Proceedings—Research on Coniferous Forest Ecosystems—A symposium.
Bellingham, Washington—March 23-24, 1972

1891

Estimation of biomass and transpiration in coniferous forests using tritiated water

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Abstract

Nondestructive measurement of biomass and transpiration rates in trees is dependent on a new application of the established theory of tracer dynamics in steady state systems. The method utilizes HTO as a tracer for H_2O *in the plant. Both transpiration and biomass measurements require experimental determination of tritium activity in the tree as a function of time. Biomass measurement requires one additional parameter—the mean residence time of water in the plant. In this paper we examine various theoretical and experimental alternatives for determining mean residence times. Data supporting one alternative is presented. The measurement of conducting tissue biomass is also discussed.*

Introduction

Kinetic theory and tracer distribution methods have been used extensively in measurement of the properties of flowing systems both in biology and engineering. The theoretical aspects and the underlying assumptions governing the practical application of tracer dynamics have been thoroughly discussed by authors in many fields. Reports by Zierler (1964), Bergner (1961, 1964a, 1964b, 1965, 1966), and Ljunggren (1967) have described the use of tracer dynamics and kinetic theory in the nondestructive measurement of flow rates, mean residence times, and compartmental volumes of steady state biological and engineering systems.

It is the purpose of this paper to present the state of the art in the application of the theory and tracer methods to the nondestructive measurement of tree biomass and transpiration, and to examine the experimental parameters that can be measured as well as the underlying theoretical assumptions on which they are based.

The nondestructive measurement of transpiration rates and biomass in trees utilizes tritiated water (HTO) as a tracer for water. Tritiated water is added to the tree water pool by injection into the trunk near ground level. The fate of the HTO tracer-labeled water in a tree is followed by monitoring tritium activity, as a function of time, in foliage and small branches. Both transpiration and biomass measurements require experimental determination of tritium concentration-time curves. Biomass requires additionally the mean residence time of water in the tree and the mean moisture content of the tree.

Theoretical Discussion

Transpiration Measurement

Application of the theory of tracer dynamics to the problem of measuring transpiration rates in plants has been previously demonstrated[1] [2] (Kline et al. 1970). The theory itself has been discussed extensively by Bergner (1961, 1964a, 1964b, 1965, 1966), Zierler (1964), Ljunggren (1967), and Orr and Gillespie (1964). Transpiration measurements depend upon use of the Stewart-Hamilton equation shown by equation 1:

$$M = F \int_0^\infty f(t)\,dt \quad , \qquad (1)$$

where M = total activity of tritium initially injected (disintegrations per minute, DPM),

F = the flow rate of water through the tree (ml/hour x tree),

f(t) = activity distribution of tritium at the points of exit from the system (DPM/ml), and

t = time (hr).

Equation 1 states simply that the product of the flow rate (F) and the total integral of the curve of activity versus time (fig. 1) is equal to the total activity of the tracer which was originally injected. In practice the activity-time curve is measured experimentally and the total activity injected is fixed by the experimenter. The flow rate (F) is the only unknown quantity in equation 1 and is solved algebraically. The value of F is the daily flow rate which is averaged over daytime and nighttime flows. The average is taken over the full time interval of residence of tritium in the tree. Shorter term resolution of transpiration is not possible with this method. Examples of transpiration rates which have been obtained using equation 1 with tritiated water as the tracer are given in table 1 for field-grown coniferous trees.

