Selection for biomass production based on respiration parameters in eucalypts: acclimation of growth and respiration to changing growth temperature

R.S. Criddle, T.S. Anekonda, R.M. Sachs, R.W. Breidenbach, and L.D. Hansen

Abstract: This paper examines the relation between respiratory physiology and growth rate and the effects of environment on this relation for the purpose of developing means for accelerating and improving selection of trees for biomass production. The relations among biomass production, respiratory metabolism, and growth temperature in controlled environments were determined for three *Eucalyptus* genotypes (clones). *Eucalyptus camaldulensis* 4016, *E. camaldulensis* C11, and *Eucalyptus gundal* (*Eucalyptus gunnii* × *Eucalyptus dalrympleana* hybrid) GD1 were selected for this study because of known qualitative differences in their field growth responses to temperature. These clones were grown in controlled environments at three temperatures. Measurements were made of growth rate, metabolic heat rate, and dark CO_2 production rate for plants grown at each of the three temperatures. This allowed determination of respiration rates of plants originally adapted for growth in different climates, but acclimated during growth at three different controlled temperatures, and also determination of respiration changes resulting from short-term changes in temperature. Growth rates of the three clones differed in their patterns of response to changes in growth temperature. For example, C11 grew most rapidly at the highest temperature, while GD1 was slowest at high temperature. Metabolic rates and the temperature dependence of metabolic rates of the clones differed and the pattern of differences changed when plants became acclimated to growth at different temperatures. Changes in metabolic properties of the three clones with growth and measurement temperatures are consistent with the growth rate changes. In general, increased growth rate was accompanied by increased respiration rate measured either as heat rate or as rate of CO_2 production. Growth rates were inversely related to two measures of metabolic energy use efficiency. Growth rates decreased as values of heat loss per gram dry weight produced and values of heat loss per mole of CO_2 produced increased. Recognition of these relations between growth rate and respiration parameters at different temperatures in controlled environement may allow prediction of relative growth rate performance of *Eucalyptus* clones over a range of growth climates.

Résumé : Nous examinons ici la relation entre la physiologie de la respiration et le taux de croissance, ainsi que l'effet de l'environnement sur cette relation, dans le but de développer des méthodes visant à accélérer et à améliorer la sélection des arbres pour la production de biomasse. Nous avons déterminé sous environnement contrôlé les relations entre la production de biomasse, le métabolisme respiratoire et la température de croissance chez trois génotypes (clones) d'*Eucalyptus*. Le choix de *Eucalyptus camaldulensis* 4016, *Eucalyptus camaldulensis* C11 et *Eucalyptus gundal* (*Eucalyptus gunnii* × *Eucalyptus dalrympleana*) GD1 était basé sur notre connaissance préalable de différences qualitatives dans leur réaction en croissance au champ à la température. Nous avons cultivé ces clones dans un environnement contrôlé sous trois températures, et nous avons mesuré leur taux de croissance, leur taux de production de chaleur métabolique et leur taux de production de CO_2 en condition de noirceur à ces trois températures. Ces mesures permettaient de déterminer le taux de respiration de plantes préalablement adaptées à des conditions de croissance différentes, mais acclimatées en cours de croissance à trois températures, ainsi que les changements dans le taux de respiration suite à des changements de température de courte durée. Les trois clones ont montré des réponses différentes dans leur taux de croissance lorsque la température de croissance changeait. Par exemple, la température de croissance la plus

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élevée a produit la croissance la plus forte chez le clone C11, mais la plus faible chez le clone GD1. Les clones différaient dans leur taux de production de chaleur métabolique ainsi que dans la dépendance thermique de ce taux, et le patron de ces différences changeait selon la température à laquelle les clones étaient acclimatés. Les changements dans les propriétés métaboliques des trois clones en fonction de leur croissance et de la température sont consistants avec les changements observés dans leur taux de croissance. En général, une augmentation du taux de croissance était accompagnée d'une augmentation du taux de respiration déterminé soit par le taux de production calorifique, soit par le taux de production de CO_2. Le taux de croissance était inversement relié à deux mesures d'efficacité d'utilisation de l'énergie métabolique : la perte calorifique par gramme de masse sèche produite, et la perte calorifique par mole de CO_2 produit. La détermination de ces relations entre le taux de croissance et les paramètres de la respiration à des températures différentes dans des conditions environnementales contrôlées pourrait nous permettre de prédire les performances de croissance relative de différents clones d'*Eucalyptus* sous différentes conditions climatiques.

[Traduit par la Rédaction]

Introduction

Both genotype and environment influence the relation between growth and respiration in plants (Wilson and Jones 1982; Amthor 1989; Hansen et al. 1989, 1992, 1994; Thornley and Johnson 1990; Criddle et al. 1991*a*, 1991*b*, 1994; Anekonda et al. 1993, 1994*a*, 1996). A previous paper presented a mechanistic model describing the relations among metabolic heat rate, metabolic energy use efficiency, and growth rate of plants (Hansen et al. 1994). Measured growth and respiration properties of several species have been used to test predictions of this model (Criddle et al. 1994; Hansen et al. 1992; Anekonda et al. 1994*b*). In addition to growth predictions, the model allows description of the effect of temperature on the growth–respiration relationship. Anekonda et al. (1996) demonstrated the significance of matching the temperature dependence of respiratory metabolism to growth environment for maximizing growth; however, the precise nature of the growth–respiration–temperature relationship was not explicitly examined. This study, therefore, extends examination of respiration–growth relations in selected *Eucalyptus* clones under well-defined, controlled conditions to determine whether it will be possible to use short respiration measurements to predict optimum growth climates for individual clones.

