Two interesting points have emerged from this work. The first lies in the fact that the *Rh*-antigen extracts can be removed from the saline by centrifuging at 10,000 r.p.m. These extracts can therefore be washed by repeated centrifugation through saline, and can be concentrated so that only very small injections of material are needed for immunization of guinea pigs.

The second and really surprising finding is that good anti-*D* sera were produced in 50 per cent of the animals immunized with concentrated extracts of *Rh*-negative blood. A single absorption only with an equivalent volume of *Rh*-negative cells was required when the sera were diluted 1/10 to produce good reacting anti-*D*. This curious finding has been repeated using different *Rh*-negative bloods, and the same results were obtained on each occasion.

Paradoxically, then, it appears possible to produce anti-*D* sera by the injection of extracts from cells supposedly lacking *D* antigen.

One possible explanation of this finding may be that a single substance forms the basic material for all *Rh*-antigens, and that their specificity is imparted by special side-chains or groups. However, the position is complicated considerably by the fact that in the last experiment made, the *Rh*-negative blood used was obtained from a donor who was herself immunized against the *D*-antigen during pregnancy.

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Immediate and After-effects of Carbon Dioxide Concentrations upon the Growth of Etiolated *Avena* Seedlings

THE influence of high concentrations of carbon dioxide on the extension growth of the *Avena* seedling was reported in an earlier communication[1]. Further observations, briefly noted here, have been made; but the work has been interrupted pending the completion of a thorough examination of the factors contributing to variability in the growth of the control plants and means devised for eliminating the sources of error.

The plants were grown in darkness in an incubator and they were continuously swept either with air (for the controls) or with known different concentrations of carbon dioxide. Pure carbon dioxide was supplied from a cylinder, and diluted with air by mixing streams of known rate of flow.

Increasing the concentration up to 10 per cent had a progressively depressant effect on the mesocotyls up to the third day after planting; the measurements were: controls, 16·0 mm.; 2½ per cent CO_2, 9·9 mm.; 5 per cent, 6·0 mm., and 10 per cent, 3·5 mm. This was followed by a high rate of growth and the attainment of a final length greater than that of the controls. The optimum concentration seems to be about 5 per cent, for with 10 per cent the early slow growth persisted and the length reached on the seventh day was less than when 5 per cent was employed, namely: controls, 71·5 mm.; 2½ per cent CO_2, 86·3 mm.; 5 per cent, 106·1 mm.; and 10 per cent, 60·0 mm. If growth of these last plants was allowed to continue, they attained at nine days old a length of 98·8 mm., so that stimulation had taken place but its expression was delayed.

Growth of the coleoptiles was also depressed during the first three days, and the slower rate of growth persisted so that the final lengths were shorter than those of the controls, which measured 69·0 mm. In 2½ per cent CO_2 they were 70·0 mm.; in 5 per cent, 46·7 mm.; in 10 per cent, 18·0 mm.; again, these last plants continued to elongate slowly and when measured on the ninth day they were 30·9 mm. long.

Further analysis of the phenomenon was undertaken to find whether stimulation of the mesocotyl could take place at any time during the eight-day growth period under examination. Plants were grown for three days in air, after which 5 per cent carbon dioxide was continuously supplied, and three days after this treatment was begun a slight stimulation of the mesocotyl was apparent with a correlated depression of the coleoptile. When, however, carbon dioxide was introduced after five days growth in air, that is, when the rate of mesocotyl elongation had passed its maximum, neither stimulation of the mesocotyl nor depression of the coleoptile was noted.

Perhaps the most interesting observation was that, when plants were grown in 5 per cent carbon dioxide for the first three days only and then transferred to air, the initial depression of the mesocotyl was again followed by an enhanced rate of growth as occurred when the gas was supplied for the whole growing period. Increase in length tailed off after the fifth day, however, and the mesocotyls (seven days old) measured 78·5 mm. It would appear, therefore, that the effect took place during the time when elongation was slow, and the greater part of the stimulation was consequently an after-effect.

The opportunity may perhaps be taken to comment on the suggestion recently made by Burton[2] that carbon dioxide might not be the agent causing this stimulation of mesocotyl growth.

In the first experiments on the effects of carbon dioxide using continuous gas flow (10 litres/hour per 57 plants), the gas was generated from sodium carbonate and sulphuric acid (both 'Analar'). The stimulatory effect was thus established in pure carbon dioxide, and it was only later that compressed carbon dioxide from cylinders was used to avoid the cumbersome preparation and storage of large quantities of gas. As similar results were obtained for a given concentration of carbon dioxide, the absence of other volatile stimulants was assumed. Moreover, Burton quoted examples of the stimulatory effect of ethylene, and seemed to imply that it might similarly be concerned in the case of *Avena*, but this is not so as ethylene in small quantities has a marked depressant effect[3,4]. The possibility of contamination of commercial carbon dioxide by products of respiration when the gas is prepared from the carbon dioxide of yeast fermentation cannot altogether be excluded, but as stated above there is no evidence of such a different effect from that of pure carbon dioxide.

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