

CARBON ISOTOPIC TECHNIQUES FOR AGING/DATING ( $^{13}\text{C}/^{12}\text{C}$ )  
AND GROWTH RATE DETERMINATION ( $^{14}\text{C}$ ) OF TROPICAL WOODS

A Proposal by:

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## ABSTRACT

Tree growth rings have been the key to "aging" (dating) of wood materials, particularly in the temperate zone. Unfortunately, in tropical regions these methods do not work well because several flushes of growth may occur during a single season giving rise to several rings, and in the extreme there are some species which show no growth rings at all. These problems are not peculiar to tropical trees only, as some temperate species such as Prosopis (mesquite), Acacia and evergreen Quercus (oak) may exhibit similar tendencies. Standard methods of wood chemistry and wood structure have yet to prove effective in aging these woods, but we believe that seasonal variations of the  $^{13}\text{C}/^{12}\text{C}$  ratio within the wood may provide a powerful new tool for dating (aging) such samples.

Such a seasonal variation has already been reported in leaves from Canada and tree rings from New Zealand. Our most recent research has verified this seasonal isotopic variation in leaves and rings of juniper from Arizona, suggesting that a) it is a repeating pattern which may be oblivious to "false" growth rings, and b) the signal in the wood is linked to changes in the leaves. The source of this variation, however, is currently unknown. It may represent a physiological effect or it may be environmentally motivated by such parameters as temperature, sunlight, moisture and/or seasonal carbon dioxide variations in the atmosphere.

We propose to conduct experiments over the next two years which will a) reveal the suitability and limitations of this method to the aging of tropical wood, and b) resolve the source of these seasonal variations as physiological or environmental. On one hand, we plan primarily to use temperate trees for controlled growth and field experiments required to determine the source of this seasonal isotopic signal and its natural variation among trees of the

same species (and same site). On the other hand, we will analyze a number of tropical species (some of known age) in order to gauge the universality of application, potential problems, and the kind of precision that may be expected.

In a separate technique, radiocarbon measurements may allow us to rapidly and precisely determine recent growth rates without long-term dendrometric monitoring. Atmospheric atomic weapons testing has put a  $^{14}\text{C}$  "label" onto air  $\text{CO}_2$ . This label is transmitted to the wood of trees through carbon fixation and assimilation, so that they should also contain a characteristic pattern of excess  $^{14}\text{C}$  input and decay for the post-1950 period. We plan to test the applicability of using as few as 2 radiocarbon measurements to constrain the age of a given portion of wood with respect to the atmospheric  $^{14}\text{C}$  pattern, and to apply the method to determining average growth rates over the most recent 20-30 years.

## I. INTRODUCTION

The dating or "aging" of temperate woods is commonly accomplished by dendrochronological methods based on variation in tree-ring widths. However, standard dendrochronology is subject to limitations. Certain species may be considered non-datable, and under special circumstances, normally datable species may be considered undatable (e.g., "complacent" ring series).

We have completed some preliminary research which suggests a potentially powerful new tool to aid in dating what heretofore have been considered non-datable species. In a variety of woods we have examined, the stable carbon isotopic composition ( $\delta^{13}\text{C}^*$ ) changes over the width of a ring in what may be a characteristic, repeating manner. Wilson and Grinsted (1977) first observed this phenomenon, which we have now examined in greater detail (Leavitt and Long, 1982b) with some dating applications in mind (Leavitt and Long, 1982a). The early results seem to indicate this phenomenon may be particularly useful in dating otherwise "non-datable" trees or species without ring structure, and in distinguishing "false" from true tree rings where ring structure is present. The environmental source of this isotopic variation is not yet known, although some of the observed  $\delta^{13}\text{C}$  variation correlates well with seasonal temperature changes. At this point, however, the possibility that this phenomenon has a physiological basis cannot be excluded.

Most of the next section outlines what little is known about these intra-annual  $\delta^{13}\text{C}$  signatures. It is evident that understanding more fully the capabilities and limitations of this tool will require a full-scale examination of this isotopic phenomenon. This proposal outlines a two-year program

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$$(*\delta^{13}\text{C}(\%)) = \left[ \frac{^{13}\text{C}/^{12}\text{C} \text{ sample}}{^{13}\text{C}/^{12}\text{C} \text{ std}} - 1 \right] \times 1000, \text{ where the PDB calcite is the accepted standard (Craig, 1957).}$$

we feel is needed to realize the potential of this tool, particularly in regard to tropical applications. Generally, it is designed to determine (1) the cause(s) of the seasonal isotopic variation, (2) the variability and ubiquity of variation, and (3) the suitability of this variation for dating tropical woods.

Finally, for studies where recent growth rates are required, they may rapidly be determined with as few as 2 to 3 radiocarbon measurements per tree. This is because atomic weapons testing has artificially charged the atmosphere with various radioactive tracers, including  $^{14}\text{C}$ . The characteristic post-1950 pattern of this excess radiocarbon should also appear in the wood of trees, allowing us to determine the age of the outer portions of wood by comparison and thereby its growth rate.

We feel the outcome of this project may benefit not only the fields of tropical ecology, forest management and silviculture, but perhaps the fields of anthropology and carbon-cycle studies as well.

## II. BACKGROUND AND STATUS OF WORK

### A. Basis for Seasonal $\delta^{13}\text{C}$ Variation

For the  $\delta^{13}\text{C}$  pattern within a ring to be dependable for dating, some consistent seasonal pattern of variation of an environmental parameter must positively or negatively affect the carbon isotopic composition laid down in the xylem tissue. For stable carbon isotopes such parameters might include temperature, precipitation, sunlight and atmospheric carbon dioxide concentration. For example, at northern hemisphere temperate latitudes the temperature trends would consist of cool temperatures at initiation of growth, temperature increase to a July-August maximum, then cooling in September and October. Several in vivo and in vitro studies with various plants and plant

enzymes have shown good correlations of  $\delta^{13}\text{C}$  with temperature (Table 1). Coefficients are both positive and negative, although most are negative when marine plants are excluded. Long (1982) notes that there may be difficulties detecting a direct temperature effect because the primary step controlling carbon fixation involves enzymes and kinetic processes. Experiments by Schmidt and Winkler (1979) also support no direct temperature effect on enzyme fixation, but show a pH dependence such that strongly illuminated plants in hot climates would have high pH values and isotopically light  $\delta^{13}\text{C}$  values.

