sponding flapping-plane vertical momentum; the reverse is true (Fig. 4).

The secondary importance of drag is corroborated by the low turbulence level of the downstream recordings. Should the basic vertical force stem from drag, a large drag coefficient would be necessary—one equivalent to a full stall throughout the downstroke. Comparison of the downstream turbulence levels of the wing flapping at a deliberately stalled attitude with those obtained at proper incidence indicates that, in a simulated condition, the stall or near-stall region is small, covering less than 20 percent of the total effective azimuth; it follows that the average drag coefficient must be small. As the findings regarding both momentum and turbulence oppose the drag hypothesis of vertical force, the hypothesis appears to be an unlikely mechanism.

Unsteady effects, significant to both circulatory and virtual mass forces, were evaluated through a series of tests in which the unsteady quality was reduced in steps. Noting that the greatest vertical force is sensed at mid-downstroke azimuth, I chose as a performance index the vertical impulse experienced by air moving through a unit area located at this position in the course of a single downstroke. This value was obtained by integrating the instantaneous vertical force, with respect to time, as the wing swept the hot-wire sensor located at the $0.7R$-radius midstroke azimuth; the appropriate value for a simulated flight is 3.32×10^{-3} g·sec/cm^2.

The wing was then arranged to represent near-steady conditions. Constant tangential velocity was obtained by removing the oscillating drive and image plane and substituting a continuous drive; the resultant configuration may be described as a single-bladed propeller operating with its axis inclined at a large angle of pitch. Values of fixed-blade incidence and tangential velocity were set equal to those developed by the simulated insect at mid-downstroke. The vertical-unit-area impulse imparted by the descending blade was determined at 1.67×10^{-3} g·sec/cm^2.

True steady-state conditions were then established by setting the wind tunnel axis normal to the propeller disc; wind speed was altered to the normal component of the above tests, but all other experimental conditions were unchanged. Vertical-unit-area impulse measured 1.19×10^{-3} g sec/cm^2.

These experiments do not support Jensen's (*14*) conclusions that insect flight in general may be treated as a sequence of stationary flow situations. It is apparent, from the unit-area-impulse results, that unsteady effects dominate simulated performance of *M. vulgaris*. Whether this influence is manifested through vigorous destalling (circulation concept) or virtual mass forces remains to be demonstrated.

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Root Pressure and Leaf Water Potential

Abstract. *Measurements with thermocouple psychrometers were made of the water potentials of leaves from sunflower and pepper plants which had stood overnight in the dark in nutrient solutions containing carbowax. Similar measurements on the solutions showed that they had lower water potentials than the leaves, although the plants were measurably transpiring. Evidence that root pressure plays a part in this inversion of water potential gradient is presented.*

In studies of the movement of water through plants it is generally accepted that the gradient of water potential increases (negatively) from the soil, through the plant to the atmosphere, and that water (or in the final stage water vapor) flows passively down this gradient (*1*). The energy source for maintaining the gradient is the evaporative demand of the plant's aerial environment. If this view is correct, then leaf water potential must always be lower (more negative) than soil water potential, as long as the atmosphere is the final sink. I now report that, under certain conditions, leaf water potential may in fact be higher (less negative) than the water potential of the rooting medium, although the overall direction of flow is still into the atmosphere. Supplementary experiments suggest that root pressure may be responsible for this inversion of part of the water potential gradient.

Pepper (*Capsicum frutescens* L. 'Californian Wonder') and sunflower (*Helianthus annuus* L. 'Large Grey') plants were grown in the greenhouse in full strength Hoagland culture solution with iron supplied as sequestrene 138 Fe-chelate. When used for experiments the pepper plants were 2 to 3 months old, and the sunflower plants were about 6 weeks old. In the experiments, plants were transferred to a humidity- and temperature-controlled growth cabinet and kept in the dark for 8 to 12 hours prior to sampling. At the time of transfer, water potential of the culture solution (about −0.5 bar) was lowered further by the addition of varying amounts of carbowax (1540 or 4000) to give a range of water potentials of −3 to −10 bars. Cabinet temperature was 23°C and relative humidity high (90 percent) or low (30 percent). Leaf water potentials were measured with Spanner-type psychrometers and corrected for tissue respiration (*2*). Duplicate measurements of solution water potentials were made with Spanner or Richards and Ogata psychrometers, or both (*3*). Experiments of this type show (Table 1), that for pepper at high relative humidity, leaf water potential is always higher than that of the solution bathing the roots, even when the solution water potential is nearly −10 bars. At low relative humidity, this inverted gradient is less steep, but is still maintained in all cases except for the −10 bar solution. Similar inverted gradients are found at high relative humidity in sunflower plants grown in full strength Hoagland solution, but for sunflower plants grown in one-fourth strength solution, these gradients are not inverted.