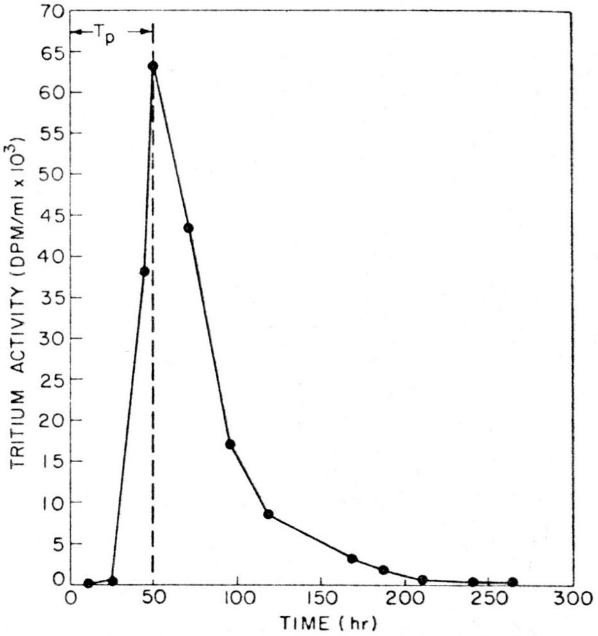

Figure 1. Typical activity-time response curve obtained by injecting a jack pine *(Pinus banksiana)* tree with tritiated water and sampling twigs as a function of time for tritium content. Peak arrival time is indicated by T_p.

Biomass Measurement

The measurement of tree biomass is based on that part of the theory of tracer dynamics which permits calculation of the pool size of the compartment through which flow takes place. In trees the pool refers simply to the average total water content of the tree. When pool size has been computed, it is a simple matter to convert to biomass using the average moisture percentage of the wood.

Derivation of an expression which permits computation of compartmental pool size is given by Zierler (1964). Zierler's expression is given by equation 2:

$$C = F \cdot T_m \quad , \qquad (2)$$

where C = compartment volume (ml),

F = flow rate through the compartment (ml/hr), and

T_m = mean residence time of the flowing substance (hr).

Equation 2 states simply that the compartment pool size is given by the product of the

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[1] J. R. Kline, C. F. Jordan, and R. C. Rose. Transpiration measurements in pines using tritiated water as a tracer. *In* D. J. Nelson (ed.), Third National Symposium on Radioecology, Proc., May 10-12, 1971, Oak Ridge, Tenn. (In press.)

[2] J. R. Kline, M. L. Stewart, C. F. Jordan, and Patricia Kovac. Use of tritiated water for determination of plant transpiration and biomass under field conditions. *In* Symposium on the Use of Isotopes and Radiation in Soil-Plant Research Including Applications in Forestry, Proc. Int. At. Energy Agency Conf. SM-151, December 13-17, 1971, Vienna, Austria. (In press.)

Table 1.—Transpiration rates, mean residence times, computed biomasses, and observed biomasses for field-grown red and jack pine trees

Red pine *(Pinus resinosa)* (1970)

Tree number	Dbh (CM)	Transpiration rate (F) (ml/hr)	Mean residence time (T_p) (hr)	Computed dry biomass (kg)	Observed dry biomass (kg)
3	14.1	1170	50	42.4	42.6
4	9.5	222	110	17.7	12.2
5	12.1	935	42	28.4	26.3
6	13.3	1280	42	38.9	35.1
7	12.3	725	49	25.7	26.7
8	8.2	413	49	14.6	12.1
10	12.3	254	98	18.0	20.6
	Mean tree weight ± SE			26.5 ± 4.1	25.1 ± 4.3
	Mean forest biomass ± SE (kg/ha)			7.7 x 10^4 ± 1.2	7.3 x 10^4 ± 1.2

Jack pine *(Pinus banksiana)* (1971)

Tree number	Dbh (CM)	Transpiration rate (F) (ml/hr)	Mean residence time (T_p) (hr)	Computed dry biomass (kg)	Observed dry biomass (kg)
1	10.5	807	49	38.5	19.9
2	12.0	833	52	33.5	26.7
3	10.0	540	48	21.5	19.9
4	10.0	941	27	25.6	32.0
5	8.9	570	45	25.4	18.1
6	11.0	833	25	19.2	26.0
7	11.0	609	23	17.0	32.3
8	9.5	506	46	24.6	24.6
9	10.4	492	27	18.3	20.9
11	11.4	1083	26	28.0	28.5
12	9.4	903	24	27.2	20.0
	Mean tree weight ± SE			23.3 ± 2.0	24.4 ± 1.5
	Mean forest biomass ± SE (kg/ha)			6.3 x 10^4 ± 0.5	6.1 x 10^4 ± 0.4

flow rate and the mean residence time of the system.