Light intensity could affect rate of photosynthesis and in turn influence the carbon isotopic composition. This could be manifested seasonally by sun angle, day length, and perhaps diffusion of sunlight by seasonal cloudiness. Park and Epstein (1960), however, found tomato plants exposed to high light intensity only slightly heavier ( $\sim 1\%$ ) than those grown under lower intensities. Smith, Oliver and McMillan (1976) found one  $\text{C}_3$  species whose  $\delta^{13}\text{C}$  became isotopically heavier with increasing light intensity, but another whose  $\delta^{13}\text{C}$  became lighter.

Precipitation (or water stress) may also have a seasonal character and could influence both stomatal opening for carbon dioxide and the water available for photosynthesis. In the southwestern U.S. the pattern might typically be abundant moisture at initiation of growth, moisture stress in late May and June, then abundant moisture in July and August with the onset of summer rains. Little is known of this effect on carbon isotope fractionation although a study of Cicer arietinum L. (chickpea) by Winter (1981) indicates water stress induces less negative  $\delta^{13}\text{C}$  values. He interprets this as an effect of restricted  $\text{CO}_2$  diffusion.

Finally, a seasonal pattern in atmospheric carbon dioxide concentration results in lower  $\text{CO}_2$  concentrations in summer than winter over the Northern

Hemisphere, primarily due to CO<sub>2</sub> uptake by the temperate biosphere. The carbon isotopic composition of the atmospheric CO<sub>2</sub> also changes seasonally, and is related to the CO<sub>2</sub> concentration by an approximate coefficient of -0.053‰/ppmCO<sub>2</sub> (Pearman and Hyson, 1980). Because seasonal CO<sub>2</sub> concentrations are latitude-dependent, the seasonal δ<sup>13</sup>C variation at Hawaii is about 0.4‰ while at Pt. Barrow, Alaska, it is about 1.0‰ (Broecker et al., 1979). In Arizona, trees will be exposed to an annual CO<sub>2</sub> δ<sup>13</sup>C fluctuation of about 0.5‰ (C.D. Keeling, 1982, pers. comm.). In the tropics, the seasonal changes in atmospheric CO<sub>2</sub> concentrations are even lower. Changes in CO<sub>2</sub> concentration alone, however, seem to influence δ<sup>13</sup>C of plants as Park and Epstein (1960) found tomato plants were isotopically lighter with increasing CO<sub>2</sub> concentrations.

Francey and Farquhar (1982) have devised a model in which the δ<sup>13</sup>C of a plant is ultimately dependent on both the δ<sup>13</sup>C of atmospheric CO<sub>2</sub> and the ratio of plant internal to external CO<sub>2</sub> concentrations:

$$\delta_p^{13} = \delta_a^{13} - a - (b-a) C_i/C_a ,$$

where subscript p = plant, a = air and i = internal, variable C = CO<sub>2</sub> concentration, variable a is the fractionation of <sup>13</sup>CO<sub>2</sub> relative to <sup>12</sup>CO<sub>2</sub> during diffusion in air (~4‰) and variable b is the isotopic fractionation associated with the enzyme fixation of carbon (~30‰). The ratio of internal to external CO<sub>2</sub> concentrations are in turn affected by the rate of CO<sub>2</sub> assimilation (A) and the leaf conductance to CO<sub>2</sub> diffusion (g):

$$C_i = C_a - A/g .$$



Isotopically light  $\delta^{13}\text{C}$  values are favored by more negative atmospheric  $\delta^{13}\text{C}$  and by any factors which may decrease the rate of  $\text{CO}_2$  assimilation (A) and/or increase the stomatal conductance of  $\text{CO}_2$  (g). Very low light intensities would decrease assimilation and lower plant  $\delta^{13}\text{C}$ , but water deficits would reduce stomatal conductance and increase plant  $\delta^{13}\text{C}$ . Given this model, one can envision mechanisms by which all of the environmental variables discussed in this section may directly or indirectly influence plant  $\delta^{13}\text{C}$ . For other factors which apparently may affect plant  $\delta^{13}\text{C}$ , such as nutrient levels (Bender and Berge, 1979), the mechanisms may be less straight-forward.

There remains the possibility that the seasonal plant  $\delta^{13}\text{C}$  signature is physiologically rather than environmentally motivated. In tropical woods a physiologically-controlled isotopic signal might have a greater seasonal amplitude than a temperature-controlled isotopic signature dependent only on relatively small variations in temperature and/or sunlight intensity. Part of our proposed research is designed to distinguish between physiologic and environment controls on these signatures.

## B. Evidence for Seasonal $\delta^{13}\text{C}$ Variation

### 1. Canadian leaves

Several studies with leaves and tree rings have provided evidence of variability of the stable-carbon isotopic composition of plants during the growing season. Lowden and Dyck (1974) analyzed  $\delta^{13}\text{C}$  of grass and maple leaves in Canada, collected at various times during several growing seasons. They analyzed whole-tissue which shows a decrease in  $\delta^{13}\text{C}$  as the growing season progresses, typically about 2-3‰ (Figure 1). They postulated that this change is due to seasonal changes in  $\delta^{13}\text{C}$  of atmospheric  $\text{CO}_2$ . However, as the growing season advances,  $\delta^{13}\text{C}$  of the atmosphere should actually increase so the  $\delta^{13}\text{C}$  of plants might be expected to increase as well.

## 2. New Zealand Monterey pine

Wilson and Grinsted (1977) examined the  $\delta^{13}\text{C}$  changes in the cellulose and lignin components of two large rings (~2 cm) of Pinus radiata (Monterey pine) in a part of New Zealand where their growth is nearly year-round. Each ring was subdivided into six segments, yielding similar annual trends of a ~1.5‰  $\delta^{13}\text{C}$  increase at the beginning of the growing season and a rapid ~1.5‰ decrease at the end (Fig. 2). Because this fluctuation is much greater than the annual fluctuation of  $\delta^{13}\text{C}$  of atmospheric  $\text{CO}_2$  and because of abundant (uniform) rainfall at the site, they considered the  $\delta^{13}\text{C}$  variation to be temperature-related with a coefficient of +0.2‰/°C.