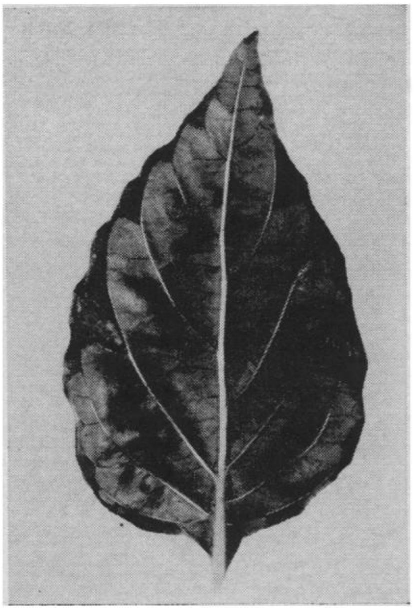

Fig. 1. Underside of pepper leaf showing injection of intercellular spaces due to guttation from the vein endings in the leaf margins.

Rawlins (*4*) has described an inverted potential gradient for pepper (solution −7.2 bars, leaves (average) −4.2 bars), similar to those in Table 1, using the Richards and Ogata psychrometer. He attributed his result to error arising from a combination of low leaf permeability and the use of the Richards and Ogata psychrometer. His interpretation implied that use of the Spanner psychrometer would have given lower leaf water potentials, the gradients being returned to their conventional direction. I reported good agreement between leaf water potentials as measured by these two types of psychrometer and concluded Rawlins' explanation was incorrect (*5*). I also pointed out that, even after applying a normal correction for leaf respiration, the gradient would have remained inverted, and at the time I was not able to offer a wholly satisfactory explanation for this. In view of leaf water potentials measured with Spanner psychrometers (Table 1), it now seems that Rawlins' water potential measurements were valid, although a small correction for respiration should be applied to them. The apparent equality of solution and pepper leaf water potentials observed in my repetition of Rawlins' experiment is also consistent with the data of Table 1. The solution used had a potential of −10 bars; for this solution potential, depending on the humidity, leaf water potential may be higher or lower than that of the solution. Presumably the humidity during this experiment must, rather fortuitously, have been the cause of the observed approximate equality between the leaf and solution water potentials.

These observations of inverted water potential gradients imply that there is a stage in the passage of water through the darkened plant when metabolic energy is used, either directly to give a truly active transport of water against a water potential gradient, or indirectly —perhaps by coupling water movement to ion movement—as suggested by Crafts and Broyer (*6*) in their explanation of the origin of root pressure. Initial experiments have suggested that such a site for metabolic intervention may be situated in the roots rather than in the leaves. Thus, root systems (plants decapitated as close to the roots as possible) exude from the cut stump while standing in a solution of −6.0 bars (pepper) or −5.0 bars (sunflower), but the water potentials of detached leaves standing in the dark in high humidity, with their petioles in similar solutions, have never shown the inverted gradients. Also, detached leaves standing in culture solution only, failed to guttate in high humidity in the dark, but leaves on intact plants of both pepper and sunflower guttated freely under these conditions, with eventual marked injection of the marginal intercellular spaces where the vein endings are situated (Fig. 1). In old pepper plants, guttation has even been observed from cracks in the stem. These results are to be expected if, as is generally held (*7, 8*), root pressure is responsible for guttation. Vaadia (*9*) has reported that $2.5 \times 10^{-5} M$ 2,4-dinitrophenol applied to the roots inhibits root pressure in sunflower; it should therefore also inhibit guttation, and I have observed this within 1 hour of applying it to the roots of a previously guttating sunflower. Hence, I adopted the working hypothesis that root pressure is responsible for the observed raising of leaf water potential above that of the solution bathing the roots. The alternative explanation that the inverted gradients were due to the reversal of the direction of water flow, that is, from the air to the plant, was ruled out by experiments in which transpirational losses were measured over 6 hours from plants having inverted gradients. At 90 percent relative humidity a sunflower plant lost 2.1 g/hr (ψ_{plant}, −4.9; ψ_{solution}, −6.8 bars), and a pepper plant lost 1.0 g/hr. (ψ_{plant}, −5.3; ψ_{solution}, −7.3 bars). At 20 percent relative humidity, a pepper plant lost 4.1 g/hr, (ψ_{plant}, −4.1; ψ_{solution}, −5.9 bars).

