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Physiological ecology of seed respiration in some tropical species

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SUMMARY

We examine the relationship of seed oxygen consumption rate ($\dot{V}_{O_2}$) to seed moisture content, seed mass, and seed age in 22 ecologically diverse tropical species. These seeds vary greatly in moisture content, age, mass, mechanism of dormancy and length of viability. We quantify each relationship with a power equation, $\dot{V}_{O_2} = aX^b$, where X is the independent variable. Seed moisture content (MC) explains 80 % of the variation in mass-specific $\dot{V}_{O_2}$ (ml O_2 g^{-1} h^{-1}) among seeds of all species, whereas seed mass explains < 1 %. However, when seeds are reclassified as moist (> 28 % MC) or dry (≤ 28 % MC), seed mass explains 54 % of the variation in mass-specific $\dot{V}_{O_2}$ (ml O_2 g^{-1} h^{-1}) within dry seeds, but no significant variation within moist seeds. In dry seeds, seed age explains only 27 % of the variance in mass-specific $\dot{V}_{O_2}$, although seed age and moisture content are negatively correlated. On a per seed basis, seed mass explains 56 % of the variation in $\dot{V}_{O_2}$ (ml O_2 h^{-1} per seed) in dry seeds and 83 % of the variation in moist seeds: the exponents of the power function, 0·54 in dry seeds and 0·78 in moist seeds, are within the range reported for the allometric relationship of oxygen consumption and body size in animals and prokaryotes. We present a framework for future studies that recognizes seed respiration as an important, yet unstudied, component of tropical seed ecophysiology. We discuss the ecological significance of seed respiration in three groups of tropical species which differ in seed moisture content, mass, viability, and post-dispersal moisture regimes: (1) shade-intolerant pioneer species with small, dry, orthodox seeds; (2) seasonally dormant species; and (3) shade-tolerant primary forest species with large, moist recalcitrant seeds.

Key words: Seed respiration, tropical species, seed mass, moisture content.

INTRODUCTION

Respiration of dry, imbibing and germinating seeds has been extensively studied in many agriculturally important temperate species (reviewed by Stiles, 1960; Bewley & Black, 1978, 1982; Priestley, 1986) and in a few economically important tropical species such as avocado (Zauberman & Schiffmann-Nadel, 1972). Considerable variation among species has been found. We know little of the ecological and evolutionary significance of this variation, in part because few species from natural ecosystems have been examined, but also because seed respiration has not been examined from the perspective of physiological ecology.

In this study, we compare seed oxygen consumption rates of 22 tropical species that vary in several ecological characteristics important in seed germination and the dynamics of seeds in tropical soil seed banks (Whitmore, 1983; Garwood, 1989), including life form, successional stage, seed size, mechanism of seed dormancy and length of seed viability (Table 1). Our objectives were (1) to determine the extent of variation in seed respiration among tropical species; (2) to determine which ecologically-important seed attributes, particularly seed size, age, moisture content, viability, and dormancy, best explain interspecific variation in respiration; and (3) to develop a framework of ecological relevance for further studies of seed respiration of tropical species.

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Table 1. *Ecological characteristics of species used in the study*

Species[a]	Family	Seed dry mass (mg)	Recalcitrant (R) or orthodox (O)[b] (seed viability period[c])	Life form (habitat[d])	Dispersal agent[e]	Site[f]	Date[g]
Apeiba membranacea	Tiliaceae	13·9	O (> 9·5 yr***)	Tree (F*)	Mammal	B	1975
Arrabidaea patellifera	Bignoniaceae	22·5	O (> 1·8 yr***)	Liana (F)	Wind*	G	1983
Cissus sicyoides	Vitaceae	18·1	—[h] (> 1·8 yr*)	Vine (C)	Bird	E	1983
Cochlospermum vitifolium	Cochlospermaceae	32·7	O (> 9·5 yr***)	Tree (C*)	Wind	B	1975
Connarus turczaninowii	Connaraceae	366·0	R (< 3 months)	Liana (F)	Bird	B	1984
Diospyros nicaraguensis	Ebenaceae	172·8	O (> 1·8 yr***)	Tree (F)	Mammal	G	1983
Gustavia superba	Lecythidaceae	3180·0	R (< 1 month)	Tree (F)	Mammal	B	1984
Hemiangium excelsum	Hippocrateaceae	80·1	—[h] (1–1·8 yr)	Tree (F)	Wind*	G	1983
Jacaranda copaia	Bignoniaceae	6·5	O[l] (> 5 months**)	Tree (F*)	Wind*	B	1984
Luffa aegyptiaca	Cucurbitaceae	92·2	O (> 1·8 yr***)	Vine (C)	—	G	1983
Maripa panamensis	Convolulaceae	391·0	R (< 1 month)	Liana (F)	Mammal	B	1984
Miconia affinis	Melastomataceae	0·2	O[l] (> 3 months***)	Shrub/tree (F*)	Bird	B	1984
Ochroma pyramidale	Bombacaceae	5·9	O (> 9·5 yr***)	Tree (C/F*)	Wind	B	1984, 1975
Ormosia sp.	Leguminosae	357·0	O[i]	Tree (F)	Bird**	B	1984
Pentagonia macrophylla	Rubiaceae	6·6	R (< 3 months)	Shrub (F)	Bird/ mammal	B	1984
Piper reticulatum	Piperaceae	1·1	R (< 3 months)	Shrub/tree (F*)	Bat	B	1984
Protium tenuifolium	Burseraceae	84·0	R[k]	Tree (F)	Bird/ mammal	B	1984
Protium panamense	Burseraceae	536·0	R (< 1 month)	Tree (F)	Bird/ mammal	B	1984
Psychotria marginata	Rubiaceae	8·4	R[j] (> 3 months*)	Shrub (F)	Bird	B	1984, 1975
Psychotria limonensis	Rubiaceae	7·3	R[k]	Shrub (F)	Bird	B	1984
Randia armata	Rubiaceae	88·9	R (< 5 months)	Tree (F)	Mammal	B	1984
Rhynchosia pyramidalis	Leguminosae	64·0	O[i] (> 3 months*)	Liana (F)	Bird**	B	1984

[a] Nomenclature follows Croat (1978) for Panamanian species and Janzen & Liesner (1980) for Costa Rican species. Four species that never germinated were excluded: *Annona acuminata*, Annonaceae (seed age = 1·8 yr), *Byrsonima crassifolia*, Malpighiaceae (2–64 days), *Doliocarpus olivaceus*, Dilleniaceae (9·5 yr), and *Tetracera* sp., Dilleniaceae (9·5 yr). Data on life form, habitat, and dispersal agent are from Croat (1978), Foster & Brokaw (1982), and Garwood (personal observations).

