Short Communication

Light as a Source of Error in Estimates of Water Potential by Vapor Equilibration

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A common method for estimating plant water potential is the vapor equilibration technique. This procedure, described originally by Archichovskij and Archichovskaja (1), and modified by Slatyer (4) and Kreeb (3), involves equilibration of plant tissue in the vapor phase above solutions of sucrose, mannitol, or salts of known concentration. Humidity chambers or microdesiccators, containing the plant tissue and control solutions, are immersed in a temperature-controlled water bath, for maintenance of a constant water potential, during the equilibration period. Sensitive control of the water-bath temperature required in this procedure is normally accomplished by using one or more incandescent light bulbs as a source of intermittent heat.

Although Kaufmann and Kramer (2) have reported on the importance of respiration and the heat of respiration during the equilibration period, no mention has been made in the literature regarding the influence of light from incandescent bulbs used to control the water-bath temperature. Since the microdesiccators are clear glass laboratory jars which permit incident light to be absorbed by the plant tissue inside, it would seem important to assess the influence of light in estimates of plant water potential using the vapor equilibration procedure. This paper reports the results of such a study.

Materials and Methods

The vapor equilibration apparatus employed in these experiments was essentially the same as that described by Slatyer (4) with the exception that the water bath was maintained at 3 to 5° above ambient temperature, thus eliminating the need for mechanical cooling. The present apparatus consisted of 2 aquaria (10 gal inside a 30 gal); 2 stirrers; a micro-set thermoregulator operating through a relay; a differential thermometer; two 100-watt incandescent bulbs; and a 2-liter separatory funnel, which served as a water reservoir by which a constant water level could be maintained in the bath during operation. The water bath was maintained at a temperature of 26.0 ± 0.005° by placing the entire apparatus in a constant temperature room held at 22 ± 0.5°. The humidity chambers (microdesiccators) used in these studies were identical to those described by Slatyer (4).

To study the effect of incident light on measurements of plant water potential by vapor equilibration, duplicate sets of leaf disks were sampled from a variety of plant species including elm (Ulmus americana, L.), bean (Phaseolus vulgaris, L.), Coleus blumei, Benth., Philodendron cordatum, Kunth, and northern red oak (Quercus rubra, L.). Each sample consisted of ten 1.0-cm leaf disks cut with a sharp cork borer, weighed and placed in microdesiccators above sodium chloride solutions ranging in water potential from zero (pure water) to −27 bars. One set of disks was placed in aluminum foil-covered microdesiccators to shield out all light; the duplicate set was placed in identical jars without the foil covering. Thermocouples were imbedded in one disk from each of the shielded and unshielded containers. Thermocouples were also positioned in the air surrounding these disks, thus giving a measurement of disk temperature relative to air temperature in the 2 types of jars.

All samples were taken from well-watered plants. In addition, the water potential of moderately and severely wilted elm seedlings was also determined. In the latter instance, control solutions ranging from zero to −55 bars were used. The equilibration period in all determinations was 24 hr. Estimations of dry weight loss during equilibration were made using the procedure outlined by Slatyer and Barrs (5).

Results

Vapor equilibration measurements using leaf tissue contained in aluminum foil-covered microdesiccators gave water potential values substantially lower (more negative) than similar determinations using unshielded microdesiccators (table I). This was true with all the plant species studied. Experiments conducted with elm leaf tissue of varying turgidity (turgid, moderately and severely wilted) indicated that the effect of excluding light became less pronounced as plant water stress increased (table II). Throughout the study, water potential values were always more negative in those jars from which light was excluded, even though the magnitude of this
Table I. Influence of Light on Vapor Equilibration Measurements of Leaf Water Potential in Well-Watered Plants

Duplicate sets of 1.0-cm disks were cut from leaves of well-watered plants and equilibrated for 24 hr over NaCl solutions ranging in water potential from zero to $-27$ bars. One set of disks was equilibrated in light-shielded (S) microdesiccators, and the corresponding set in unshielded (U) microdesiccators. The values for U were always higher (less negative) than those for S.