Each *Eucalyptus* genotype tested had a unique pattern of change in growth and respiration parameters with change in temperature. Differences in the respiratory parameters could be related to known differences in climates best suited for growth of the clones. Growth of plants under temperature-stressed conditions resulted in poor growth and was accompanied by changes in respiration that reflected the changed growth rates.

Eucalypts acclimate by adjusting respiration parameters when growth temperatures are changed. The range of temperatures to which individual clones can acclimate for efficient growth is small, however, and differs for the three clones that we studied. The patterns of change in respiratory parameters during acclimation to a fixed average growth temperature (days) and the changes in respiration during short-term (hours) temperature fluctuations both provide information useful for predicting growth rate responses of individual clones to different temperature regimens. Measurements of respiration as a function of temperature can be used to rapidly identify suitable growth temperatures and thereby aid identification of genotypes most suited for growth at a selected location. This ability could decrease time for selection of eucalypts suited for rapid growth at proposed planting locations.

Materials and methods

Eucalyptus clones studied

Three *Eucalyptus* clones, *Eucalyptus camaldulensis* 4016, *E. camaldulensis* C11, and *Eucalyptus gundal* GD1, were used in this investigation. Clone 4016 was selected by the Eucalyptus Improvement Association of California from a plantation maintained by San Diego State University at Temacula, Calif. C11 was selected from the seed source of Lake Albacutya provenance, Australia, and grown at the University of California, Davis. GD1 is an interspecific hybrid of *Eucalyptus gunnii* × *Eucalyptus dalrympleana*. These clones were initially propagated in a greenhouse at the University of California at Davis, using standard procedures for rooting stem cuttings by mist propagation (Sachs et al. 1988). Shoots of ramets of all the clones were trimmed to similar heights before beginning growth-chamber studies. Between-clone leaf sizes were different for the three clones. Of the three clones, GD1 had the smallest leaves. Hence, leaf numbers and sizes of 4016 and C11 were visually normalized by removing extra leaves and trimming leaf size to achieve the same leaf area index for all three clones, thus minimizing differences in light absorption as a possible variable in growth rate differences.

The three *Eucalyptus* clones studied were selected because of observed differences in their field growth rates in different climates. GD1 is a cold-tolerant clone. C11 and 4016 both grow better in warmer climates; however, C11 trees survived a 1990 freeze better than 4016 at Davis, while 4016 grew faster than C11 at Corning, Calif., where summer temperatures are higher and winter temperatures are lower than at Davis (unpublished observations by R.M. Sachs and M.J. Booth). Thus, these three clones represented different patterns of growth responses to field temperature regimes.

Growth conditions

Growth conditions, including light intensity, were carefully controlled. Only growth temperature was varied so that changes in growth rates could be related to differences in the responses of these clones to temperature. Eight to 10 rooted cuttings (ramets) from each of the three genotypes (clones) were examined to define within-genotype variation in growth rates and respiration. Because of the limited number of available cuttings, we could not maintain an equal ramet number for each clone. The same ramets and clones were used for measurements

Table 1. Pearson correlation coefficients between growth and respiratory traits for paired ramets of the same clone at different growth temperatures.

Temperature (°C)[a]	Light ($\mu mol \cdot m^{-2} \cdot s^{-1}$)	Ramets	$\dot{q}_{15}$	$\dot{q}$	μ_q	R_{CO_2}	$\dot{q}/R_{CO_2}$	R_{SG}	$\dot{q}$/biomass
15:10	700	22	0.30	0.34	0.26	0.41	−0.30	0.39	−0.74
			++	++	ns	*	++	+	****
20:15	700	18	−0.01	0.00	0.04	0.17	−0.22	0.19	−0.71
			ns	ns	ns	ns	ns	ns	***
30:25	700	22	0.61	0.66	0.28	0.62	−0.39	0.56	−0.28
			**	**	ns	**	+	**	ns
30:25	150	22	−0.20	−0.01	0.17	0.21	−0.42	0.32	−0.87
			ns	ns	ns	ns	+	ns	***

Note: Units of respiration traits were as follows: $\dot{q}_{15}$ and q, $\mu W \cdot (g \times 10^{-3})^{-1}$; μ_q, kK; R_{CO_2} and R_{SG}, $pmol \cdot (g \times 10^{-3})^{-1} \cdot s^{-1}$; $\dot{q}/R_{CO_2}$, $kJ \cdot mol^{-1}$; $\dot{q}$/biomass, $\mu W \cdot (g \times 10^{4})^{-2} \cdot d$ (all mass measurements were determined on a dry-weight basis). ns, not significant, by *t*-test; ++, $0.15 \geq p > 0.10$; +, $0.10 \geq p > 0.05$; *, $0.05 \geq p > 0.01$; ***, $0.001 \geq p > 0.0001$; and ****, $p < 0.0001$.

[a]Light:dark.

at all growth temperatures to minimize nongenetic differences in temperature responses.