Table 1. Comparison of water potentials (bars) of leaves from plants, after standing 8 to 12 hours in the dark (23°C) in Hoagland solution containing carbowax, with the water potentials (bars) of the solutions.

Water potential (bars)				
Leaves			Solution*	
Upper	Middle	Lower	1	2
		Pepper leaves, 90% RH		
−1.9	−2.3	−1.9	−3.6	−3.8
−3.3	−3.1	−3.3	−6.0†	−6.1†
−2.8	−3.2	−2.9	−7.5†	−7.6†
−8.6	−8.0	−7.1	−10.0	−9.5†
		Pepper leaves, 30% RH		
−3.4	−3.7	−3.2	−4.0	−3.9†
−5.5	−4.6	−4.6	−6.0	−6.0†
−6.8	−6.0	−5.5	−7.9	−7.6†
−11.3	−10.4	−9.6	−10.0	−9.5†
		Sunflower leaves, 90% RH		
−3.3	−3.3	−3.0	−5.7†	−5.7
−4.1	−3.9	−4.0	−8.2	−7.8†
−4.6‡	−5.4‡	−5.2‡	−3.2	−3.6†
−9.1‡	−9.7‡	−6.7‡	−5.2	−5.6
−8.0‡	−11.1‡	−9.3‡	−6.9	−6.9

* Duplicate determinations. † Richards and Ogata psychrometer used here, all other determinations by Spanner psychrometer. ‡ These plants grown in one-fourth strength Hoagland solution, all others in full strength solutions.

If root pressure were indeed responsible for these inverted gradients, then the gradients themselves might be expected to exhibit some of the known properties of root pressures. These properties include (i) an effect of nutrient concentration, root pressure being higher in plants grown in moderately concentrated solution than in plants in dilute solution (*8*); (ii) a rhythmic diurnal variation with a maximum at midday and a minimum at midnight, persisting even under constant conditions (*9, 10*); (iii) inhibition by metabolic inhibitors (*9, 11, 12*).

The sunflower data (Table 1) provide evidence in support of the influence of nutrient concentration. Plants grown in full strength Hoagland solution showed higher leaf water potentials than plants grown in one-fourth strength solution. In fact, the water potential gradients were no longer inverted in the one-fourth strength plants. The role of rhythmic diurnal variation was investigated by placing a pepper plant in the dark at 23°C and 90 percent relative humidity in a solution of −7.5 bars potential. The plant was allowed a 12-hour equilibration period, and the water potentials of upper and middle leaves were measured at noon, midnight, and the following noon. The observed potentials were −2.8, −3.7 and −2.9 bars (upper leaves), and