## 3. European earlywood and latewood

Freyer (1980) examined  $\delta^{13}\text{C}$  of cellulose from the earlywood and latewood of 50 individual tree rings (unspecified species). In the mean,  $\delta^{13}\text{C}$  of the earlywood was isotopically lighter than the latewood.

## 4. Arizona juniper tree rings and leaves

We examined the  $\delta^{13}\text{C}$  variation within the 1978 and 1979 ring of a Juniperus deppeana (alligator juniper) individual which grew near Prescott, Arizona. The carbon isotopic pattern of both whole wood and the cellulose component is one of increasing  $\delta^{13}\text{C}$  over the growing season (Fig. 3). The whole-wood values are isotopically lighter than cellulose, presumably due to the presence of lignins which tend to be relatively light (Park and Epstein, 1961). An important feature of the pattern in the 1979 ring is that it was unperturbed by the presence of a diffuse latewood band within the ring. Such latewood bands when fully developed in the middle of a growing season may give the appearance of an extra (false) ring. Although a positive temperature response (+0.13‰/°C) was estimated for the early half of the ring, the continued increase of  $\delta^{13}\text{C}$  after the diffuse latewood deposition (representing

times when Prescott mean monthly temperatures had leveled off), suggests independence of  $\delta^{13}\text{C}$  and temperature.

We also measured the  $\delta^{13}\text{C}$  of the 1981 growth (wood and leaves) from a Juniperus monosperma in Tucson, Arizona. Representative samples of the incremental growth of the leaves were taken at approximately monthly intervals. At the end of the growing season, cores of the 1981 ring were taken. Wood and leaves yield similar  $\delta^{13}\text{C}$  patterns for both whole-tissue and cellulose (Fig. 4). The  $\delta^{13}\text{C}$  values decrease and then more slowly increase at the end of the season, a trend the reverse of that seen in the J. deppeana and P. radiata samples previously described. The timing of  $\delta^{13}\text{C}$  change in the leaves and wood is nearly identical, although a persistent offset of  $\sim 2.5\text{--}3.5\%$  suggests fractionation of carbon between leaves and wood. Because this tree received regular irrigation, water stress was not considered a causal factor and a significant ( $r^2 = 0.86$ ) temperature coefficient of  $-0.27\%/^{\circ}\text{C}$  was computed.

#### 5. Other studies

We have made several additional measurements on Pinus ponderosa (ponderosa pine) and Pseudotsuga menziesii (Douglas fir) which have revealed promising results (Figs. 5-8). All seem to follow the pattern of Wilson and Grinsted's P. radiata with increasing  $\delta^{13}\text{C}$  at the beginning of the growing season followed by a  $\delta^{13}\text{C}$  decrease at the end. The reverse trend of the Tucson J. monosperma thus stands out as somewhat anomalous. However at 2500' (mean summer Tucson temperatures about  $30^{\circ}\text{C}$ ) it is by far the hottest site of any reviewed here. Furthermore, water availability at the Tucson site (irrigation) was almost certainly greater throughout the growing season than that seen at the P. ponderosa and P. menziesii sites. An additional possible explanation for the apparent uniqueness of the J. monosperma is that it grew within the city of Tucson, and although over 200 m from any major highways,

there could be some local pollution effect. The negative temperature coefficient for the J. monosperma, however, is quite similar in magnitude to that found in several in vitro studies with the carbon-fixing enzyme RuBP-C (Christeller et. al., 1976; Estep et. al., 1978).

The set of measurements from P. ponderosa and P. menziesii represent the same years so that isotopic signatures from (a) the same species at the same site, (b) the same species at different sites, and (c) different species at the same site may be compared. The Hitchcock and Radio Ridge Douglas firs (Figs. 5, 6) are both from the Santa Catalina Mountains near Tucson, Arizona, separated by several kilometers. The Hitchcock signatures have very low amplitude relative to the Radio Ridge rings. At a lower elevation, however, the Hitchcock site is warm and dry relative to Radio Ridge, which may contribute to the difference. The Radio Ridge and Gila Douglas firs (Figs. 6, 7) are separated by several hundred kilometers, yet both show high-amplitude signatures. In fact, in both trees the 1909 amplitude is greater than that of the 1908 isotopic signature. Furthermore, in both trees the initial isotopic rise is over a shorter distance in the growth direction than is the  $\delta^{13}\text{C}$  fall at the end.

The amplitude of the isotopic signatures of the Gila ponderosa pine is lower than those of the Gila Douglas fir, which could be a species effect. However, the asymmetry of both sets of signatures is quite similar so that the proximity of sites and elevations suggests some common environmental influence on both of the trees, but with different coefficients.

Lastly, a cross-section of wood from Prosopis juliflora (mesquite) which was growing near the San Pedro River in southern Arizona was tested. This sample was interesting in that it showed no clear ring structure demarcating annual growth. Equal subdivisions were made on an inner portion of the

mesquite section which appeared to show faint evidence for two rings. The isotopic results in Fig. 9, however, suggest three rings may actually be represented.

#### C. Probable Limitations of Seasonal $\delta^{13}\text{C}$ Variation

Despite the minor amount of research which has been done with these isotope signatures, there are some recognized limitations to dating. Chief among these is that the method could probably not be used to locate missing rings or to date wood which contains missing rings. For wood samples from the humid tropics, such missing rings may not be a problem. Furthermore, to assign absolute dates to a sequence it will probably be necessary to know the age of one ring (perhaps the outside), unless, as in standard dendrochronology, there are peculiar "index" years which are very characteristic. This latter possibility as well as technical limitations are discussed further in the next section.