Plants were grown in 10-cm diameter, 200-mL pots in a peat–sand–perlite soil mix in a controlled growth chamber at the University of California at Davis, Calif. All plants were irrigated with a 50% Hoagland nutrient solution. Photoperiods of 16 h light : 8 h dark were used. Light in the chamber was maintained at a constant 700 $\mu mol \cdot Q \cdot m^{-2} \cdot s^{-1}$ photon flux density during daylight illumination for all but one growth experiment. For this experiment, light was at 150 $\mu mol \cdot Q \cdot m^{-2} \cdot s^{-1}$. First, the ramets were grown in the low light, at 30:25°C light:dark temperature for 15 days. Then, the same ramets were grown sequentially at 20:15°C for 14 days, at 15:10°C for 23 days, and 30:25°C for 15 days. Shoots formed during growth at each temperature regimen were carefully harvested, leaving behind the original shoot and leaves. Our study does not address the effects of sequential harvest or the influence of belowground biomass on respiration traits. The harvested shoots of each plant were vacuum dried overnight at 75°C and weighed. Biomass production per day, i.e., total dry mass per days of growth, was used as the standard unit of growth rate.

Sample collection for calorespirometric measurement

Shoot apices, including terminal buds and subapical portions of rapidly growing tissues, were collected from each ramet a few minutes before the remainder of new biomass was harvested. The samples were collected near 08:30 and placed in small vials with cold, half-strength Hoagland's solution containing 1% sucrose. The vials were placed on ice during transport to the laboratory and then stored in an aerated refrigerator at 5°C until use. All samples collected from the chambers were stored at least 0.5 h in the refrigerator before starting the first measurements. Respiratory rates of the tissues declined during the first 0.5 h of storage after cutting and then remained nearly constant for the next 2–3 days.

Calorespirometric measurements

Calorespirometric measurements were made using a Hart Scientific model 7707, heat-conduction, differential, scanning calorimeter operated in the isothermal mode (Criddle et al. 1991*a*). Approximately 1 cm long sections, including the apical meristem with subtending developing stem and leaves, were sealed in the 1-cm^3 calorimeter ampules along with a 50-μL vial. Metabolic heat rates were measured with 40 μL of H_2O in the vial. Then, the H_2O was removed and replaced with 40 μL of 0.4 M NaOH. CO_2 produced by the respiring tissues was absorbed by the NaOH to produce carbonate ion and liberate additional heat at a rate proportional to the CO_2 production rate. These sequential measurements provide two, partially overlapping measures of tissue respiration rates. The ratio of heat rate/CO_2 rate ($\dot{q}/R_{CO_2}$), i.e., the enthalpy loss per mole of CO_2 formed, provides information on metabolic energy use efficiency. For further details of the methods refer to Criddle et al. (1991*a*, 1991*b*) and Hansen et al. (1994). Throughout this study, temperature at which a given respiratory parameter is measured is identified by a subscript if other than at 25°C. For example, metabolic heat rate measured at 15°C is denoted $\dot{q}_{15}$ and CO_2 production rate at 25°C as R_{CO_2}. Heat rates at 15 and 25°C and the rate of CO_2 production rate at 25°C were all measured directly. Values of the temperature coefficient (μ_q) from the Arrhenius equation defining reaction rate changes with temperature, and ($\dot{q}/R_{CO_2}$) at 25°C, were calculated from the measured values.

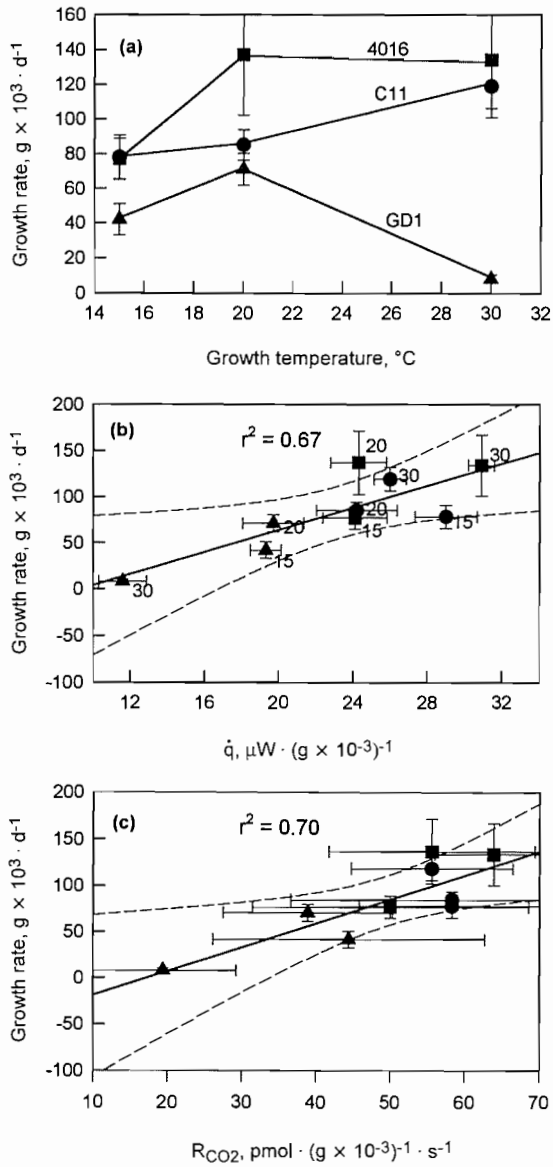
Data analysis

Growth of the same ramets of the three clones sequentially in each of the three different temperature regimens allowed pairing of values for the same ramet at each growth temperature to minimize intraclonal variations and allowed two-way analysis of variance using VARCOMP procedures for unbalanced classification (SAS Institute Inc. 1992). This analysis allows estimation of variance due to temperature, clones, and interaction between them.

