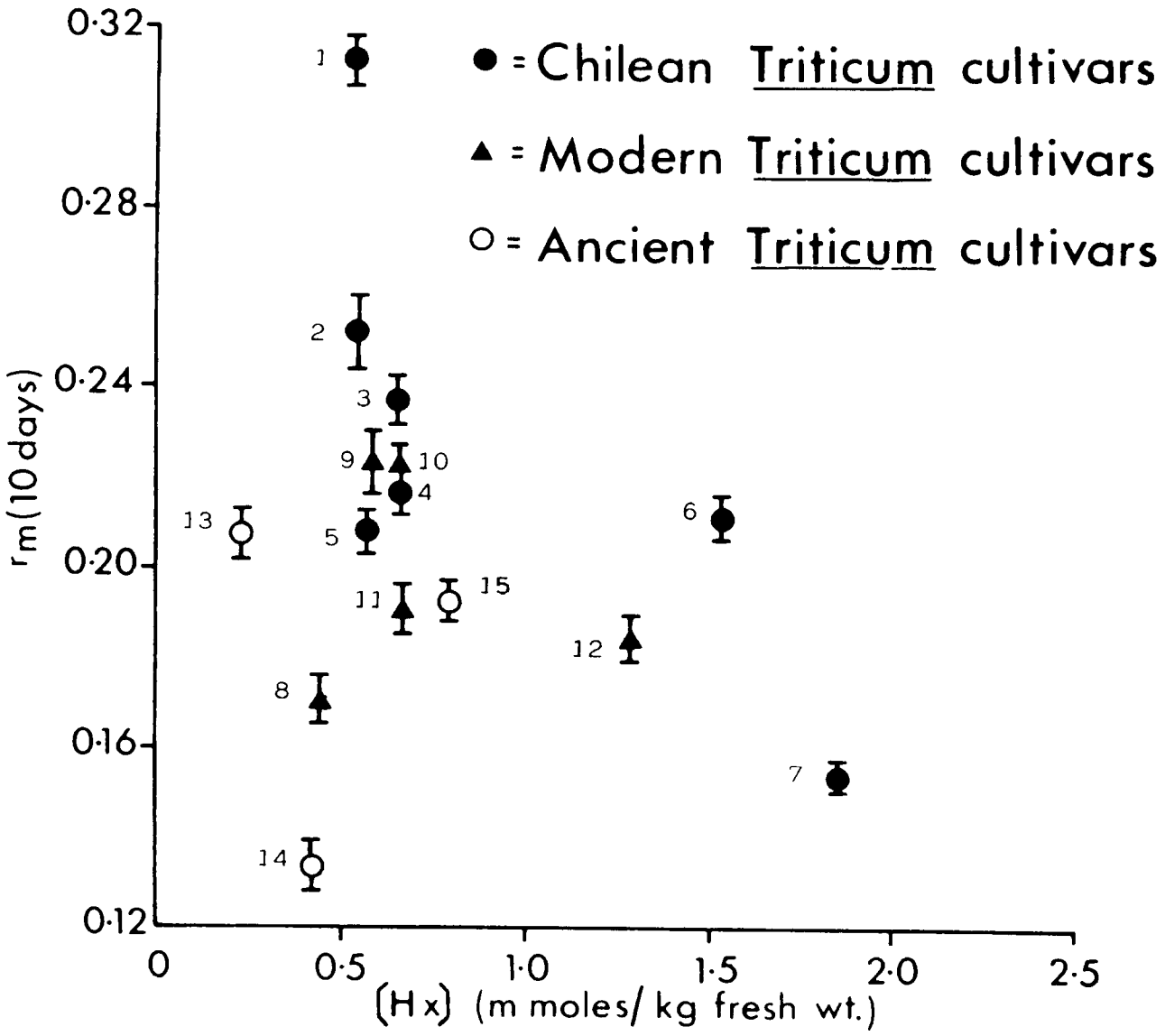

Fig. 1. The relationship between r_m and Hx levels in 13 Triticum taxa.

Chilean cultivars: 1) Huenufen; 2) Sonka; 3) Likay; 4) SNA2 (T. durum); 5) Naofen; 6) Quilofen;7) SNA3 (T. durum)
Modern European Cultivars: 8) Jerico; 9) Armada; 10) Hobbit; 11) Musket; 12) Avalon.
Ancient Cultivars: 13) Triticum aestivum 15189: 14) Triticum monococcum 377666; 15) Triticum polonicum, 384345.

phases were evaporated to dryness. Hx forms a blue complex (λ_{max} = 590 nm) upon the addition of ferric chloride reagent (50 g $FeFl_3.6H_2O$, 500 ml 95% ethanol and 5 ml 15 M HCl). The concentration of Hx in the tissues was determined by comparing the absorbance of extracts with a standard curve made with DIMBOA from maize (cv. Fola Union). Details of aphid culturing and bioassay of the plant material are given in(2).

1.3 Results and Discussion

Values for intrinsic rate of increase (with 95% confidence limits) and hydroxamic acid levels are plotted in Fig. 1. The relationship between

intrinsic rate of increase and hydroxamic acid levels is much less clear than that previously demonstrated (2). There are at least two possible reasons for this. One is that the range of acid concentrations detected in the current plant material is much smaller than that published in earlier work (2). Secondly, the growth-form of the non-commercial Triticum species used in this study differs markedly from the commercial Chilean and European cultivars which are also included in Fig. 1. The closeness of the growth-form of 'wild' Triticum species to that of non-agricultural grasses may mean that there is a higher ratio of cell wall and fibrous material to that of cell contents than in commercial cultivars. The crude acid assay, based on fresh plant weight, may lead to difficulties of interpretation when old and modern Triticum material is compared in the same data set, as in Fig. 1.

Differences between values calculated for the Hx concentrations in the same cultivars grown in Chile and in Southampton may be attributed to differences in assay techniques, and in plant ages. During Hx extraction from different cultivars, ether evaporation in Chile was achieved rapidly by rotary evaporation. This technique was not employed in Southampton, and consequently the much slower evaporation may have allowed for considerable Hx degradation. Although plants were harvested at the 2 leaf stage in both laboratories, plant heights may not have been the same, and slight differences in age could lead to the detection of quite different Hx levels in the same cultivars. Hx concentrations are known to decline rapidly in seedlings of most modern wheats.

Future work will expand the existing screen in four major ways: 1) increase the genetic range of the Triticum (and Aegilops) material investigated; 2) investigate modified analyses of hydroxamic acid levels which will permit more realistic comparisons between modern and ancient cultivars and species; 3) screen the plants for acid levels and aphid resistance at early and late growth stages; 4) re-evaluate the plant material for resistance to dipterous and molluscan pests of seedling cereals.

1.4 Acknowledgements

We thank Dr. C. Law of the Plant Breeding Institute, Cambridge, Great Britain for advice and for the provision of Triticum seed.

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