#### D. Radiocarbon and Recent Tree Growth Rates

In addition to the stable carbon isotopes,  $^{13}\text{C}$  and  $^{12}\text{C}$ , the radioisotope  $^{14}\text{C}$  exists and is taken up by all living matter. There are natural fluctuations of radiocarbon in the atmosphere, but in the post-1950 period there has been a large, artificial  $^{14}\text{C}$  "pulse" from atmospheric hydrogen bomb testing. The curve for this input of excess radiocarbon and its subsequent decay is well-defined from high-precision measurements, and is depicted in Figure 10.

Because trees will sample atmospheric  $\text{CO}_2$  they will acquire this radiocarbon "signature" within the wood laid down in the post-bomb era. Stuiver et al. (1981) have suggested that by sampling as few as 2 outer adjacent segments of a tree radius whose cutting date is known, their position in time will be uniquely defined on the  $^{14}\text{C}$  curve (Figure 11) provided growth rate over this period has been approximately constant. Given this age of wood together with

the length of these segments, a growth rate may be determined for the last 20-30 years if the tree was cored or harvested within the past few years.

It may thus be possible to get an integrated growth rate estimate for a period of decades with just a couple of radiocarbon measurements, as opposed to standard methods of circumference measurements for 5-10 years to obtain a growth rate. With this method one could go into virtually any area and make an estimate of recent growth rates without such continuous monitoring. A key to the practicality of this technique will be the use of the Tandem Accelerator Mass Spectrometer (TAMS) which allows determination of radiocarbon concentrations in very small samples. With this technique, non-destructive coring would provide sufficient wood material for the growth rate determinations.

### III. PROPOSED RESEARCH

#### A. General Plan for $\delta^{13}\text{C}$ Isotopic Signatures

Our research plan with the seasonal isotopic variation involves (1) further testing to determine how widespread this annual isotopic signature is in trees, (2) determining the extent to which these signatures may be valuable for dating of woods, and (3) resolving the environmental or physiological cause(s) of these signatures. In many cases, dendrochronologically dated (temperate) wood materials will be required to resolve some of the related fundamental questions because analysis of wood associated with a specific year is assured. Tropical wood will be tested for aging suitability throughout the project.

We are fortunate to be associated closely with the internationally renowned Laboratory of Tree-Ring Research at the University of Arizona. The Tree-Ring Lab contains a large repository of wood samples of various ages from

all over the world. In pursuing this proposed research, we expect to continue our already established close cooperation with several members of the staff of the Laboratory of Tree-Ring Research, especially Prof. H. C. Fritts and Prof. V. C. LaMarche (specialists in tree growth and dendroclimatology), Prof. M A. Stokes and Mr. T. P. Harlan (dendrochronology), and Prof. J. Dean and Mr. D. Bowden (anthropology-archaeology). We will also work closely with Dr. Ariel Lugo of the Institute of Tropical Forestry (Puerto Rico) for appropriate tropical samples and applications.

#### B. Universality of $\delta^{13}\text{C}$ Isotopic Signatures

All the wood (and leaf) materials examined thus far (see Background Section) have shown some type of seasonal isotopic variation. We now propose to continue examining these signatures by testing (1) temperate species such as those most commonly used in dendrochronology (bristlecone pine, pinyon pine, white fir, sequoia, oak (Europe), and others), and (2) tropical species which may have several growth flushes or which may show no ring structure. Similar temperate species such as Prosopis (mesquite) and evergreen Quercus (oak) will also be examined. This will also allow us to determine any inherent differences between hardwoods and softwoods, and temperate and tropical trees. Furthermore, in this stage of research we will be able to test how the number of subdivisions are related to defining detail of these isotopic signatures.

The natural variability of these signatures must also be examined with regard to tree growth, and, at this stage, it is important that this be done with ~~datable~~ temperate species where age is constrained. For instance, the signatures of one or more rings must be measured along different radii of a section. Studies have shown a radial variability of  $\delta^{13}\text{C}$  for whole individual-ring (or group of rings) sequences (Mazany et. al., 1980; Tans and

Mook, 1980). We already have available to us an oriented section from a ponderosa pine from the Santa Catalina Mountains for this purpose.

Another question we will answer is whether there is any "growth effect" which may influence the character (e.g., amplitude) of these signatures. Ring widths progressively decrease in thickness from the inner, early years to the outer growth years, so that the seasonal signatures may also show some age dependence. At the same time, signatures within the heartwood and sapwood may be compared. Furthermore, by collecting wood from an old tree and a young tree at the same site, we will be able to compare the signatures of the same year when they occur as small rings on the outside of the old tree but as relatively large rings on the outside of the young tree.

#### C. Isotopic Signatures and Dating

Regardless of whether the cause of these signatures is physiologic or environmental, their ubiquity would offer a powerful dating tool. For two datable (temperate) trees from the same site we would develop a long, continuous record of isotopic signatures of at least 20 years in length. From this we will be able to judge just how similar this seasonal variation is from one tree to the next. Furthermore, this would allow us to check for unusual or anomalous "index" signatures which could aid in dating. Such distinctive signatures would be equivalent to the importance of narrow rings in standard dendrochronology, and might eventually allow absolute dating of wood by the character of isotopic signatures.

Additional tests will be made on false rings, preferably on dated woods where the false rings have been identified. We should be able to obtain wood materials where growth has essentially stopped in mid-season but resumed after a latewood band with a sharp terminal boundary has been deposited. These may be rings within which more than one such latewood band has developed, as is apparently common among some tropical woods.



We will also examine some of the temperate species which have been difficult to date in the past, such as Populus (cottonwood), Cupressus (cypress), evergreen Quercus, Prosopis and/or other species which may have useful applications. Most importantly, we have been in contact Dr. Ariel Lugo of the Institute of Tropical Forestry in Puerto Rico in regard to providing tropical wood materials for dating. He has offered tropical wood of several species from locations where there are stands of known age. A recent conference on the dating of tropical woods (Bormann and Berlyn, 1981) concluded that of the radioisotope methods only radiocarbon measurement methods might be useful for this dating application. With this set of tropical woods, however, we will be able to determine the applicability and precision of our stable-carbon aging method for these woods. For other tropical samples whose cutting date is known but whose overall age is unknown, we will use accelerator radiocarbon analysis in order to locate the distinctively "tagged" wood representing the 1950's and 1960's (see Section IIIA). We will then test the correspondence of the isotopic signatures between the outside and this radiocarbon dated period for precision of our analysis.