Results

An estimate of the strength of correlations between biomass production and respiratory variables for all three clones without considering differences among the genotypes is given by Pearson correlation coefficients (Table 1). Statistically significant, positive correlations were observed between biomass production and many respiration variables ($r = 0.30–0.66$) at both the highest (30:25°C) and the lowest (15:10°C) of the three temperatures. The values of $\dot{q}_{15}$, $\dot{q}$, R_{CO_2}, and μ_q were positively correlated with

Fig. 1. Average growth rates of ramets of clones 4016, C11, and GD1 plotted as functions of (*a*) growth temperature in the controlled environment chambers, (*b*) metabolic heat rates measured at 25°C, and (*c*) CO_2 rates measured at 25°C. In all figures, ▲ represents GD1, ● represents C11, and ■ represents 4016. Of the two values available for metabolic parameter at each temperature (Table 1), the value with lower standard error, or if the standard error is equal for both, the value estimated using the large number of ramets, is plotted in this and subsequent figures. Mass measurements were on a dry-weight basis.



biomass growth rate, whereas $\dot{q}/R_{CO_2}$, which is a measure of energy use inefficiency, was negatively correlated. In all growth conditions, heat lost per unit of biomass ($\dot{q}$/biomass) was strongly ($p < 0.001$) and negatively correlated ($r = -0.74$ to -0.87) with biomass. These overall correlations between the respiration parameters and biomass production occur largely in spite of clonal differences in responses to temperature. The strengths of the correlations vary with growth and measurement temperatures, however, showing that major genotype specific differences exist in respiration responses to temperature. Little correlation was found between respiration parameters and biomass production for plants grown at intermediate temperatures of 20:15°C and plants grown in limiting light of 150 μmol·Q·m^{-2}·s^{-1}.

Table 2. Analysis of variance model for variance-component estimation.

Source[a]	Expected mean square
T	Var(E) + 7.07 Var(T×C) + 0.01 Var(C) + 21.19 Var(T)
C	Var(E) + 7.21 Var(T×C) + 21.32 Var(C)
T×C	Var(E) + 7.06 Var(T×C)
E	Var(E)

[a]T, temperature; C, clone; E, error (among ramets within clones).

Table 2 provides analysis of variance model for variance-component estimation. Variance-component estimates and percentage of total variance for respiration and growth traits are presented in Table 3. Variance-component estimate due to differences in growth temperatures, Var(T), is significant ($p < 0.0001$), accounting 27% of the total variance observed in μ_q. This means changes in growth temperatures significantly influences changes in μ_q but not in other respiration parameters or in biomass. Differences in values of variance components due to the clones, Var(C), are significant ($0.01 > p > 0.0001$) in all but the ratio of heat lost per unit of biomass produced ($\dot{q}$/biomass). Percent Var(C) ranges from 13 ($\dot{q}/R_{CO_2}$) to 42 (R_{CO_2}). Differences in values of variance components due to the interaction between growth temperature and clone, Var($T \times C$), are also significant ($0.05 > p > 0.0001$) in $\dot{q}_{15}$, $\dot{q}$, R_{CO_2}, R_{SG}, and biomass. The variance components that estimated negative values, with their true values likely near zero, are set to zero and their F-tests are presented as nonsignificant in Table 3. The error component of variance, Var(E), measures differences between ramets taken from the same clone.

Figure 1 illustrates growth rates of the three genotypes as functions of temperature, metabolic heat rate, and CO_2 rate. In these and subsequent plots, values of growth temperature as well as discussions of the data all refer to temperatures during daytime growth. Figure 1*a* illustrates the differences in growth rate responses of the three genotypes to growth temperature. GD1 grows moderately well at lower temperatures, but growth rate goes to near zero when the temperature is raised to 30°C. Clone 4016 increases growth rate with temperature rapidly at lower temperatures, but does not increase significantly between the 20 and 30°C data points. C11 growth rate increases more rapidly with temperature between 20 and 30°C than at lower temperatures. Error bars in Fig. 1 and subsequent figures represent clonal means and their standard errors for all the measurements. The broken lines indicate 99% confidence intervals for regression lines. Figures 1*b* and 1*c* show that growth rates increase with increasing respiration rates measured as either $\dot{q}$ or R_{CO_2}.

Table 3. Variance-component estimates and estimated variance components as percentage of total variance for measured and calculated respiration and biomass traits.

Traits[a]	Var(T)	%	Var(C)	%	Var($T\times C$)	%	Var(E)	%
$\dot{q}_{15}$	0	0ns	1.749	29****	2.458	40****	1.909	31
$\dot{q}_{25}$	0	0ns	19.12	38****	17.25	34****	14.16	28
R_{CO_2}	0	0ns	140.4	42****	76.88	23***	116.5	35
$\dot{q}/R_{CO_2}$	0	0ns	162.9	13**	138.3	11ns	928.0	75
μ_q	0.235	27****	0.139	16**	0	0ns	0.492	57
R_{SG}	0	0ns	678.8	31****	310	14*	1168	54
Biomass	0	0ns	1160	28****	614.5	15*	2380	57
$\dot{q}$/Biomass	0	0ns	47.36	1ns	221.8	4ns	5259	95

Note: Statistical significances indicated for the Var(T) and Var(C) percentages are for the associated variance components. ns, not significant; *, $0.05 > p > 0.01$; **, $0.01 < p < 0.001$; ***, $0.001 < p < 0.0001$; and ****, $p < 0.0001$.

[a]See Table 1 for measurement units.