In plants the pool size (C) is also given by the difference between wet and dry weight (W-D) of the plant. Moisture fraction is conventionally calculated by equation 3:

$$\frac{W-D}{W} = f \; , \qquad (3)$$

where W = wet weight of sample (gm),

D = dry weight of sample (gm), and

f = fractional moisture content.

Equation 3 holds equally well for the case where the determination is done on the entire plant or for a representative subsample of the plant. In the case where the entire plant is the sample, W-D can be substituted for C in equation 1 resulting in equation 4, which is an expression for the moist weight of the plant:

$$W = \frac{1}{f} F \cdot T_m \; . \qquad (4)$$

Moist weight can be converted to dry weight using equation 3. This results in equation 5 which is an expression for dry biomass of the plant:

$$D = \frac{1-f}{f} F \cdot T_m \; . \qquad (5)$$

Equation 5 requires the experimental determination of f, F, and T_m for its solution. In the absence of a feasible method for determining f on the entire tree, it is necessary to measure it on subsamples. Ideally the subsamples should be weighted for different tree parts such as trunk, branches, and leaves. Since there is usually no method available for measuring weighting factors, we have followed the practice of estimating moisture content of the tree trunk since this represents the greater portion of the biomass of the tree. In our experience, moisture content of plant parts has not been greatly different from one another and no serious errors are introduced by following this procedure. If plants are found where unweighted estimates of f differ from the true weighted value, then this could be a significant source of error in the estimate of biomass.

The value of F in equation 5 is the mean flow rate which has previously been calculated using equation 1. This means that the reliability of the estimate of biomass can be no better in general than the reliability of flow or transpiration rate. Biomass estimates are normally expected to have lower statistical precision than transpiration estimates since they require the use of additional measured parameters.

The most difficult parameter to measure in equation 5 is T_m, the mean residence time. There appear to be at least three possible approaches to obtain this quantity: (1) measure the slope of the declining branch of the activity-time curve; (2) compute the first moment of the curve; or (3) measure the transit time between the point of injection and the point of exit of the tracer from the system (Donato et al. 1964).

The slope method for mean residence time would be valid in the case where the plant is labeled to equilibrium with the tracer. If all of the water molecules of the trees were labeled equally with HTO, then the rate at which tritium activity declines in the tree would be proportional to the amount of tritium present. Such systems are described by an equation of the form $A = A_0 e^{-\lambda t}$ where A_0 is initial activity, A is activity at time t and λ is the rate constant of loss. The term λ is the slope when data described by this relationship are plotted on semilogarithmic coordinates. The mean residence time for such a relationship is simply the reciprocal of λ ($T_m = \frac{1}{\lambda}$). This is a frequently used relationship for obtaining T_m although in practice it is sometimes used without verification of the assumptions. In some systems it is possible to label to equilibrium by injecting the tracer into the system continuously; however, in large trees this would entail larger than desirable releases of radioactivity to the environment. Therefore, it is preferable to label by the instantaneous pulse method. When the pulse method of labeling is used, the injected material moves upward in the tree while retaining its pulse shape. The pulse may undergo considerable broadening but the system cannot be assumed to have achieved uniform labeling. In this case the slopes of activity-time curves have two components, one reflecting the turnover rate of water and the other reflecting the pulse shape. There is no method currently available for resolving these components in an activity-time curve, and therefore the slope of the curve cannot validly be used to compute T_m.

Ljunggren (1967) has described the computation of mean residence times for flowing systems using the first moment of the activity-time curve. The first moment is the centroidal axis of the activity-time distribution—that vertical axis which divides the distribution into two parts having equal areas. Equation 6 indicates the method:

$$T_m^* = \frac{\int_0^\infty t\, f(t)\, dt}{\int_0^\infty f(t)\, dt} \qquad (6)$$

