Assessing the impact of nitrogen supplementation in oats across multiple growth locations and years with targeted phenotyping and high-resolution metabolite profiling approaches

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Summary:
Oats (*Avena sativa* L.) are increasingly viewed as a healthy food source, being high in dietary fibre (e.g., β-glucans), as well as antioxidants, minerals, and vitamins. Despite its importance for the end-user and milling industry, oat milling quality and grain composition are not currently considered in UK agricultural guidelines for optimal nitrogen application levels. The selection of oat variety and crop management practices and their role in determining oat nutritional quality are important areas within which to gain further understanding. In this study, the response of four oat varieties to a wide range of added nitrogen levels (0 to 250 kg N/ha applied) was investigated across multiple locations and years with respect to yield, grain quality, and grain metabolites (assessed via high resolution GC- and LC-MS).

The application of nitrogen significantly enhanced grain yield across all trials, but this was not significant beyond the application of 200 kg N/ha. The grain yield of a cereal crop like oats can be split into three major components: the panicle population density, the number of grains per panicle, and the individual grain weight. In all three trials, although thousand grain weight (TGW) was significantly affected by nitrogen, it was not correlated with grain yield. Grain yield was, however, highly correlated with the number of grains per metre squared (m^{-2}; r=0.99). The grain number m^{-2} is a combination of panicle number m^{-2} and grain number per panicle, both of which increased significantly as nitrogen increased in all trials. These results suggest that productive tiller survival rate increases as greater levels of nitrogen are provided. However, it was found in this study that the panicle number m^{-2} did not increase beyond applications of 100 kg N/ha. Grain number per panicle did continue to increase with higher applications of nitrogen. Oats display phenotypic plasticity in response to soil-climate conditions, but this is strongly influenced by variety. Significant differences between varieties were found for the three yield component traits, with the variety 'Mascani' observed to have the lowest grain number per panicle and the highest panicle number m^{-2}, as well as the highest TGW under all treatments. The results suggest that, whilst the different oat varieties can change yield component structure in response to changing nitrogen levels, the actual grain yield remains unchanged. A significant effect of variety, nitrogen treatment, and interaction between the two was found for hectolitre

weight, which, in general, decreased at higher nitrogen application levels, as did thousand grain weight.

A novel high-resolution UHPLC-PDA-MS/MS method was developed, validated, and shown to be superior to a previous rapid (sub 15 minute) method, especially with respect to improved metabolite enrichment, resolution, and identification capabilities. Through the combination of phenotyping approaches, amino acid metabolism was shown to be upregulated by nitrogen supplementation, as were total protein and nitrogen-containing lipid levels, whereas health-beneficial avenanthramides were down-regulated. Varietal differences in metabolic phenotype were also identified. Although nitrogen addition significantly increased grain yield and β-glucan content, certain traits, such as β-glucan, total oil content, and a number of metabolites, were as much affected by oat variety as by addition of nitrogen. Conditions that result in higher yield do not necessarily result in higher grain quality or nutritional value.