Figure 2*a* shows that growth rates decrease as the amount of heat lost per milligram of biomass produced ($\dot{q}/R_{CO_2}$) increases. When the heat loss per milligram of biomass produced is large, the growth rate is slow. Note that heat rate is measured in these studies at a single time just prior to harvest of the tissues and represents only a snapshot of metabolic activity at that time. Biomass production rate is averaged over the total time at a given temperature. The heat rate/growth rate ratio is plotted here as a log function, simply to keep the numbers on a reasonable scale for presentation. The data show that the rates of biomass formation for *Eucalyptus* genotypes are strongly dependent on the fraction of metabolic energy lost as heat. This relation holds for all genotypes measured and at all growth temperatures.

Figure 2*b* shows the heat rate/growth rate ratio for each of the three genotypes as a function of growth temperature. GD1 and 4016 both have minimums at 20°C. The ratio for GD1 at 30°C is very high, indicating poor energy coupling and a large loss of heat per milligram of tissue formed at this temperature. At 30°C this genotype has a very low growth rate (Fig. 1*a*). By the criterion of heat rate/growth rate, all three genotypes are less efficient at 15°C than at 20°C. Efficiency of 4016 may increase marginally between 20 and 30°C. Efficiency of C11 increases from 20 to 30°C. GD1 efficiency decreases drastically as temperature increases from 20 to 30°C. Clearly GD1 becomes inefficient at higher temperatures, while C11 may have increased efficiency at temperatures even above 30°C. These efficiency results are consistent with the growth rate data of Fig. 1*a*, indicating that metabolic energy efficiency (also known as substrate carbon conversion efficiency) is temperature dependent and a determinant of growth rate.

The ratio of heat rate to biomass accumulation rate plotted in Figs. 2*a* and 2*b* is not generally obtainable for large plants and field growth conditions. It is used here only as a means for establishing the relation between efficiency and growth and the effect of temperature on efficiency. However, it is possible to measure efficiency without knowing growth rates by determining $\dot{q}/R_{CO_2}$ (Hansen et al. 1994; Anekonda et al. 1996). Figure 2*c* shows that growth

Fig. 2. (*a*) Average growth rate plotted versus the ln(heat production rate divided by the growth rate). (*b*) The effect of growth temperature on the heat rate per milligram of biomass produced. (*c*) Average growth rate of ramets of 4016, C11, and GD1 plotted versus $\dot{q}/R_{CO_2}$.

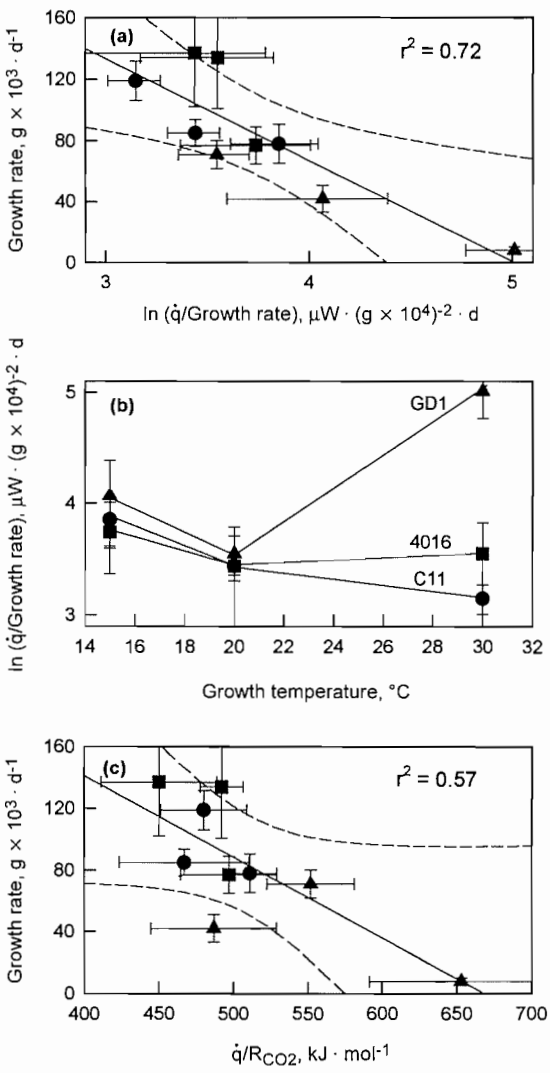
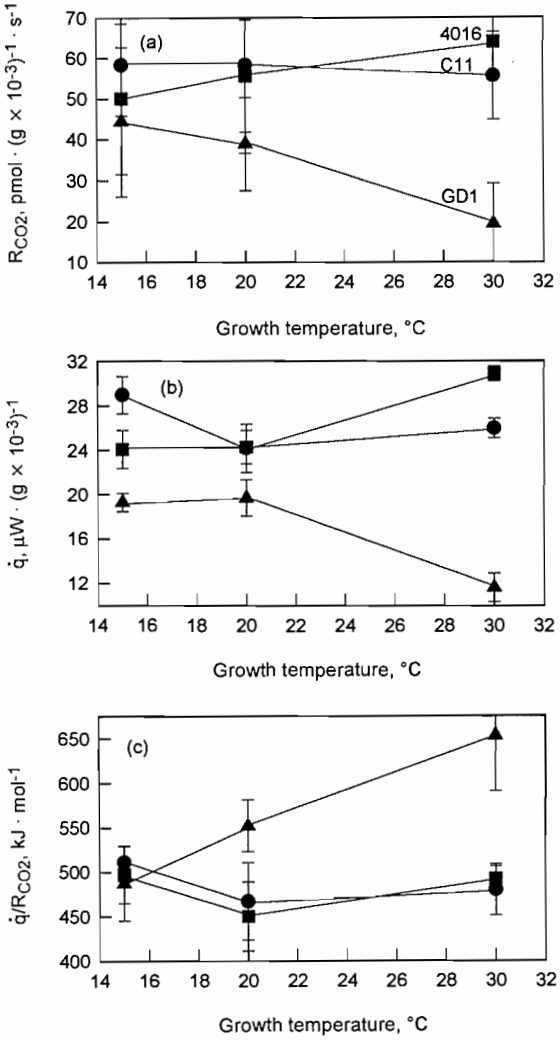


