C₄ Photosynthesis in Tree Form Euphorbia Species from Hawaiian Rainforest Sites

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ABSTRACT

The $^{13}$C/$^{12}$C isotope ratios and the leaf anatomy of 18 species and varieties of Euphorbia native to the Hawaiian Islands indicated that all possess C₄ photosynthesis. These species range from small prostrate coastal strand shrubs to shrubs and trees in rainforest and bog habitats. The results show that C₄ photosynthesis occurs in plants from a much wider range of habitats and life-forms than has been previously reported.

The C₄ dicarboxylic acid pathway has now been reported to be present in plants from a wide range of genera and families of both the monocots and dicots (7). This taxonomically diverse occurrence suggests that substantial potential for variation in life-form and environmental response should be present. Nevertheless, the species known to have C₄ photosynthesis are herbaceous or shrubby in life-form and are predominately found in hot and/or arid habitats. While C₄ plants do occur in cool moist habitats such as salt marshes and as weeds in disturbed sites, they have not been reported to be present in well developed mesic and wet forests. In spite of this, comparative studies of the photosynthetic CO₂ exchange characteristics of C₃ and C₄ plants have failed to reveal any reason why the C₄ pathway should be disadvantageous in these habitats (2, 13).

The Hawaiian species of Euphorbia consists of 14 native species, which occur in habitats ranging from arid coastal stands to rain forests and range in life-form from prostrate shrubs to trees (14). All are members of the subgenus Chamaesyce. Euphorbia has been previously shown to contain both C₃ and C₄ members (12, 16), with the species possessing C₄ photosynthesis belonging to the subgenus Chamaesyce and occurring principally in hot arid regions, whereas the C₃ species are native to moister regions. Herbert (10) has reported the occurrence of a parenchymatous vascular sheath in the Hawaiian species based on observations of cleared herbarium specimens. Since these Hawaiian species occur in habitats and have life-forms in which C₄ photosynthesis has not been previously reported, we have determined the $^{13}$C/$^{12}$C isotope ratios and leaf anatomy. High carbon isotope ratios ($δ^{13}$C = −8 to −18‰) are characteristic of plants possessing C₄ photosynthesis or Crassulacean acid metabolism (1) while the “Kranz” anatomy (8), typified by well developed, chlorenchymatous vascular bundle sheaths, is also indicative of C₄ photosynthesis. The combination of these two characteristics in the same plant material provides unequivocal evidence for the presence of the C₄ dicarboxylic acid pathway.

MATERIALS AND METHODS

Samples of leaves of the Hawaiian Euphorbia were obtained from herbarium specimens at the United States National Herbarium, Washington, D. C. The carbon isotope ratio in this material was determined following combustion to CO₂ in an O₂ atmosphere according to the method of Craig (3, 4). The isotope ratio was expressed as:

$$δ^{13}C_{	ext{rat}} = \left[ \frac{^{13}C/^{12}C \text{ sample}}{^{13}C/^{12}C \text{ standard}} - 1 \right] \times 1000$$

where the standard is CO₂ obtained from belemnite from the Peedee Formation in South Carolina (4). The expected measurement precision was ±0.1‰.

The leaf anatomy was determined by infiltrating with H₂O under vacuum for approximately 30 min. One- to two-mm wide slices were cut from the softened leaves and fixed for 2 hr in 2.5% glutaraldehyde in 0.15 M Na phosphate buffer, pH 7.6. The slices were then dehydrated with ethyl alcohol, embedded in butoxyethanol-glycol methacrylate plastic and sectioned (10 μm thick). The sections were stained with toluidine blue. Other sections were stained additionally with I₂-KI to reveal the presence of starch.

RESULTS AND DISCUSSION

All of the Hawaiian species of Euphorbia and the two South Pacific species investigated had high carbon isotope ratios and well developed “Kranz” anatomy indicating that they all possess C₄ photosynthesis. The $δ^{13}$C values ranged from −12.1 to −13.5‰ (Table I) and were well within the range reported for C₄ species (11, 15). Examination of the leaf anatomy revealed that all species had well developed vascular bundle sheaths comprised of large thick walled cells. Additional staining with I₂-KI revealed a preferential occurrence of starch in the bundle sheath cells as is typical with C₄ plants (6). While the amount of starch varied considerably between samples and was absent in several, it was exclusively or preferentially found in the bundle sheath cells when present.

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The life-forms and habitats of the Hawaiian Euphorbia contrast markedly and range from small prostrate shrubs in arid coastal strand habitats (E. degeneri) and drought-deciduous shrubs in arid habitats (E. celastroides var. amplexcens) to small trees in mesic and rainforest habitats (E. forbesii, E. rockii). Other species are shrubs in the understory of rain forests (E. clusiaefolia) or in bogs (E. remyi var. kahiliana). These species demonstrate clearly that the C₄ pathway is not restricted to herbaceous or shrubby life-forms and does occur in tree species as well.

The presence of the Hawaiian Euphorbias in wet and mesic habitats that support forests indicates that, as suggested by CO₂ exchange measurements (2, 13), C₄ photosynthesis is probably not disadvantageous under these conditions. The lack of reports at present of C₃ plants in tropical and temperate forests other than in Hawaii may be associated with an inability to compete with C₄ plants because of a lack of suitable life-forms. One cause of the competitive success of C₄ plants may be historical in that C₄ plants would have had to invade established C₃ forest communities. Slow rates of colonization typical of islands (11) probably resulted initially in few competitive interactions. Native Hawaiian Euphorbias appear to have evolved from a common ancestor, perhaps similar to the coastal strand species currently present (5, 9). After colonization of coastal strand habitats, Euphorbia probably invaded mesic and wet habitats and evolved suitable growth forms in the absence of severe competition. The subsequent increase in competition from C₃ plants failed to displace the Euphorbia species already occupying these habitats. In other situations where competition between C₃ and C₄ plants with similar life forms (e.g., grasses) occur, there is a tendency for growth of the C₄ species to be synchronized to cool, moist periods and the C₃ species to hot, moist, and dry periods (17). Under these conditions, physiological differences between C₃ and C₄ plants may be accentuated to take advantage of a wide range of conditions.

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LITERATURE CITED