#### D. Source of $\delta^{13}\text{C}$ Isotopic Signatures

We plan to perform several experiments to illuminate the cause of the seasonal isotopic variation. To determine possible effects of climate we will look at rings of the same age from the same species at a variety of sites with different climates (i.e., different latitudes, different elevations). This will be an expansion of some of the tests described in the Background Section.

We will look at the isotopic signatures with regard to cell size and cell-wall thickness. Furthermore, because active research with x-ray densitometry of wood is being conducted in the Tree-Ring Lab, we may compare wood density with the isotopic variation.

A class of experiments that is critical to our understanding of the variables responsible for  $\delta^{13}\text{C}$  changes within rings is controlled growth testing. However, growth chamber experiments on seedlings may not be relevant to responses in mature trees and controlled growth may be prohibitively expensive. During the first year of this study, we will investigate other possibilities. We will continue consultation with Dr. Boyd Strain of Duke University regarding experimental details. One possibility may be to work with certain dwarf species old enough to have passed the "juvenile" stage, yet small enough to manage in a laboratory environment. During the second year, we expect to begin small-scale growth-chamber experiments, in which we can vary temperature, water-stress, and possibly other variables.

In addition to these growth-chamber type experiments, it would be useful to perform a field experiment wherein we could monitor carefully temperature, precipitation, soil moisture and solar radiation during several growing seasons and then measure the isotopic signatures of the growth rings at the site. We are fortunate, however, that such monitoring of a site has already been done in the Santa Catalina Mountains by Prof. Hal Fritts, Brown (1968) and Budelsky (1969) and extensively illustrated in Fritts (1976). Despite the fact that this monitoring was done back during the period 1963-1967, the wood from trees of this site is, of course, still available to us. We therefore plan to analyze the isotopic signatures in light of the environmental data of the site.

#### E. Experimental Improvements Involving $\delta^{13}\text{C}$ Signatures

Working with subdivisions of an entity which may already be quite small presents practical problems. The separation of rings done in the past has been accomplished with a razor knife under a binocular microscope. We will explore possibilities for improving this process, such as perhaps through the use of a microtome slicer.

Homogeneity of these small samples must also be ensured. If the material from a subdivision contains more carbon mass than we would routinely analyze, we must further reduce it to a usable portion. Inhomogeneities in these subdivisions (related to the seasonal fluctuation) require special care to ensure that the portions analyzed are representative of the whole subdivision. Some of the points in Figure 5 which do not conform to a smooth trend may be the result of such biasing. We will seek a method such as micro-milling or careful trimming to overcome inhomogeneities.

Finally, if the results of this project reveal an important dating tool, new technical methods may be required to rapidly process these samples. Results thus far seem to indicate the same signal is contained in the whole wood as in the cellulose. If so, then advanced technologies such as laser methods may be used to accurately and rapidly convert samples to CO<sub>2</sub> for mass spectrometric analysis. Currently, in addition to the time required to manually separate the subdivisions it takes 15 to 20 minutes to convert the samples to CO<sub>2</sub> and another 20 minutes for analysis.

#### F. Radiocarbon Pattern and Growth Rates

We plan to use the TAMS to determine the radiocarbon concentrations in adjacent segments of wood from the outer portions of cores or cross-sections. Comparing these concentrations with the atmospheric <sup>14</sup>C pattern of the post-1950 period will provide unique solutions for the wood age. Once age is established, average growth rate for this period is easily calculated.

We will first test this method on temperate trees with datable rings to verify that the precision of analysis and data reduction is sufficient for accurate results. Once established, we will proceed to tropical trees. With trees of known age, we may test for uniformity of growth by determining the growth rate for the post-bomb period and comparing it to the growth rate

inferred for the inner part of the tree. Where the tree age is unknown, we may use the stable carbon isotopic signatures to assist in determining the nature of changes in growth rate.

Ideally, we could then proceed to selected stands of trees to determine growth rates over the most recent 2 or 3 decades. Such determinations may be important to testing for "fertilization" effects from atmospheric CO<sub>2</sub> increases in natural undisturbed forest stands.

#### IV. EXPECTED IMPACT OF PROPOSED RESEARCH

On the one hand, the results of this research will bridge several fields and potentially provide a wealth of information to dendrochronologists, tropical ecologists, plant physiologists, forest managers and carbon-cycle researchers. On the other, however, the broad scope of this research implies it will not fit neatly into the standard compartments of conventional funding sources. The potential yield of information for any of several different fields individually **may be sufficient** to make this project viable.

To dendrochronology this will be an additional new method to aid in dating wood. At the very least, these isotopic signatures may help in identifying false rings. This can be applied to modern studies with such woods as evergreen oaks and acacias, or to studies of ancient wood as in the extension of the bristlecone pine chronology back in time. Furthermore, if certain years have very characteristic seasonal  $\delta^{13}\text{C}$  signatures, then these may provide a new type of "index" year to aid in dating wood with rings which is not datable for some reason (e.g., missing rings or complacency). Finally, this method may open the door to dating the class of woods without ring structure.

The element of time is also important to forest managers and tropical ecologists. Effective management often entails a knowledge of the age-size distribution of certain stands. With accurate aging, the ecologist can answer questions on forest succession and growth strategies of individual species or classes of species. Up until now, growth rate measurement over a few years and ring-counting where possible have been the keys to age estimation. With the seasonal isotopic signatures there is the potential for high accuracy age determination. This technique may be applied directly to aging problems, or it may be used to test the accuracy of the other methods of age and growth rate determination. Radiocarbon measurements with the TAMS may provide a rapid growth rate estimate for the last 20-30 years.

The source of the seasonal isotopic signatures may be motivated by some **combination of internal** and external factors. These isotopic changes may thus be linked to certain aspects of plant physiology and could be a measure of seasonal physiologic changes in activity, e.g., photosynthesis or respiration or transpiration. The character of the seasonal variation may also have some taxonomic implications.