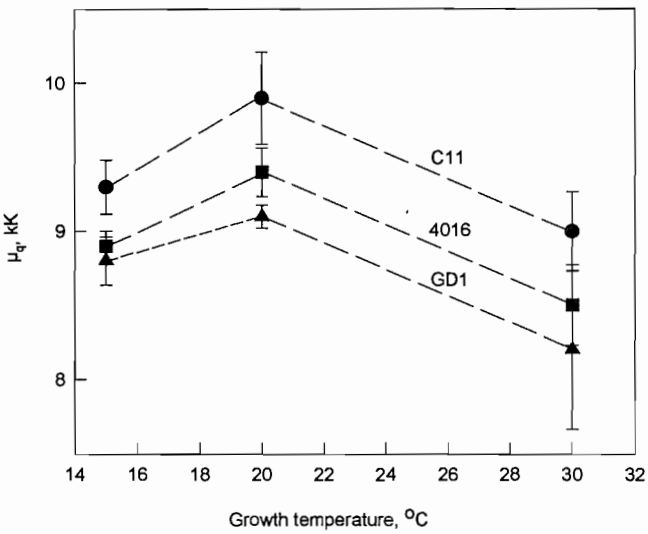
Fig. 3. Effects of growth temperature on (*a*) the dark rate of CO_2 production (R_{CO_2}), (*b*) the metabolic heat rate ($\dot{q}$), and (*c*) the heat produced per mole of CO_2 produced ($\dot{q}/R_{CO_2}$).



rates for individual genotypes and the growth rates for all three genotypes considered together decrease with increase in $\dot{q}/R_{CO_2}$ in the same way as growth rates change with $\dot{q}$/growth rate in Fig. 2*a*. Figure 2*c* shows that when heat per mole of CO_2 produced by metabolism is large, the plant is less efficient in conserving energy for use in biosynthetic pathways and growth rate is decreased. Thus $\dot{q}/R_{CO_2}$ can be used as an indicator for substrate carbon conversion efficiency in predictions of growth rate.

As an additional means of understanding the data in Fig. 2*c*, we now focus on each metabolic rate parameter as a function of growth temperature (Fig. 3). R_{CO_2} of C11 (measured at 25°C) does not change significantly with growth temperature (Fig. 3*a*). The CO_2 rate for 4016 increases steadily between 15 and 30°C growth temperatures. Values of R_{CO_2} for GD1 decrease by about one-half as growth temperature is increased from 15 to 30°C. Similarly, $\dot{q}$ values also show genotype-specific responses to growth temperature (Fig. 3*b*). A plot of $\dot{q}_{15}$ appears

Fig. 4. Variation in values of μ_q, measured between 15 and 25°C for three *Eucalyptus* genotypes, with differences in growth temperature.



much the same as Fig. 3*b*; only the expected rate differences resulting from measurements at different temperatures are apparent. The ratio $\dot{q}/R_{CO_2}$ for the three clones is plotted against growth temperature in Fig. 3*c*. Changes in this ratio with growth temperature are of course the result of the changes in both $\dot{q}$ and R_{CO_2} discussed above. The ratio increases rapidly for GD1 as growth temperature is increased, but falls and then increases slightly for 4016 and C11. Figure 3*c* demonstrates the same dependence of $\dot{q}/R_{CO_2}$ on temperature as was earlier shown for heat rate per growth rate data in Fig. 2*b*.

Another parameter measured in these studies, the Arrhenius temperature coefficient of metabolism (μ_q), has been shown previously to correlate weakly with growth rates of various *Eucalyptus* species (Anekonda et al. 1996). Values of μ_q for the three genotypes differ with growth temperature (Fig. 4). In all cases, μ_q for the plants growing at 20°C is significantly larger than that for plants growing at 15 or 30°C. These *Eucalyptus* clones adapt μ_q to current growth conditions in the same direction and maintain the same relative ranking according to μ_q over the entire temperature range studied.

Discussion

Growth rate data for the three *Eucalyptus* clones as a function of growth temperature in controlled climate chambers are consistent with the generally recognized differences in field growth rates of these three *Eucalyptus* genotypes in different climates. Figure 1*a* shows that GD1 does not grow well at the higher temperature; in fact, it virtually shuts down growth at 30°C. In the field, GD1 is cold hardy and does not grow well in warm climates. Growth rate of 4016 increases rapidly between 15 and 20°C, but does not increase much further as temperature continues to increase. C11 growth rate increases steadily with temperature. C11 and 4016 grow about equally well in the central portion of the Central Valley of California. One could speculate

from Fig. 1*a* that in the hotter, northern end of the valley C11 may surpass growth of 4016. However, rigorous field test are required to further tests these predictions.