[b] This is a provisional classification, based on seed viability data and other observations (Garwood, personal observations).

[c] Seed viability period is the time required in dry storage for percent germination to drop to < 10 % of initial values. For species in which viability did not drop to < 10 % by the last germination trial, percent viability relative to initial values is indicated as follows: * < 35 %; ** 50–60 %; *** > 95 %.

[d] Adult habitats are forests (F) or clearing (C). In pioneer species (*), seedlings are shade-intolerant and establish only in high-light environments of forest tree-fall gaps or clearings; seeds are usually facultatively dormant until a gap forms.

[e] Seeds of mammal- and bird-dispersed species are surrounded by moist edible pulp or arils, except for two bird-dispersed legumes (**) with mimetic red or red and black seeds; wind-dispersed seeds are dry. In some wind-dispersed species (*), the wing of the diaspore was included in the respiration measurements.

[f] Seeds were collected in Panamanian tropical semideciduous forests at Barro Colorado Island (B), Panama Province, and Ensenada del Guayabo (E), Darien Province, and in tropical deciduous forest in Guanacaste Province (G), Costa Rica.

[g] Date is year of seed collection.

[h] Not classified because viability was high (> 70 %) after one year of storage but was low (< 20 %) after 2 yr.

[i] The hard-seeded legumes were classified as orthodox, even though viability data were lacking or ambiguous.

[j] Probably recalcitrant, because viability was low (⩽ 30 %) after 3 months.

[k] Probably recalcitrant, based on seed characteristics and germination behaviours (Garwood, unpublished data).

[l] Probably orthodox because short-term viability (3–5 months) was high (> 50 %), but long-term data are lacking.

METHODS

Seed collection and treatments

Seeds were collected primarily from the semi-deciduous tropical forest on Barro Colorado Island (BCI), Panama, in July–August 1984: oxygen consumption was measured on BCI in August 1984. Oxygen consumption of these seeds, as well as of older seeds collected in Panama and Costa Rica, was measured in October 1984 at the University of California at Los Angeles (UCLA). Sites and dates of seed collection are in Table 1.

There were three seed treatments. (1) Air-dry: seeds were air-dried and stored in open plastic bags 1–19 days, depending on species, and in closed plastic bags thereafter. Seeds of *Gustavia superba*, however, were always stored in closed plastic bags to retard fatal desiccation. (2) Continuously moist: seeds were air-dried overnight, placed in closed plastic bags on moist filter paper, and aerated frequently. (3) Wetted: air-dry seeds 2–19 days old were wetted, kept on moist filter paper, and aerated. In the air-dry and continuously moist treatment, seeds for measurement were chosen randomly; the sample was repeatedly measured or another sample was drawn from the pool (with replacement), depending on the sample size available. In the wetted treatment, seeds were randomly chosen from the air-dried treatment, wetted, repeatedly measured, then discarded.

Seeds were air-dried, stored and germinated in an air-conditioned laboratory. Percent moisture content (MC) of whole seeds was calculated on a dry-mass basis ([mass water/dry seed mass] × 100) after drying seeds at 65 °C at least 4 days. Thus, our MC values may be somewhat higher than those based on the ISTA Rules (International Seed Testing Association, 1985). For comparison, literature citations of MC on a wet-mass basis were converted to a dry-mass basis (Roberts & Roberts, 1972, Fig. A4·1). The fleshy pulp of animal-dispersed species was removed immediately after collection.

Percent germination and mean time to germination of freshly collected seeds and stored seeds were measured by placing seeds on moist filter paper at ≈ 25 °C with a ≈ 12 hour light/dark cycle. Hard-coated seeds were pretreated in hot water (≈ 60–70 °C) for 5 min (*Ochroma pyramidale*) or by filing the seed coat (*Rhynchosia pyramidalis* and *Ormosia* sp.).

Measurement of oxygen consumption

Seed oxygen consumption rate ($\dot{V}_{O_2}$), was determined by constant-volume respirometry. Depending on the size of seeds, 1–600 seeds were placed simultaneously in polyethylene syringes with capacities ranging from 10–60 ml. The syringes were then thoroughly flushed with outside air (O_2 content, after removal of H_2O and CO_2, was 20·94 ± 0·001 %), and closed off with a plastic three-way valve. The seeds consumed oxygen within the airtight syringe for a period of 1–24 h in the laboratory, receiving diffuse light up to ≈ 12 h.

The amount of oxygen consumed by the seeds in the syringe was determined with an Applied Electrochemistry S-3A oxygen analyser. Outside air was drawn at a rate of ≈ 30 ml min^{-1} through a 1 m length of tubing (internal volume ≈ 20 ml) into a small H_2O and CO_2 scrubber with an internal volume of ≈ 5 ml, and then through the O_2 sensor. When air from a syringe was injected into the plastic tubing through a closable valve at a rate of ≈ 2 ml s^{-1}, it formed a bolus which smoothly displaced the air already present in the tubing. As the bolus was drawn through the O_2 sensor, the voltage output of the O_2 analyser changed proportionately to the oxygen concentration in the bolus. A computer monitored this change and automatically calculated $\dot{V}_{O_2}$ in ml O_2 h^{-1} corrected to STP (see Bartholomew, Lighton & Louw, 1985, for details of the calculation). The degree of O_2 depletion seldom exceeded 1 % and was generally kept in the range 0·1–0·4 % by adjusting the duration of enclosure.