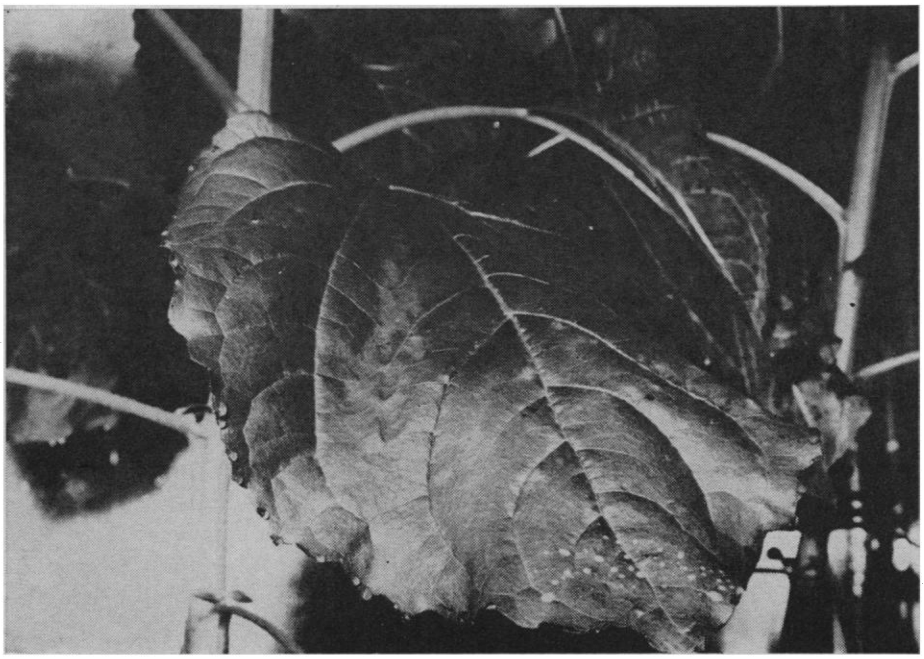

Fig. 2. Attached sunflower leaf showing copious guttation although the plant was standing in a solution with a water potential of −3 bars.

−2.5, −4.0, and −2.4 bars (middle leaves). The leaf potentials therefore varied as expected, decreasing at midnight and increasing almost exactly to their original values the following noon. Metabolic inhibition, which had already been strongly suggested in the experiment in which 2,4-dinitrophenol applied to sunflower roots inhibited guttation from the leaves, was investigated further by measuring the water potentials of pepper leaves before and after the application of $10^{-4}M$ KCN to the roots of a plant standing in a −7.7-bar solution in the dark at 23°C and 30 percent relative humidity. Leaf potentials prior to application of the inhibitor were −5.0, −5.1, −5.1, and −5.2 bars (average −5.1 bars), and after an overnight period with inhibitor, −7.1, −7.5, −7.7, and −7.8 bars (average −7.5 bars). The average leaf potentials, before and after the treatment, show that the inverted gradient between leaf and solution was almost abolished by the application of the inhibitor. This inhibitor also considerably reduces root pressure in tomato (*11*) and onion (*12*). Plants treated in this way and subsequently returned to culture solution in the greenhouse continued normal growth, their leaves and roots remaining healthy. These three experiments strongly support the view that root pressure may influence leaf water potential, at least in darkened plants.

The results presented so far have largely depended upon psychrometric measurements of leaf water potential. Shmueli and Cohen (*13*) express the view that such results are due to inadequacies of the techniques used, although they do not offer supporting experimental evidence. Kramer (*8*) has suggested that root pressure in most herbaceous plants does not exceed 1 to 2 bars, whereas the present data suggest, from the magnitude of the inverted gradients (Table 1), that it can amount to at least 4 bars in sunflower and nearly 5 bars in pepper. It therefore seems desirable to consider briefly simple supporting evidence not dependent upon leaf water potential measurements. I have reviewed evidence that guttation is due to root pressure, and it seems reasonable to use this phenomenon as a direct visual criterion for the existence of positive root pressure in the intact plant. Figure 2 shows copious guttation from an attached leaf of a sunflower plant after an overnight period in solution at −3 bar and 90 percent relative humidity in the dark. All the leaves on the plant were guttating in this way, giving a striking appearance. There was also injection of the intercellular spaces of the leaves. These observations imply that root pressure exceeded 3 bars in this plant. A similar observation has been made for a pepper plan standing in a −6-bar solution. In this case guttation was noted at 10:30 a.m. from intact leaves together with copious exudation from the petiolar stumps remaining after leaves had been removed for water potential measurements; there was also slight injection of the intercellular spaces of intact leaves. However, by 2:10 p.m. the exudation from the stumps had declined considerably and injection of the intercellular leaf spaces was reduced. In this case the observations imply that root pressure exceeded 6 bars. The appearance of injection before noon and its reduction after noon was presumably due to the midday maximum in leaf water potential already noted for this species.

My results have led to the view that, under the experimental conditions, leaf water potential in pepper and sunflower can be 4 to 5 bars higher than that of the solution bathing the roots, and that this effect is brought about by root pressure. It will be necessary to examine further the nature of root pressure before deciding whether the inverted gradients reported are due to a truly active transport of water or to coupling of water movement, perhaps with ion movement. If the latter is the case then it may be necessary to postulate, along with Eaton (*14*) and Lundergardh (*15*), that the ions causing the water movement are subsequently reabsorbed at some point higher in the plant. Whether metabolic energy is used in illuminated plants to lower leaf water potentials or can contribute significantly to their undoubtedly higher transpiration rates cannot be decided on the present evidence.

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