In general, T_m^* will always be the true mean residence time of the tracer in the system under study. The tracer mean residence time of the system will satisfy the relationship of equation 2, however, only under the conditions of identical behavior of tracer and substance traced. In trees, the nominal mean residence time for water, T_m, is not equal to the tracer mean residence time T_m^* since the tracer apparently undergoes interactions with the conducting vessels of the tree. The relationship is given by the expression

$$T_m^* = f_1\, T_m + f_2\, T_H$$

where T_H is the residence time of the fraction of tritium which has undergone some interaction with the wood and f_1 and f_2 are fractions of the total tritium which pass through the tree without interaction and with interaction, respectively. Possible interactions include isotopic exchange of tritium with hydrogen of the wood or diffusion of tritium into nonflowing compartments of plant water. Experimentally $T_m^* > T_m$ has been found for all trees which have been examined to date, indicating that the term $f_2\, T_H$ has a nonzero value in trees. This phenomenon has been termed "holdback" by others who have examined tracer dynamics in flowing vessels (Ljunggren 1967). Because of "holdback" the value of T_m^* cannot be used without correction to solve equation 2. The problem of finding an appropriate value for $f_2\, T_H$ is presently unsolved.

In general, the mean residence time for a flowing system can always be obtained by measuring the activity distribution of a tracer in the system at two points along the flow pathway (Ljunggren 1967). If T_1 is the time of passage of the tracer at point 1 and T_2 the time of passage at point 2 further downstream, then $T_m = T_2 - T_1$ where T_m is the mean residence time between the two sampling points. Since the tracer normally undergoes peak broadening, T_1 and T_2 are taken as the times when the peak of the distribution passes the sampling points. In trees we fix the initial position of the tracer by the injection at time $T_1 = 0$. In this special case $T_m = T_2$. The value of T_2 could be measured at any point in the tree downstream to the injection point. In the special case where the downstream sampling point is tree foliage, then $T_m = T_p$ where T_p is simply the time of peak arrival in the foliage.

As a first approximation it can be assumed that T_p is not affected by "holdback" as was T_m^* because tritium is probably removed from free flowing forms equally over the entire activity distribution. That is, interaction of tritium with conducting vessels could as well occur with the isotope in the leading edge of the distribution or the trailing edge. The peak position would, therefore, not be affected by these interactions. This assumption requires experimental verification which is given in the results section.

Correction for Nonconducting Tissue

The foregoing suggests that tritium tracer experiments can only be used to measure actually conducting biomass in trees. Such tissues as bark, flowers, fruit, and nonconducting heartwood will not be included in the estimate. Roots are not included in the estimate since the tracer injection is normally done in tree trunks above the roots. In practical biomass measurements for forestry purposes, the most serious problem is the omission of nonconducting heartwood from the direct measurement. The theory of tracer dynamics cannot be used for direct measurement of this quantity, and it is, therefore, necessary to make an approximation. Equation 7 expresses the relationship between total biomass and the biomass of heartwood and sapwood:

$$V_T\rho_T = V_H\rho_H + V_S\rho_S \ , \qquad (7)$$

where V_T = total volume of tree tissue (cm^3),

ρ_T = weighted mean wood density ($\frac{g}{cm^3}$),

$V_H; V_S$ = volume of heartwood and sapwood (cm^3), and

$\rho_H;\rho_S$ = density of heartwood and sapwood ($\frac{gm}{cm^3}$) .

Substituting the relationships

$$\rho_H/\rho_S = K \qquad \text{and} \qquad V_H/V_S = \lambda$$

into equation 7 results in equation 8:

$$V_T\rho_T = V_S\rho_S\,(\lambda K + 1) \ . \qquad (8)$$

Assuming that the volume of heartwood and that of the total tree can be approximated by a right circular cone, an expression for λ is derived as follows:

$$\lambda = \frac{r^2}{\Delta r\,(2r + \Delta r)} \ ,$$

where r = mean radius of heartwood at base (cm), and

Δr = mean thickness of sapwood at base (cm).

Upon substituting for λ in equation 8, a final expression for tree biomass is obtained:

$$V_T\rho_T = D = V_S\rho_S\left(\frac{r^2 K}{\Delta r(2r + \Delta r)} + 1\right). \qquad (9)$$

The numerical group $V_T\rho_T$ is the total plant biomass (D) and the group $V_S\rho_S$ is the conducting or sapwood biomass as measured by the tritium method. Equation 9 is not sensitive to the assumption that wood volumes are approximated by right circular cones. The same result is obtained for a right circular cylinder or for intermediate figures.