Determinations on BCI were carried out in an air-conditioned laboratory at 26 ± 1 °C. Determinations at UCLA were carried out at 23 ± 1 °C; these readings were corrected to 26 °C assuming a Q_{10} of 2·0. $\dot{V}_{O_2}$ was expressed as ml O_2 h^{-1} per gram dry mass of seed or as ml O_2 h^{-1} per seed. We present the $\dot{V}_{O_2}$, moisture content, and age of seeds for the 22 species in Appendix 1. No bacterial or fungal contamination was visually evident in our samples of viable seeds; therefore, we assume that oxygen consumption by other organisms was negligible.

Statistics

Regression analysis was carried out using the least squares technique. Students t-test was used to determine whether the slopes differed significantly from zero; analysis of covariance was used to compare slopes. $\dot{V}_{O_2}$ measurements were not included in the analysis if the germination test nearest the measurement date indicated that the seed sample was inviable (0 % germination). For analyses involving seed mass, $\dot{V}_{O_2}$ of dry and moist seeds of each species was averaged separately. $\dot{V}_{O_2}$ was not averaged for analyses of seed age, mean number of days until germination and percent germination.

RESULTS AND DISCUSSION

Seed viability and dormancy

Seeds are classified as orthodox or recalcitrant, based on their ability to tolerate moisture loss (Roberts, 1973). Seeds of orthodox species, which are desiccation tolerant, are usually dormant and have low

moisture content at the time of dispersal. Seeds of recalcitrant species, which are desiccation intolerant, usually have high moisture content and lack dormancy at the time of dispersal (Chin & Roberts, 1980). About half of our 22 species are probably recalcitrant and half orthodox (Table 1).

Seed dormancy in many tropical shade-intolerant pioneer species is broken by changes in light quality or temperature associated with an opening of the forest canopy (Vázquez-Yanes & Orozco Segovia, 1984). Among our species, fluctuating temperatures or scarification of the seed coat break dormancy in seeds of *Ochroma pyramidale* (Vázquez-Yanes, 1974), *Apeiba membranacea* (although older seeds germinate without pretreatment; Garwood, 1986; Acuña & Garwood, 1987), *Rhynchosia pyramidalis* and *Ormosia* sp. (see Methods). Seeds of *Jacaranda copaia* are dormant in the dark (Garwood, unpublished data). Seed dormancy has not been reported in any of the recalcitrant species in this study.

Oxygen consumption and seed moisture content

Moisture content, one of the most important factors controlling seed oxygen consumption within species (Bewley & Black, 1978, 1982), was also a good predictor of the rate of oxygen consumption among species. Values of $\dot{V}_{O_2}$ (ml O_2 g^{-1} h^{-1}) increased with the moisture content of the seeds in both the continuously moist and air-dry treatments (Fig. 1). The regressions of $\dot{V}_{O_2}$ on percent moisture content for both treatments were significant (Table 2), but the slopes (scaling exponents in the relation $Y = aX^b$) of the two treatment groups did not differ ($F = 1{\cdot}304$, d.f. = 1, 39, $P = 0{\cdot}25$). Moisture content explained 80 % of the variation in $\dot{V}_{O_2}$ among all seeds in our samples (Table 2).

$\dot{V}_{O_2}$ decreased significantly with seed age (days since seed collection) in air-dried seeds, but seed age explained only 12 % of the variance in $\dot{V}_{O_2}$ in seeds < 80 days old and 27 % of the variance in seeds of all ages (Table 2). Thus, seed age explained much less of the variance in $\dot{V}_{O_2}$ than moisture content, although percent moisture content of the air-dry seeds was negatively correlated with age ($r = -0{\cdot}53$, d.f. = 24, $P < 0{\cdot}01$, for seeds < 80 days old; $r = -0{\cdot}65$, d.f. = 32, $P < 0{\cdot}01$, for seeds of all ages; age and $\dot{V}_{O_2}$ log-transformed). $\dot{V}_{O_2}$ of air-dried seeds was not correlated with mean number of days from sowing until germination ($r = -0{\cdot}27$, $P > 0{\cdot}1$, d.f. = 24) nor with percent germination ($r = -0{\cdot}11$, $P > 0{\cdot}4$, d.f. = 40). $\dot{V}_{O_2}$ of air-dried seeds is a poor interspecific predictor of time to germination and percent germination.

In orthodox species, long-term viability of seeds in artificial storage decreases as seed moisture content increases up to a critical moisture content, then increases until seeds are fully hydrated if dormancy can be maintained (Roberts, 1973; Villiers & Edgcumbe, 1975; Roberts & Ellis, 1982; Ibrahim, Roberts & Murdoch, 1983). The critical moisture content (dry basis) is about 18–22 % in oily seeds and 35–39 % in non-oily seeds (Roberts, 1973; Roberts & Ellis, 1989). At the critical moisture content in soybean and pea seeds, the rate of change of $\dot{V}_{O_2}$ as a function of moisture content increases sharply because oxidative nonenzymatic and enzymatic (e.g. lipoxidase) reactions are replaced by mitochondrial respiration as free water becomes available (Vertucci & Leopold, 1986, 1987).