Significant differences exist in respiratory parameters among the three *Eucalyptus* genotypes studied here as well as among *Eucalyptus* species and other genotypes within a species. Moreover, we show here that the patterns and range of changes in respiratory parameters during acclimation to new growth temperatures differ with genotype. Some genotypes are more capable of acclimating growth to a given temperature regimen than are others. Analysis of the patterns of variation in metabolic parameters with growth temperature can thus reveal which genotypes are better suited for rapid growth in a given temperature environment. Acclimation changes in respiratory parameters are small, however, when compared with the range of variability of these parameters within a species. For example, changes in growth temperature from 15 to 30°C gives μ_q values ranging from 7.8 to 9.9 kK for clones in this study compared with the overall range of μ_q from 5 to 10 kK exhibited within and among *Eucalyptus* species (Anekonda et al. 1996).

The data in Fig. 1 indicate that an overall positive correlation can sometimes be found between growth rate and respiration rate. This is consistent with the experience of many workers and numerous reports in the literature (Amthor 1989) and with our model of plant growth rates (Hansen et al. 1994). Even with differences among genotype and growth temperatures in this study, an overall positive correlation exists between growth rate and CO_2 rate as well as growth rate and metabolic heat rates measured at either 15 or 25°C. There is a negative correlation between growth rate and $\dot{q}/R_{CO_2}$, as expected. Still, these correlations are not found for all genotypes at all growth temperatures. Correlations with a single measure of respiration rate do not contain sufficient information to provide a mechanistic understanding of the growth–respiration relation (Hansen et al. 1994, 1995). The absence of significant correlation at 20:15°C shows the necessity for a more complex function than a simple proportionality between respiration rate and growth rate. Changes in metabolic efficiency across this temperature range must also be considered (Hansen et al. 1994, 1995).

Figure 1 shows that a major reason for differences in relative growth rate behavior of the three genotypes with changing growth temperature is the difference in temperature dependence of the metabolic parameters. The data show that while growth rates are proportional to heat rates and rates of CO_2 production (Figs. 1*b*, 1*c*), $\dot{q}/R_{CO_2}$ does not remain constant as growth temperature changes (Fig. 3*c*). Thus, we know that acclimation to new growth temperatures affects $\dot{q}$ and R_{CO_2} differently.

Values of $\dot{q}/R_{CO_2}$ are related to substrate carbon use efficiency (ε) by eq. 1 (Hansen et al. 1994):

$$[1] \quad \frac{\dot{q}}{R_{CO_2}} = -\left(1 - \frac{\gamma_P}{4}\right)\Delta H_{O_2} - \frac{\varepsilon}{1-\varepsilon}\Delta H_B$$

where γ_P is the average chemical oxidation state of stored photosynthate, ΔH_{O_2} is a constant equal to −455 kJ/mol, and ΔH_B is the overall enthalpy change per mole of C (photosynthate) incorporated into biomass. Lower $\dot{q}/R_{CO_2}$ translates to higher efficiency in this equation if γ_P is constant and ΔH_B is positive and constant, and ultimately to faster growth (Figs. 2*a*, 2*b*), since growth rate depends on efficiency. Differences in $\dot{q}/R_{CO_2}$ with growth temperature, therefore, indicate changes in relative growth rates of the clones at different temperatures. The consequences of these conclusions are apparent when examples of growth rates of the genotypes under specific conditions are considered. For example, as growth temperature increases, $\dot{q}/R_{CO_2}$ for GD1 becomes slightly higher (less efficient) at 20°C, then even larger (highly inefficient) at 30°C (Fig. 1*a*). Metabolic energy is not coupled effectively to production of biomass at 30°C, and growth nearly ceases. The energy lost as heat per milligram of tissue formed for GD1 at 30°C over the period of growth is nearly seven times that for 4016 grown at this temperature (Fig. 2*b*). This same relation is evident from Fig. 3*c*, where efficiency, as measured by heat produced per CO_2, is again very low for GD1 grown at 30°C and higher at 15 and 20°C, temperatures where growth is faster.

Hansen et al. (1995) have shown that growth rate of plants is a product of respiratory rate multiplied times the substrate carbon conversion efficiency. GD1 has low efficiency and low metabolic rate at 30°C and consequently grows slowly. Both efficiency and metabolic rate of GD1 are higher at 20°C to account for the higher growth rate at this temperature. Clearly this is a genotype-specific response shown by calorespirometric as well as growth rate analysis to be adapted to growth in cool climates. Also the studies show that GD1 does acclimate during growth at different temperatures, but this acclimation is limited. In contrast, 4016 has high respiratory rate, high efficiency, and consequently high growth rate at 30°C and lower metabolic rate with near the same efficiency at 15°C. These values predict good growth across the temperature range with improved growth rates at the higher temperatures as observed. An important consequence of the relation between $\dot{q}$, $\dot{q}/R_{CO_2}$, and growth rate is that the respiratory values can be readily and rapidly measured in the laboratory on small tissue segments from plants of nearly any size or age as a function of temperature. This provides a simple rapid screening procedure for potential growth rate at a given temperature.

The temperature dependence of metabolism has previously been shown to be an important variable in adaptation of woody shrubs (Criddle et al. 1994) and of *Eucalyptus* (Anekonda et al. 1996) species to different temperature regimens. Values of μ_q differ within a species for plants from different locations, and these differences persist in common gardens (Criddle et al. 1994). Values of μ_q are generally lower for genotypes from higher latitudes and elevations (Criddle et al. 1994). This is consistent with Fig. 4, showing that GD1, which is adapted to cooler climates, has lower values of μ_q.