One of the important questions in carbon-cycle studies is the status of the biosphere as net carbon source or sink. With a rapid measure of growth rates such as that proposed here with radiocarbon, post-bomb growth rates may be readily calculated for a tropical forest stand. If growth rates had previously been measured in the same stand 50-100 years ago, the results may be compared to test for a "fertilization" effect from increasing atmospheric CO<sub>2</sub>. Age and rate determination are also a key to estimating how fast tropical forests reestablish (and sequester carbon) after a disturbance such as clearing for agriculture.

Table 1. Experimentally-derived temperature coefficients for  $^{13}\text{C}$  fractionation in plants.

A. in vitro

<u>Enzyme (source)</u>	<u>Temperature Coefficient</u>	<u>Source</u>
RuBP-C (sorghum)	+1.2‰ °C <sup>-1</sup>	Whelan, Sackett and Benedict (1973)
RuBP-C (sorghum)	-0.22	Christeller, Laing and Troughton (1976)
RuBP-C (algae and spinach)	-0.2 to -0.6	Estep et al. (1978)
PEP-C (wheat and maize)	+0.03	Schmidt et al. (1978)

B. in vivo

<u>Species</u>	<u>Material</u>	<u>Temperature Coefficient</u>	<u>Source</u>
Xanthium strumarium	leaf	-0.1‰ °C <sup>-1</sup> (for 13-18°C)	Smith, Oliver and McMillan (1976)
pea, rape, barley	whole plant	-0.13 to -0.29 (non-linear)	Smith, Herath and Chase (1973)
wheat	leaf	-0.4	Schmidt et al. (1978)
15 species	whole plant	-0.01 to -0.13	Troughton and Card (1975)
marine plankton	whole cell	+0.23	Sackett et al. (1965)
phytoplankton	whole cell	+0.35	Degens et al. (1968)
timothy	whole plant cellulose	-0.15 -0.03	Bender and Berge (1979)

## FIGURE CAPTIONS

- Fig. 1. Seasonal  $\delta^{13}\text{C}$  variation in whole tissue of maple leaves and grass from Quebec, Canada (from Lowden and Dyck, 1974).
- Fig. 2. Intra-annual  $\delta^{13}\text{C}$  variation in lignin (open circles) and cellulose (solid circles) within tree rings of Pinus radiata from New Zealand (from Wilson and Grinsted, 1977).
- Fig. 3. Intra-annual  $\delta^{13}\text{C}$  variation in whole wood (solid squares) and cellulose (open squares) from rings of Juniperus deppeana from Prescott, Arizona. A diffuse, false latewood band occurred in subdivision III of the 1979 ring (from Leavitt and Long, 1982b).
- Fig. 4. Seasonal  $\delta^{13}\text{C}$  variation within the 1981 leaves and ring for (a) whole tissue and (b) cellulose from a Juniperus monosperma growing in Tucson, Arizona (from Leavitt and Long, 1982a).
- Fig. 5. Intra-annual  $\delta^{13}\text{C}$  variation for the 1908 and 1909 growth rings from a Pseudotsuga menziessi from the Santa Catalina Mts. (Radio Ridge site), Arizona. (LW = latewood).
- Fig. 6. Intra-annual  $\delta^{13}\text{C}$  variation for the 1908 and 1909 rings from a Pseudotsuga menziessi from the Santa Catalina Mts. (Hitchcock site), Arizona.
- Fig. 7. Intra-annual  $\delta^{13}\text{C}$  variation for the 1908 and 1909 rings from a Pseudotsuga menziessi from the Gila National Forest, New Mexico.
- Fig. 8. Intra-annual  $\delta^{13}\text{C}$  variation for the 1908 and 1909 rings from a Pinus ponderosa from the Gila National Forest, New Mexico.
- Fig. 9. The  $\delta^{13}\text{C}$  variation within a 7.4 mm segment of Prosopis juliflora which showed no sharply-defined ring structure (from the San Pedro River Valley, Arizona).
- Fig. 10. Variation of  $^{14}\text{C}$  in the atmosphere (from Stuiver et al., 1981)
- Fig. 11. Illustration of how radiocarbon activities of 2 samples could uniquely define the position in time on the atmospheric  $^{14}\text{C}$  curve (from Stuiver et al., 1981).