There was no obvious change in $\dot{V}_{O_2}$ within the

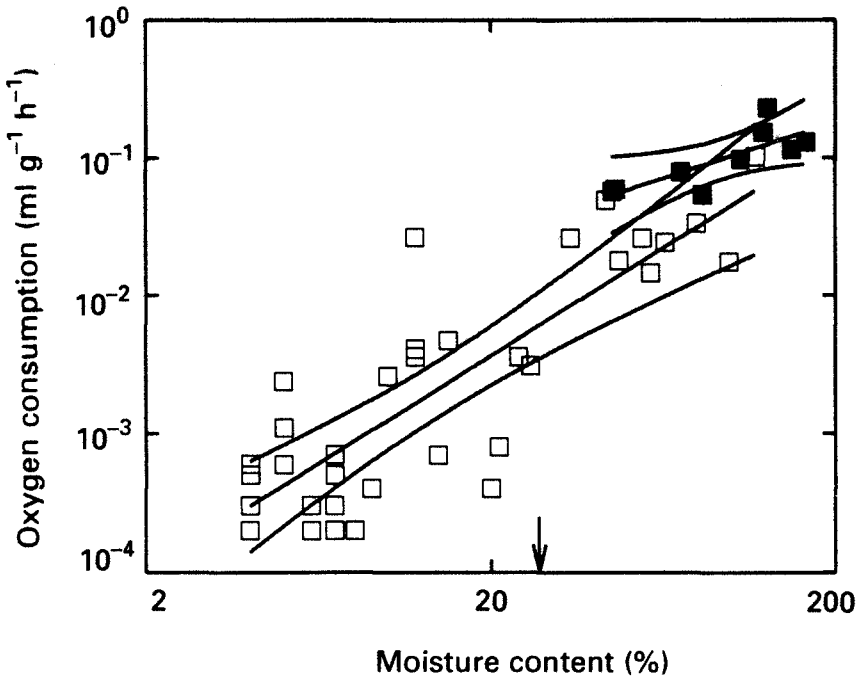

Figure 1. Oxygen consumption rate per gram dry mass as a function of seed moisture content (dry basis). The regression lines and 95 % confidence intervals of air-dry seeds (open squares) and continuously moist seeds (solid squares) are shown. Both regressions were highly significant (Table 2). An arrow indicates 28 % moisture content (MC), which was used to reclassify seeds as moist (> 28 % MC) or dry (≤ 28 % MC) (see text).

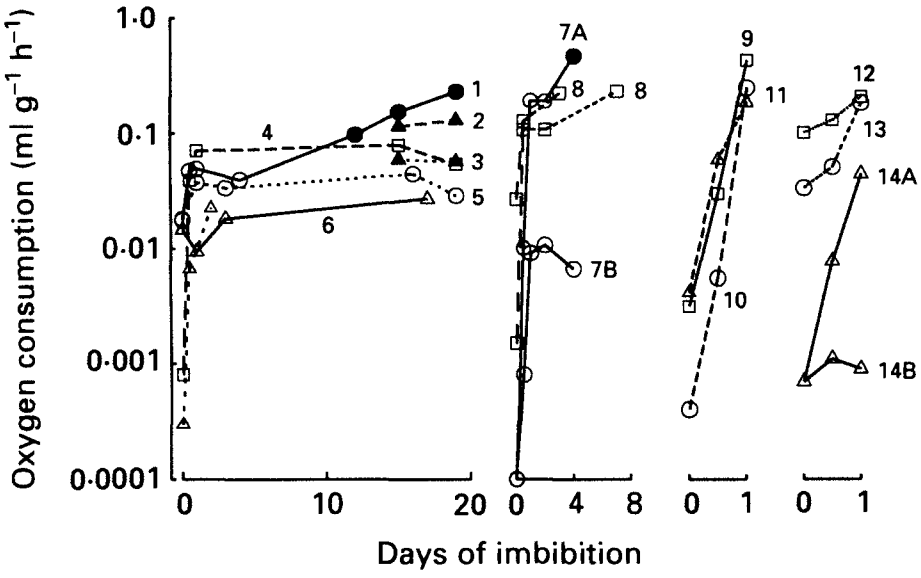

Figure 2. Oxygen consumption rate per gram dry mass as a function of days of imbibition. Solid symbols indicate seeds that are germinating. Species are (1) *Connarus turczaninowii*; (2) *Maripa panamensis*; (3) *Protium panamense*; (4) *Psychotria marginata*; (5) *Gustavia superba*; (6) *Randia armata*; (7) *Rhynchosia pyramidalis*, seeds scarified (A) or not scarified (B); (8) *Jacaranda copaia*; (9) *Piper reticulatum*; (10) *Ochroma pyramidale*; (11) *Psychotria limonensis*; (12) *Protium tenuifolium*; (13) *Pentagonia macrophylla*; and (14) *Ormosia* sp., seeds scarified (A) or not scarified (B).

Table 2. *Regression statistics for the relationship of $\dot{V}_{O_2}$ to moisture content of seeds (percent, dry-mass basis), age of seeds (days), and seed dry mass (g)*

Independent variable	$\dot{V}_{O_2}$ (ml O_2 h^{-1})	Comparison	*b*	*a*	r^2	*t*	d.f.
Moisture content[a]	$\dot{V}_{O_2}$ g^{-1}	All seeds	1·761	0·0000219	0·803	12·93***	41
		Air-dry	1·609	0·0000295	0·687	8·38***	32
		Cont. moist	0·817	0·00239	0·620	3·38*	7
		Dry (⩽ 28 % MC)	1·240	0·0000592	0·262	2·86*	23
		Moist(> 28 % MC)	1·140	0·00380	0·432	3·49**	16
Age[a]	$\dot{V}_{O_2}$ g^{-1}	Air-dry, < 80 d old	−0·561	0·0134	0·120	2·30*	39
		Air-dry, all ages	−0·499	0·0116	0·271	4·18***	47
Seed dry mass[b,c]	$\dot{V}_{O_2}$ $seed^{-1}$	All seeds	0·995	0·00525	0·471	4·99***	28
		Air-dry	0·980	0·00185	0·519	4·76***	21
		Cont. moist	0·748	0·0682	0·941	8·96***	5
		Dry (⩽ 28 % MC)	0·542	0·000120	0·565	4·22***	15
		Moist (> 28 % MC)	0·785	0·0385	0·833	7·41***	11
	$\dot{V}_{O_2}$ g^{-1}	All seeds	−0·00061	0·00483	< 0·001	< 0·01[ns]	30
		Air-dry	−0·026	0·00177	0·001	0·13[ns]	23
		Cont. moist	−0·210	0·0537	0·339	1·60[ns]	7
		Dry (⩽ 28 % MC)	−0·484	0·000104	0·538	4·18***	15
		Moist (> 28 % MC)	−0·176	0·0356	0·194	1·63[ns]	11

[a] All values of $\dot{V}_{O_2}$ for each species were included in each appropriate category of the comparison.