Three important conclusions relating μ_q to growth of three clones follow from these controlled environment results. (*i*) The temperature dependence of metabolism is a genetically defined parameter linked to the ability of a genotype to grow well and survive in a given climate. (*ii*) Values of μ_q can change over a small range to allow

limited acclimation of plants to changing seasonal growth conditions. (*iii*) The range of acclimation is specific to a genotype. As seasonal changes in μ_q are small and roughly parallel for plants in a common garden, measurements of μ_q may serve as useful indicators for predicting which genotypes will prosper at a selected growth site.

Anekonda et al. (1996) demonstrated that growth rates of trees from 17 *Eucalyptus* species increased with increasing values of μ_q up to some maximum value, then decreased as μ_q increased further. They postulated that the optimal value for μ_q would be different if the average growth temperature was changed. In Fig. 4, values of μ_q do in fact change with growth temperature. Changes in μ_q with growth temperature appear to be parallel for the genotypes in this plot, but there are insufficient temperature points to define the curves at intermediate temperatures. It is clear that μ_q reaches a maximal value somewhere between 20 and 30°C. The ability to alter μ_q toward the optimal value for growth in a given climate is a critical factor in growth and survival of plants and an important variable to consider when selecting plants for growth at locations distant from the production nursery or a breeding nursery. The optimal value of μ_q for growth depends on both average temperature and on the diurnal temperature range (Criddle et al. 1994). This paper demonstrates the effect of average temperature; the effect of diurnal temperature fluctuations on values of μ_q is briefly considered elsewhere (Anekonda et al. 1996) but still needs to be examined in detail.

No previous studies have coupled careful examination of growth rates in controlled environments at different temperatures with measurement of multiple respiratory measurements over a range of temperature. The results presented here show a positive linear correlation between metabolic rate and CO_2 rate and growth rate. The relation is retained for all three genotypes studied and at the three growth temperatures. In addition, growth rates are inversely related to the heat production per mole of CO_2 produced, a measure of energy inefficiency in metabolic processes. Such correlations show that it may be possible to use rapid respiration measurements to accurately identify climates suitable for growth of these and other genotypes and to predict their relative growth rates in these climates.

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References

Amthor, J.S. 1989. Respiration and crop productivity. Springer-Verlag, Berlin.

Anekonda, T.S., Criddle, R.S., Libby, W.J., and Hansen, L.D. 1993. Spatial and temporal relationships between growth traits and metabolic heat rates in coast redwoods. Can. J. For. Res. **23**: 1793–1798.

Anekonda, T.S., Criddle, R.S., Libby, W.J., Breidenbach, R.W., and Hansen, L.D. 1994*a*. Respiration rates predict differences in growth of coast redwood. Plant Cell Environ. **17**: 197–203.

Anekonda, T.S., Criddle, R.S., and Libby, W.J. 1994*b*. Calorimetric evidence for site adapted biosynthetic metabolism in coast redwood. Can. J. For. Res. **24**: 380–389.

Anekonda, T.S., Hansen, L.D., Bacca, M., and Criddle, R.S. 1996. Selection for biomass production based on respiration parameters in eucalypts: effects of origin and growth climates on growth rates. Can. J. For. Res. **26**: 1556–1568.

Criddle, R.S., Fontana, A.J., Rank, D.R., Paige, D., Hansen, L.D., and Breidenbach, R.W. 1991*a*. Simultaneous measurement of metabolic heat rate, CO_2 production, and O_2 consumption by microcalorimetry. Anal. Biochem. **194**: 413–417.

Criddle, R.S., Breidenbach, R.W., and Hansen, L.D. 1991*b*. Plant calorimetry: how to quantitatively compare apples and oranges. Thermochim. Acta, **193**: 67–90.

Hansen, L.D., Lewis, E.A., Etough, D.J., Fowler, D.P., and Criddle, R.S. 1989. Prediction of long-term growth rates of larch clones by calorimetric measurement of metabolic heat rates. Can. J. For. Res. **19**: 606–611.

Hansen, L.D., Woodward, R.A., Breidenbach, R.W., and Criddle, R.S. 1992. Dark metabolic heat rates and integrated growth rates of coast redwood clones are correlated. Thermochim. Acta, **211**: 21–32.

Hansen, L.D., Hopkin, M.S., Rank, D., Anekonda, T.S., Breidenbach, R.W., and Criddle, R.S. 1994. The relation between plant growth and respiration: a thermodynamic model. Planta, **194**: 77–85.

Hansen, L.D., Hopkin, M.S., Taylor, D.K., Anekonda, T.S., Rank, D.R., Breidenbach, R.W., and Criddle, R.S. 1995. Plant calorimetry II: modeling the difference between apples and oranges. Thermochim. Acta, **250**: 215–232.

Sachs, R.M., Lee, C., Ripperda, J., and Woodward, R. 1988. Selection and clonalpropagation of eucalypts. Calif. Agric. **42**: 27–31.

SAS Institute Inc. 1992. SAS/STAT® Guide for personal computers, version 6 edition. SAS Institute Inc. Cary, N.C.

Thornley, J.H.M., and Johnson, I.R. 1990. Plant and crop modeling: a mathematical approach to plant and crop physiology. Oxford Science Publications, Clarendon Press, Oxford.

Wilson, D., and Jones, J.G. 1982. Effects of selection for dark respiration rate of mature leaves on crop yields of *Lolium perenne* cv. S23. Ann. Bot. (London), **49**: 313–320.