Equation 9 has not yet been evaluated experimentally. It is proposed here to suggest some of the anticipated lines of research to be undertaken in the Coniferous Biome. A principal problem for the solution of equation 9 lies in accurate measurement of the quantities r and Δr. These quantities fundamentally refer to the radii of nonconducting and conducting wood, respectively. In the simplest case they may be coincident with heartwood and sapwood as observed visually. Their evaluation could then be done by straightforward measurement of tree cores.

An accurate solution also depends on the nature of the transition zone between conducting and nonconducting tissue. If this is sharply defined, then r and Δr will be well defined and an accurate solution to equation 9 can be obtained. If the conduction undergoes a gradual transition across the radius of the tree, there may be no practical method for assigning values to r and Δr. It is possible that reasonable values can be obtained by studying tritium distribution along wood cores which have been taken from tritium-labeled trees. These considerations apply to large trees which have appreciable volumes of heartwood. In the trees for which we have experimental data, heartwood was a minor part of the total tree volume and was not considered.

Results

The mean residence time T_m is the most difficult parameter of equation 5 to obtain, principally because it is not generally known a priori which of several possible means of computing it is the correct one. The desired value is the nominal mean residence time T_m (equation 2); however, in the usual nondestructive experiment this is not directly obtainable. Where tritium-injected trees have been harvested, however, the water pool size (C) is directly obtainable from biomass and moisture measurements, and it is possible to tion; however, the mean of a group of such determinations was a reliable and unbiased estimate of the mean obtained by direct harvest. With additional research on the problem it may be possible to reduce the experimental error of the method and to improve the reliability of individual estimates.

Acknowledgments

The work reported in this paper was performed under the auspices of the U.S. Atomic Energy Commission and supported in part by National Science Foundation Grant No. GB-20963 to the Coniferous Forest Biome, U.S. Analysis of Ecosystems, International Biological Program. This is Contribution No. 32 to the Coniferous Forest Biome.

Literature Cited

Bergner, P.—E. E. 1961. Tracer dynamics: I. A tentative approach and definition of fundamental concepts. J. Theor. Biol. 2: 120-140.

__________. 1964a. Tracer dynamics and the determination of pool sizes and turnover factors in metabolic systems. J. Theor. Biol. 6: 137-158.

__________. 1964b. Kinetic theory: Some aspects on the study of metabolic processes. *In* R. M. Kniseley and W. N. Tauxe (eds.), Dynamic clinical studies with radioisotopes, p. 1-18. U.S. At. Energy Comm. TID-7678. Germantown, Md.

__________. 1965. Exchangeable mass: Determination without assumption of isotopic equilibrium. Science 150: 1048-1050.

__________. 1966. Tracer theory: A review. Isotope & Radiat. Tech. 3: 245-262.

Donato, L., C. Giuntini, R. Bianchi, and A. Maseri. 1964. Quantitative radiocardiography for the measurement of pulmonary blood volume. *In* R. M. Kniseley and W. N. Tauxe (eds.), Dynamic clinical studies with radioisotopes, p. 267-283. U.S. At. Energy Comm. TID-7678. Germantown, Md.

Kline, J. R., J. R. Martin, C. F. Jordan, and J. J. Koranda. 1970. Measurement of transpiration in tropical trees using tritiated water. Ecology 5(6): 1068-1073.

Ljunggren, K. 1967. A review of the use of radioisotope tracers for evaluating parameters pertaining to the flow of materials in plant and natural systems. Isotope & Radiat. Tech. 5(1): 3-24.

Orr, J. S., and F. C. Gillespie. 1968. Occupancy principle for radioactive tracers in steady state biological systems. Science 162: 138-139.

Zierler, K. L. 1964. Basic aspects of kinetic theory as applied to tracer distribution studies. *In* R. M. Kniseley and W. N. Tauxe (eds.), Dynamic clinical studies with radioisotopes, p. 55-79. U.S. At. Energy Comm. TID-7678. Germantown, Md.

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