[b] For each species, values of $\dot{V}_{O_2}$ were averaged separately for dry and moist seeds.

[c] To reclassify seeds as dry or moist when moisture content was unknown, we assumed it was ⩽ 28 % if $\dot{V}_{O_2}$ was ⩽ 0·0077 ml O_2 g^{-1} h^{-1}, the value corresponding to 28 % moisture content in the regression of $\dot{V}_{O_2}$ on moisture content (Fig. 1).

$\dot{V}_{O_2}$ is the dependent variable in the regression equation: $Y = aX^b$. Air-dried and continuously moist treatments are discussed in the Methods. Seeds were reclassified by moisture contents (MC) as moist (> 28 % MC) or dry (⩽ 28 % MC) (see text). Levels of significance: *** $P < 0·001$; ** $P < 0·01$; * $P < 0·05$; n.s., $P > 0·10$.

range of critical moisture contents (18–39 %) in our sample of 22 species (Fig. 1). Using the midpoint (28 %) of the range, we reclassified seeds in the continuously moist and air-dry treatments moisture content (MC) as dry (⩽ 28 % MC) or moist (> 28 % MC). The regressions of $\dot{V}_{O_2}$ on moisture content were significant in both dry and moist seeds (Table 2), but the slopes were not significantly different ($F = 0·024$, d.f. = 1, 39, $P > 0·75$). (Moisture content, however, explained a greater proportion of the variation in $\dot{V}_{O_2}$ in the metabolically more active moist seeds than in the less active dry seeds [43 % versus 26 %; Table 2]). The expected change may be obscured because our curve averages across oily and non-oily seeds and orthodox and recalcitrant species. We cannot rule out, however, that the tropical species studied may lack this abrupt change in physiology.

Oxygen consumption during imbibition and germination

The temporal pattern of seed respiration as seeds imbibe water and germinate is divided into four phases (Bewley & Black, 1978; Pradet, 1982). Respiration rapidly increases as seeds imbibe water (Phase I), remains nearly constant for a time thereafter (Phase II, the lag phase), then rapidly increases again as seeds germinate (Phase III). Post-germination changes in respiration (Phase IV) are variable. Phase II is sometimes absent. Respiration rates and the lengths of each phase differ widely among the few species studied, but the causes of these differences are not well understood (Bewley & Black, 1978).

We used the data from continuously moist seeds and rewetted seeds to construct the time course of $\dot{V}_{O_2}$ during the first 4 to 19 days in six species, during the first day only in six additional species, and during germination (Phase III) of two species (Fig. 2). Phase I generally lasted 1–2 days but sometimes 3 days (Fig. 2). Although many species did not germinate (and begin Phase III) during our respiration measurements (Fig. 2), mean time until germination ranged from 3–111 days (Garwood, unpublished data). Thus, the length of the lag phase (Phase II) in these nondormant tropical seeds varied greatly among species and was often much longer than that of nondormant temperature crop species, which usually lasts a few hours or days (Bewley & Black, 1978).

Respiration during Phase I increased more in seeds with initially low $\dot{V}_{O_2}$ than in those with high rates (Fig. 2). This is primarily an effect of seed moisture content. Initial seed moisture content was negatively correlated ($r = -0·69$, d.f. = 11, $P < 0·01$, log-transformed) with the relative increase in respiration during Phase I [estimated using the ratio

($\dot{V}_{O_2}$ of seeds after < 1 day of imbibition)/($\dot{V}_{O_2}$ of air-dried, nonimbibed control seeds)]. $\dot{V}_{O_2}$ increased only 2–5-fold during Phase I in seeds of *Pentagonia macrophylla*, *Randia armata*, *Protium tenuifolium*, *Gustavia superba*, and *Psychotria marginata*, which have high moisture contents (64–117 % in 2–3 day old seeds) and are probably recalcitrant. This small increase indicates that the metabolic machinery is already functional or nearly so at the time of dispersal. In contrast, $\dot{V}_{O_2}$ increased 1–3 orders of magnitude during Phase I in seeds of *Jacaranda copaia*, *Ochroma pyramidale*, and *Rhynchosia pyramidalis*, which have low moisture contents (< 24 % in 2–3 day old seeds). *J. copaia* and *O. pyramidale* have orthodox seeds with facultative dormancy; *R. pyramidalis* has hard-coated seeds.

$\dot{V}_{O_2}$ during Phase II was not considerably higher or lower in species that required the longest times to germinate (*R. armata* and *P. marginata*) than in those that germinated more quickly (*G. superba*, *Connarus turczaninowii*, *Maripa panemensis*, and *Protium panamense*).

Changes in oxygen consumption during drying

Temporal changes in $\dot{V}_{O_2}$ during drying varied considerably among nine species (Fig. 3), but this variation was not clearly associated with any one factor. $\dot{V}_{O_2}$ and moisture content of seeds drying in the air decreased rapidly through time in some orthodox as well as recalcitrant species (*Ochroma pyramidale* and *Jacaranda copaia* versus *Randia armata* and *Psychotria marginata*; Fig. 3), although seed moisture content (measured on days 2–3) was lower in the orthodox species (12–24 %) than in the recalcitrant species (58–64 %). The initial large decrease in $\dot{V}_{O_2}$ preceded substantial moisture loss in *O. pyramidale*, *J. copaia*, and *R. armata* (Fig. 3), although only the two orthodox species survive drying.

