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ON STARCH METABOLISM IN WHITE CLOVER STOLONS.**

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The Effect of Elevated CO₂ under Field Conditions on Starch Metabolism in White Clover Stolons.

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Climate change models predict substantial increases in atmospheric carbon dioxide (CO₂) concentration over the next fifty years. Elevated CO₂ will affect plant metabolism directly *via* photosynthesis and assimilate abundance and indirectly by greenhouse forcing. Perennial crops like white clover (*Trifolium repens* L.) often show high sensitivity to environment during overwintering. The winter survival of white clover is associated with the deposition of starch reserves in autumn, and their utilization during winter and spring. In the laboratory it is possible to produce white clover stolon material showing different patterns of starch metabolism, and to study starch accumulation and starch degradation in different environments. However, it is difficult to assess whether the effects of elevated CO₂ in controlled environments are a real reflection of the consequences of this component of climate change. Therefore, samples from field experiments have been analysed. Some samples came from open-topped chambers at IGER, North Wyke, but the main experiment is under realistic field conditions in the Free Air Carbon Dioxide Enrichment (FACE) rings at the ETH-Eschikon Experimental Station near Zurich. These rings provide good temporal and spatial control of CO₂ concentration in an environment otherwise unchanged from field conditions. There are three replicate rings: the controls have ambient CO₂ concentration (350ppm) and the high CO₂ rings are fumigated to 600ppm.

Cloned stolon tip cuttings of Menna were planted in April, 1994. Plant establishment was good: a similar number of plants survived per plot in control and fumigated rings (Table 1). By mid-July there was nearly complete cover. In mid-October the plots were defoliated for the winter. The dry matter (leaf and petiole) removed from a 40 by 50 cm sub-plot during this defoliation was 45% greater for fumigated rings but this was not significant (Table 1). There may have been insufficient sink demand for prolonged stimulation of carbon fixation as these plots were cut only infrequently during the summer. The greatest stimulation of clover growth by high CO₂ has been observed in the dry matter removed by regular harvesting. There was little canopy to remove when the plots were cut in March this year after overwintering. Fumigated plots again yielded more dry matter but again this was not significant (Table 1).

At both these harvests stolon material was collected and transported to Aberystwyth for detailed analysis of starch and soluble sugar content. Older stolon sections (internodes 4-6) are the major site of storage for starch. In October these sections had a high starch content, and CO₂ fumigation had increased starch and non-reducing sugars although not significantly (Table 2). By March, as spring growth began, the major part of these starch reserves had been utilised. High CO₂ and control plants now had the same stolon starch content. Soluble sugar content of stolons was higher in spring than in autumn but there were no significant effects of fumigation (Table 2). Overall high CO₂ had few effects. In contrast preliminary results for samples from weekly-cut clover plots in open-topped chambers during active growth in early summer suggest that elevated CO₂ may significantly reduce the deposition of starch reserves in stolons. Dry matter production was increased by high CO₂ in this experiment. The sink demand that drives stimulation of photosynthesis under these conditions may also have shifted carbon allocation toward growth rather than storage.

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TABLE 1. PLANT ESTABLISHMENT AND GROWTH.

Survival out of 25 through the establishment phase, and dry matter yield (g.m^{-2}) at October and March harvests for white clover plants grown in control and high CO_2 fumigated *FACE* rings. $n=3$.

	Control	Fumigated	se	
Establishment	20	22	3	NS
Autumn DM	95.3	138.1	13.1	NS
Spring DM	11.7	15.0	7.9	NS

TABLE 2. STOLON CARBOHYDRATES.

Reducing(R), non-reducing(NR) sugars and starch content (mg.g⁻¹dw) of older (internodes 4-6) stolon sections in autumn and spring harvested from white clover grown in control and high CO₂ fumigated *FACE* rings. n=3.

	Autumn		Spring		se
	Control	Fumigated	Control	Fumigated	
Starch	234.8	260.0	5.0	5.0	11.1
NR	99.4	109.7	125.8	151.9	10.9
R	14.7	12.8	61.9	72.4	5.9