<table>
<thead>
<tr>
<th>Species</th>
<th>Water potential in bars</th>
<th>Shielded</th>
<th>Unshielded</th>
<th>Difference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bean</td>
<td>-0.5</td>
<td>-4.8</td>
<td>4.3 ± 1.2</td>
<td></td>
</tr>
<tr>
<td>Coleus</td>
<td>-1.2</td>
<td>-4.2</td>
<td>3.0 ± 2.1</td>
<td></td>
</tr>
<tr>
<td>Elm</td>
<td>-2.0</td>
<td>-6.1</td>
<td>4.1 ± 1.3</td>
<td></td>
</tr>
<tr>
<td>Oak</td>
<td>-3.0</td>
<td>-7.9</td>
<td>4.9 ± 3.1</td>
<td></td>
</tr>
<tr>
<td>Philodendron</td>
<td>-1.7</td>
<td>-5.8</td>
<td>4.1 ± 2.4</td>
<td></td>
</tr>
</tbody>
</table>

Table II. Influence of Light on Vapor Equilibration Measurements of Leaf Water Potential in Elm Seedlings Subjected to Moisture Stress

Duplicate sets of 1.0-cm disks were cut from leaves of elm seedlings under a range of water regimens. Samples were equilibrated for 24 hr over NaCl solutions ranging in water potential from zero to $-55$ bars. The values for U were always higher (less negative) than those for S.

<table>
<thead>
<tr>
<th>Plant water stress</th>
<th>Water potential in bars</th>
<th>Shielded</th>
<th>Unshielded</th>
<th>Difference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Well-watered</td>
<td>-2.0</td>
<td>-6.1</td>
<td>4.1 ± 1.3</td>
<td></td>
</tr>
<tr>
<td>Moderately wilted</td>
<td>-19.4</td>
<td>-21.7</td>
<td>2.3 ± 0.8</td>
<td></td>
</tr>
<tr>
<td>Severely wilted</td>
<td>-46.0</td>
<td>-47.5</td>
<td>1.5 ± 0.8</td>
<td></td>
</tr>
</tbody>
</table>

difference changed with the level of plant water stress.

In some instances, especially at plant water potentials approaching zero, the discrepancy between covered and uncovered microdesiccators became particularly apparent. In some cases, leaf disks in unshielded humidity jars actually lost weight after equilibration in a saturated atmosphere above pure water. Corresponding leaf disks from shielded microdesiccators exhibited water potentials of $-5$ to $-6$ bars in some experiments.

Temperature measurements with thermocouples placed in leaf tissue and in air surrounding the tissue showed that the difference between leaf and air temperatures in unshielded jars was always greater than corresponding differences in shielded jars. The differences between the 2 types of jars varied from 0.099° to 0.176°.

Determinations of dry weight loss during equilibration indicated that respiration was significant for most species tested. However, differences in dry weight loss of leaf tissue in the 2 types of microdesiccators were not significant.

Discussion

It seems apparent from the data collected in this study that exposure to light causes appreciable error in estimates of leaf water potential by the vapor equilibration technique. The leaf tissue in unshielded microdesiccators absorbs incident light resulting in an increase in leaf temperature above corresponding tissue in shielded containers. Although one would expect greater heat of respiration in leaf tissue exposed to the light, the higher water potential values observed in unshielded containers cannot be explained on the basis of increased respiration caused by higher leaf temperature. Determinations of dry weight loss during equilibration indicated no significant difference in respiration between the 2 types of chambers: and yet the temperature differences between tissue and air were 0.099° to 0.176° greater in unshielded jars.

Since no appreciable differences in dry weight loss were observed between leaf tissue in shielded and unshielded microdesiccators, the data suggest that the primary factor responsible for higher water potentials was an increase in water loss from the warmer leaf tissue in unshielded chambers. Calculations using the equation in Kaufmann and Kramer (2) indicate that temperature differences observed in the present study are sufficient to account for the higher water potentials reported for leaf tissue in unshielded microdesiccators.

Although some question may still exist regarding the mechanism by which incident radiation influences the determination of water potential by vapor equilibration, the fact remains that light does constitute a substantial source of error in this technique, especially at plant water potentials approaching zero. Other methods for eliminating incident light (blackened light bulbs, shielded light bulbs, electric heating elements) were also effective in reducing errors attributable to this phenomenon. On the basis of these studies, it is recommended that light be excluded during measurements of vapor equilibration, since this source of error can cause significantly higher estimates of plant water potential than actually exist.

Literature Cited