$\dot{V}_{O_2}$ changed little through time in three recalcitrant species (*Gustavia superba*, *Connarus turczaninowii*, *Piper reticulatum*), although the earliest stages of drying were not monitored. In *Gustavia superba*, the largest seeded species (≈ 3 g), $\dot{V}_{O_2}$ remained nearly constant over two months while moisture content dropped by nearly half. (Note that seeds of this species were stored in closed plastic bags to slow drying and extend viability). As expected, both seed moisture content and $\dot{V}_{O_2}$ remained low in the unscarified seeds of *Rhynchosia pyramidalis*, a hard-seed legume. Unexpectedly, $\dot{V}_{O_2}$ in *Miconia affinis*, the species with the smallest seed (≈ 0·2 mg), increased an order of magnitude during drying. (The cause of this increase is not known, but unseen microbial contamination or problems in handling the very small seeds cannot be ruled out.)

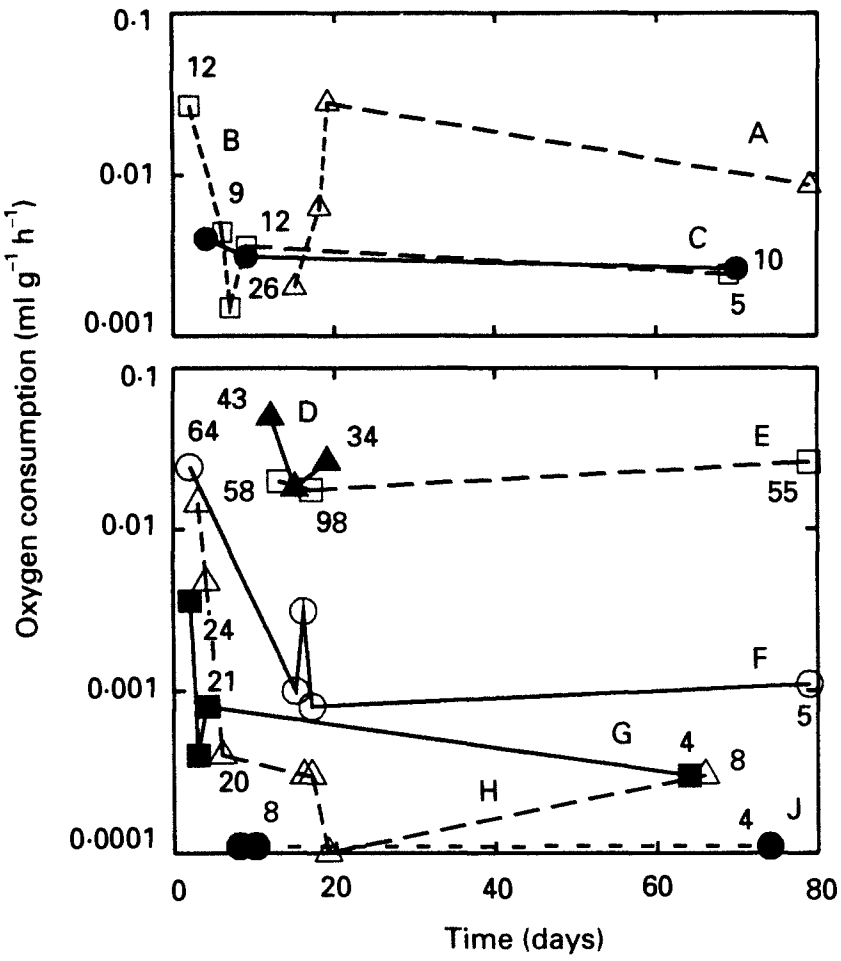

Figure 3. Oxygen consumption rate per gram dry mass ($\dot{V}_{O_2}$) through time as seeds air-dry in the laboratory at ≈ 25 °C. Numbers adjacent to symbols are percent moisture content of seeds. Species for which we have three or more measurements of $\dot{V}_{O_2}$ through time are shown: (A) *Miconia affinis*; (B) *Jacaranda copaia*; (C) *Piper reticulatum*; (D) *Connarus turczaninowii*; (E) *Gustavia superba*; (F) *Psychotria marginata*; (G) *Ochroma pyramidale*; (H) *Randia armata*; and (J) *Rhynchosia pyramidalis*, unscarified seeds.

Seeds of similar age and moisture content often had very different values of $\dot{V}_{O_2}$. For example, seeds of *J. copaia* and *O. pyramidale* air-dried for 60–70 days had similar moisture contents (4–5 %) and masses (≈ 6 mg), but $\dot{V}_{O_2}$ differed by an order of magnitude (Fig. 3). The two pairs of species with the most similar curves of $\dot{V}_{O_2}$ over time (*P. reticulatum* and *J. copaia*; *R. armata* and *O. pyramidale*) each included one recalcitrant and one orthodox species.

Oxygen consumption and seed mass

Rates of oxygen consumption should depend in part on seed size, measured as either volume or mass. Seed volume influences surface area/volume processes, such as rates of exchange of oxygen and other gases with seed tissues as well as rates of imbibition, thereby altering rates of respiration. Seed mass, which is positively correlated with seed volume, is also a partial indicator of the total reserves of the seed. How long the germinating seed and seedling rely on these reserves depends on how fast they are mobilized and respired.