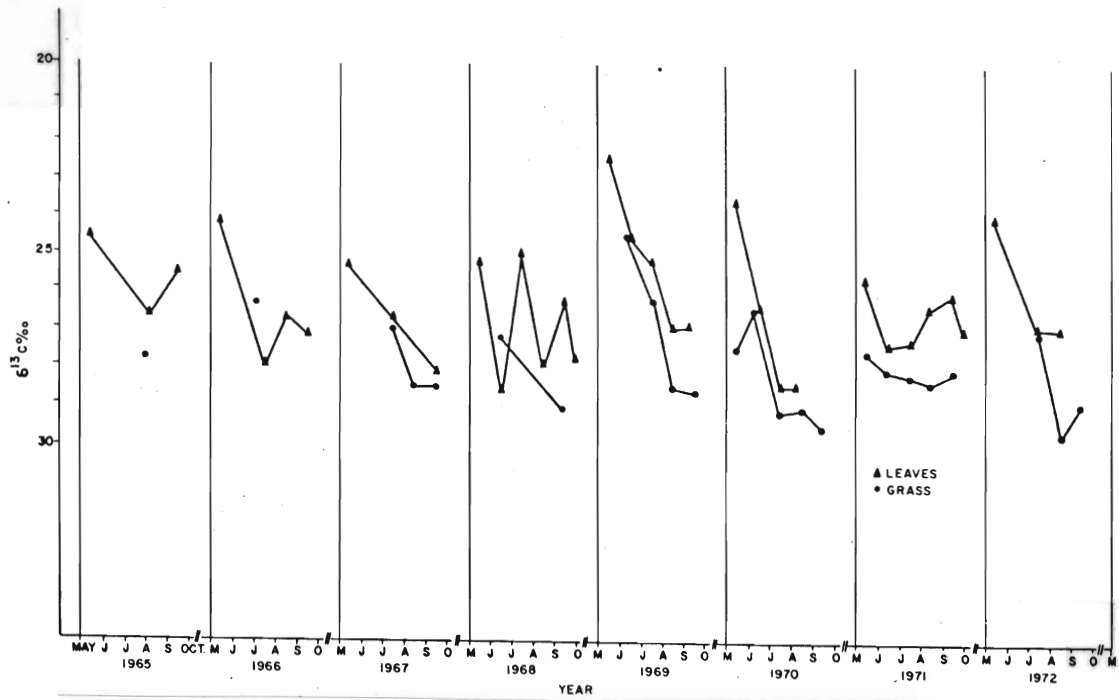


Fig. 1

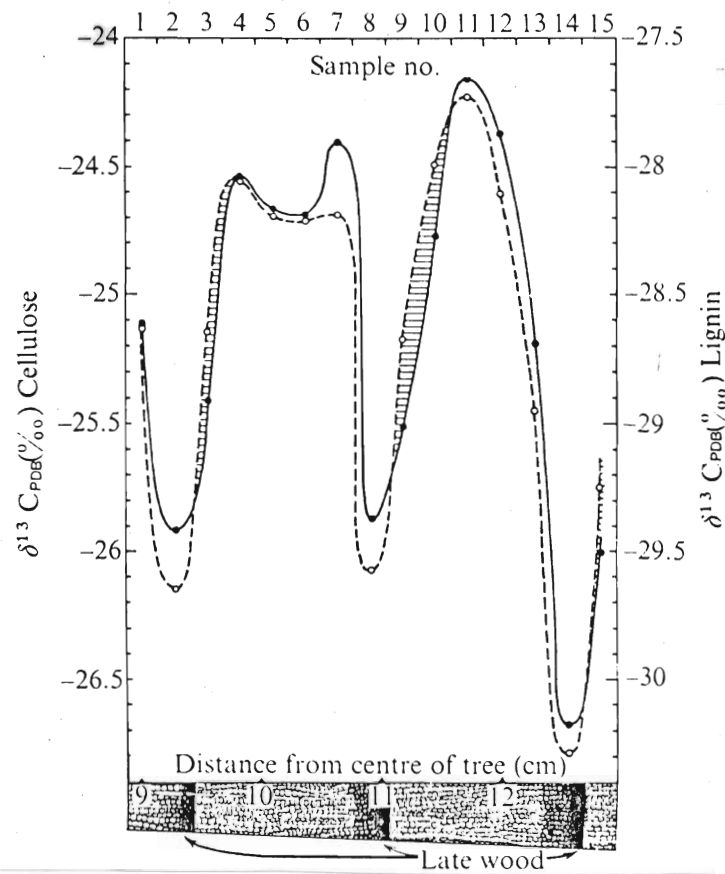


Fig. 2



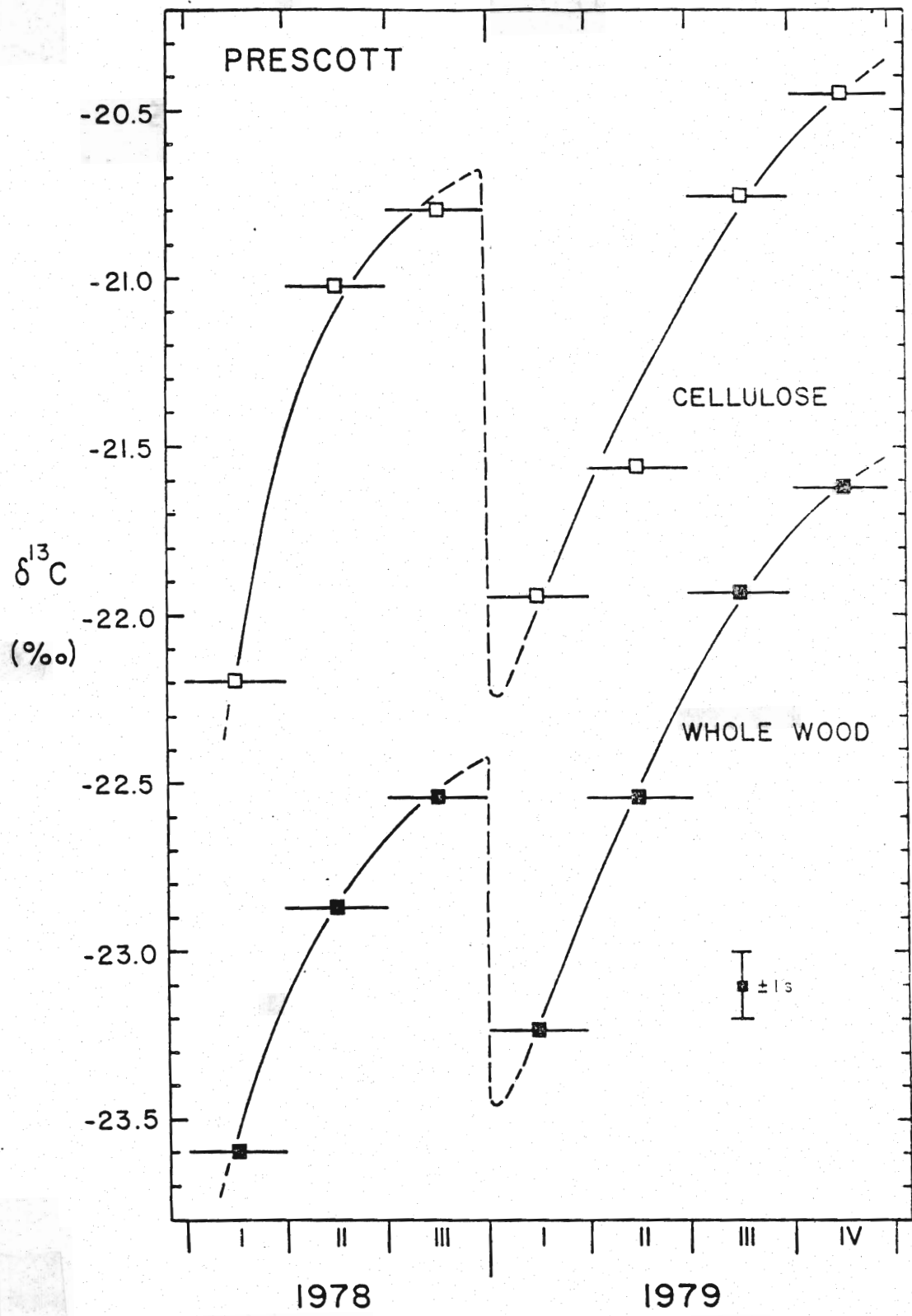


Fig. 3

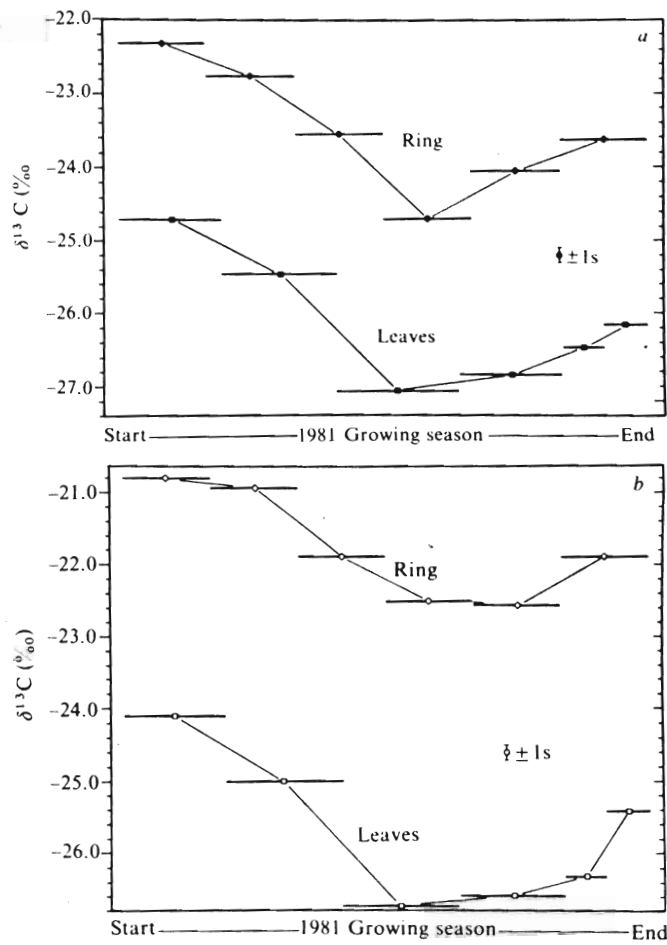


Fig. 4

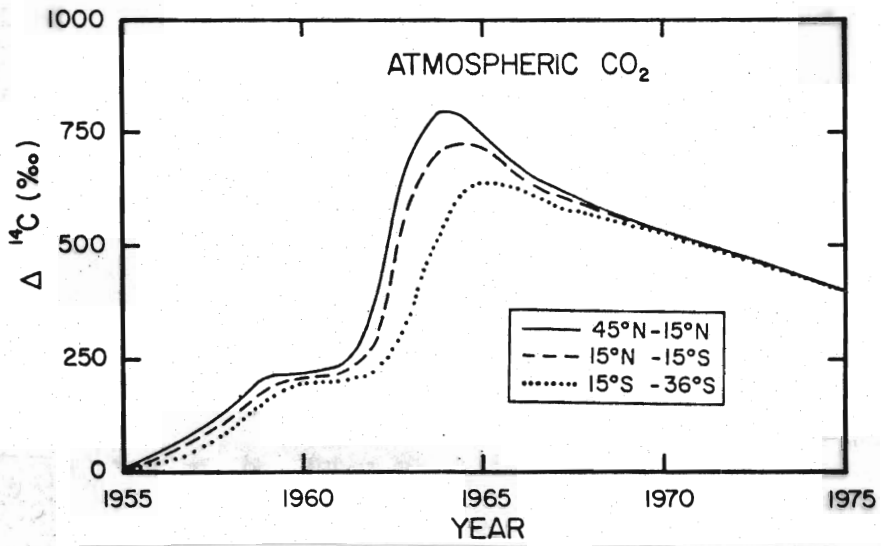


Fig. 10

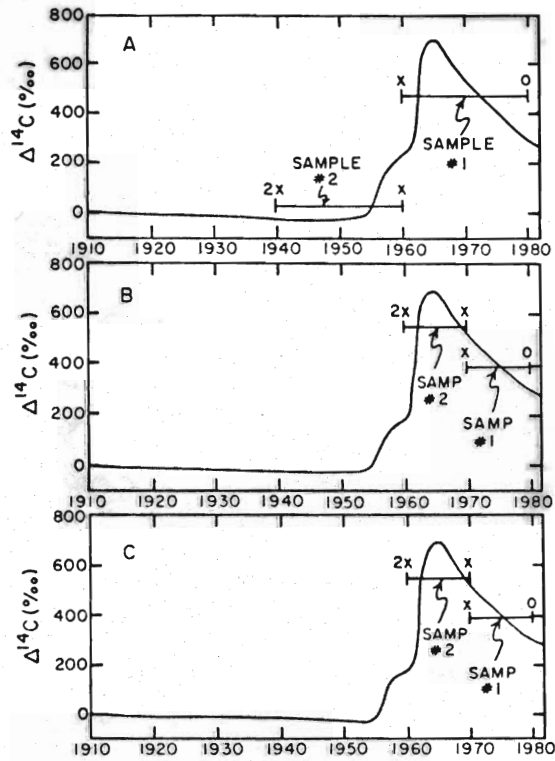


Figure 3 As shown in this diagram, if two depth-integrated samples are analyzed for radiocarbon, as long as the growth rate of the tree has been reasonably uniform from year to year over the sampling interval, the results will uniquely define the mean growth rate of the tree. If the samples are of identical width, a graphical solution to the deconvolution could be constructed so that the growth rate could be obtained from the  $\Delta^{14}\text{C}$  values for the two samples. As can be seen by comparing the acrotyes for the first sample in cases (B and C), a single measurement does not provide a unique answer. The  $\Delta^{14}\text{C}$  curve shown is that for the tropical atmosphere (Nydal et al., 1979).

Fig. 11

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