The allometric relationship of respiration and mass has apparently not been investigated in seeds, although it is well-studied in animals (Heusner, 1985). McNab (1988) has emphasized the need to analyse physiologically homogeneous subsets of organisms in allometric studies. We compared dry (⩽ 28 % MC) and moist (> 28 % MC) seeds because

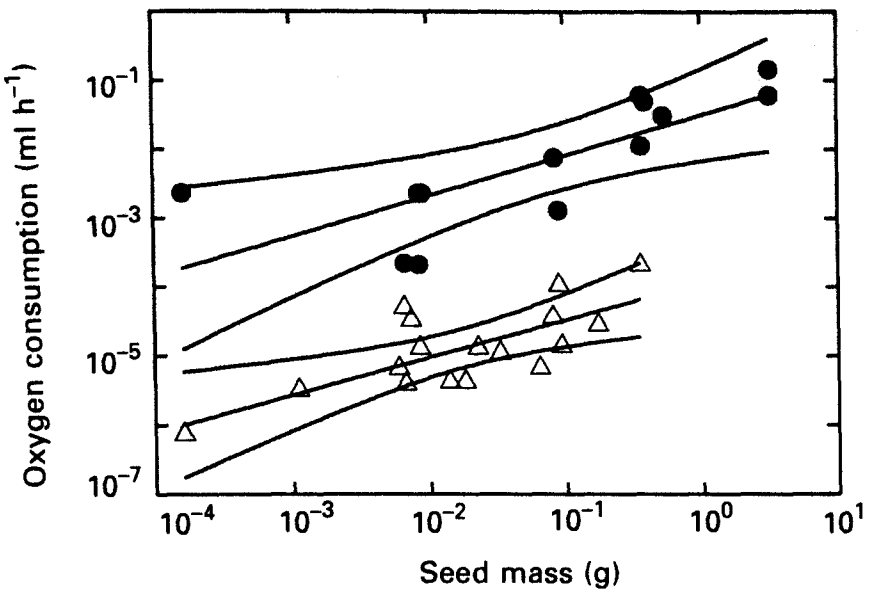

Figure 4. Oxygen consumption rate per seed ($\dot{V}_{O_2}$) as a function of dry seed mass. The regression lines and 95 % confidence intervals of moist seeds (solid circles) and dry seeds (open triangles) are shown. The regression of $\dot{V}_{O_2}$ on seed mass is significant for both dry and moist seeds (Table 2).

seed mass explains more of the variation in mass-specific $\dot{V}_{O_2}$ in these groups than in the original treatment groups (Table 2). For $\dot{V}_{O_2}$ per seed, however, seed mass explains about the same amount of variation in the two groupings (Table 2).

In our sample, $\dot{V}_{O_2}$ per seed significantly increased with seed mass in both dry and moist seeds (Fig. 4; Table 2). Seed mass explained 83 % of the variation in $\dot{V}_{O_2}$ in moist seeds, but only 56 % in dry seeds (Table 2). $\dot{V}_{O_2}$ was ≈ 2 orders of magnitude lower in dry seeds compared to moist seeds over the range of seed sizes sampled, reflecting the greater portion of metabolically inactive tissue in dry than moist seeds. $\dot{V}_{O_2}$ in dry seeds may represent the metabolic activity of a small, hydrated portion within the seeds or the non-metabolic oxidation of seed coat components (Priestley, 1986), which should increase with the surface area of seeds.

Oxygen consumption increases proportionately faster with seed size in moist seeds than in dry seeds (comparison of regression coefficients, $F_s = 2{\cdot}60$, $P < 0{\cdot}05$, d.f. = 13, 17). The exponents of this power function ($Y = aX^b$), 0·78 for moist seeds and 0·54 for dry seeds, are within the range reported for animals and prokaryotes (Heusner, 1985). In dry seeds, seed mass (log-transformed) was not correlated with percent germination ($r = 0{\cdot}28$, $P > 0{\cdot}2$) or percent moisture content of seeds ($r = 0{\cdot}33$, $P > 0{\cdot}2$). Thus, the lower slope of the regression of $\dot{V}_{O_2}$ per seed on the mass of dry seeds is unlikely to be a secondary effect of mass-specific differences in moisture content or viability. Lastly, the mass specific rates of respiration ($Y/X = aX^{b-1}$) are less strongly dependent on seed mass in hydrated, metabolically active seeds (where $b-1 = 0{\cdot}22$) than in drier seeds (where $b-1 = 0{\cdot}46$).

The seeds used in these allometric comparisons were not all in the same stage of germination (e.g., Phases I–III). This physiological heterogeneity might explain some of the residual variation in the allometric relations. In future comparisons, we suggest that seeds in Phase II be used because $\dot{V}_{O_2}$ is relatively stable during this period.

General considerations

The technology now exists to measure rather easily the oxygen consumption of dry, imbibed and germinating seeds and has been modified to measure CO_2 evolution as well (Garwood & Lighton, unpublished data). Respiration of individual seeds, rather than large samples, can be measured over relatively short time periods, especially if the seeds are large or have high moisture content. This is important ecologically. Unlike most agricultural varieties, seeds of many natural species do not germinate synchronously. Respiration rates of heterogeneous samples of seeds in different phases of germination are difficult to interpret.

Comparative studies of seed gas exchange in the three ecologically well-defined groups discussed below should contribute substantially to our understanding of tropical seed germination and its role in the life cycles of tropical species. These groups, which encompass a large proportion of tropical forest species, differ in seed moisture content and seed mass, the two most important factors predicting rates of oxygen consumption among species in our study, or experience different environmental moisture regimes after dispersal.

(1) Many fast-growing, shade-intolerant tropical pioneer species have small, dry, orthodox seeds which may remain viable for several years in the soil beneath the forest canopy (Whitmore, 1983, Garwood, 1989). The seed viability equations, though useful for predicting seed longevity of agricultural species in storage (Roberts & Ellis, 1989), cannot be used to predict potential seed longevity in the soil, because soil temperature and moisture levels change continually. Although seed moisture content will reach an equilibrium value dependent on soil water potential (Roberts & Ellis, 1989), we do not know equilibrium values, the time taken to reach them, average moisture contents over short periods, let alone changes in moisture content over the life span of seeds in the soil, for any tropical species. During periods of high moisture content, seeds may repair cellular damage accumulated during dormancy at low moisture contents (Villiers, 1973; Villiers & Edgcumbe, 1975). At the same time, high respiration rates of fully imbibed dormant seeds would deplete reserves unless the rates decrease with time, as found in a few temperate species (Barton, 1945; Ibrahim *et al.*, 1983). We need to know whether this decrease is common, whether it is caused by changes within the seed or within the surrounding soil microenvironment, and how rapidly seeds can repair damage at different moisture levels.

(2) In seasonal tropical forests, seeds of many species dispersed in the late rainy season are dormant through the remainder of the rainy season and the following dry season and germinate in the next rainy season (Garwood, 1983). Interspecific differences in dry season seed survival were related to the timing, length, and frequency of rainfall during the dry season (see Garwood, 1989, Fig. 1). To understand the interspecific differences in survival, we need further studies of the changing moisture and metabolic status of seeds under different rainfall regimes.

(3) Many slow-growing, shade-tolerant primary forest species have relatively large, moist, recalcitrant seeds that are never components of the persistent soil seed bank, but germinate in the moist shaded understory of the forest (Whitmore, 1983; Garwood, 1989). The difficulty of maintaining recalcitrant seeds in even intermediate-term storage is impeding silvicultural improvement of tropical timber and fruit tree crops and slowing the development of more rational tropical land-use policies. Seed respiration rates should provide new insights into the physiology of recalcitrant species, since viability apparently depends on the availability of oxygen (King & Roberts, 1980; Tompsett, 1983), as is the case with hydrated, dormant orthodox seeds (Ibrahim & Roberts, 1983). Although most recalcitrant species germinate very rapidly, a few species germinate (often asynchronously) only after several months (Garwood, 1989). We need to know whether the physiology of these two groups differs.

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Appendix 1. *Oxygen consumption rate (ml O_2 g^{-1} h^{-1}) of dry, continuously moist and rewetted seeds*

		Dry seeds			Rewetted seeds			Continuously moist seeds		
Species	Seeds per sample	Age (d)	Moist. (%)	Oxygen consumption rate	Age (d)	Imb. time (d)	Oxygen consumption rate	Age (d)	Moist (%)	Oxygen consumption rate
Apeiba membranacea	64	9·5Y	7	0·0003	—	—	—	—	—	—
Arrabidaea patellifera	20	1·8Y	4	0·0006	—	—	—	—	—	—
Cissus sicyoides	30	1·8Y	7	0·0005	—	—	—	—	—	—
Cochlospermum vitifolium	14	9·5Y	6	0·0003	—	—	—	—	—	—
Connarus turczaninowii	5–11	12	43	0·0495	16	< 1	0·0470	12	106	0·0975g
		15	47	0·0179	17	1	0·0492	15	123	0·1539g
		19	34	0·0261	19	4	0·0395	19	127	0·2303g
Diospyros nicaraguensis	7	1·8Y	7	0·0002	—	—	—	—	—	—
Gustavia suberba	1	13	—	0·0198	17	1	0·0372	16	—	0·0445
		16	98	0·0175	19	3	0·0334	19	—	0·0286
		79	55	0·0261	—	—	—	—	—	—
Hemiangium excelsum	18	1·8Y	4	0·0005	—	—	—	—	—	—
Jacaranda copaia	9–30	2	12	0·0266	2	< 1	0·1269	—	—	—
		6	9	0·0044	5	3	0·2228	—	—	—
		7	—	0·0015	7	< 1	0·1077	—	—	—
		9	12	0·0036	9	2	0·1077	—	—	—
		69	5	0·0024	9	7	0·2304	—	—	—
Luffa aegyptiaca	30	1·8Y	6	0·0002	—	—	—	—	—	—
Maripa panamensis	5–14	—	—	—	—	—	—	15	149	0·1151g
		—	—	—	—	—	—	19	164	0·1302g
Miconia affinis	20–240	15	—	0·0020	—	—	—	18	—	1·4690g
		18	—	0·0062	—	—	—	19	—	0·6786g
		19	—	0·0276	—	—	—	—	—	—
		79	—	0·0086	—	—	—	—	—	—
Ochroma pyramidale	20–70	2	24	0·0036	3	< 1	0·0055	—	—	—
		3	—	0·0004	4	1	0·2474	—	—	—
		4	21	0·0008	—	—	—	—	—	—
		64	4	0·0003	—	—	—	—	—	—
		9·5Y	7	0·0007	—	—	—	—	—	—
Ormosia sp.	1–3	3	14	0·0007	3	< 1	0·0011	—	—	—
					4	1	0·0009	—	—	—
					3	< 1f	0·0078	—	—	—
					4	1f	0·0444	—	—	—
Pentagonia macrophylla	18–40	2	79	0·0336	2	< 1	0·0512	—	—	—
		63	5	0·0006	3	1	0·1857	—	—	—
Piper reticulatum	100–600	4	—	0·0040	9	< 1	0·0296	—	—	—
		9	26	0·0031	10	1	0·4281	—	—	—
		70	10	0·0026	—	—	—	—	—	—
Protium panamense	5–17	—	—	—	—	—	—	15	46	0·0591g
		—	—	—	—	—	—	19	45	0·0570g
Protium tenuifolium	5	2	117	0·1016	2	< 1	0·1307	—	—	—
		3	—	0·0967	3	1	0·2086	—	—	—
Psychotria limonensis	10–12	12	15	0·0047	15	< 1	0·0584	—	—	—

Appendix 1 (*cont.*)

		15	12	0·0041	16	1	0·1848	—	—	—
Psychotria marginata	9–46	2	64	0·0243	2	< 1	0·0391	15	71	0·0792
		15	—	0·0010	3	1	0·0705	19	82	0·0540
		16	—	0·0031	17	< 1	0·0188	—	—	—
		17	—	0·0008	—	—	—	—	—	—
		79	5	0·0011	—	—	—	—	—	—
Randia armata	8–30	3	58	0·0145	4	1	0·0094	17	—	0·0268
		4	—	0·0047	6	3	0·0180	—	—	—
		6	20	0·0004	17	< 1	0·0067	—	—	—
		16–17	—	0·0003	19	2	0·0224	—	—	—
		19	—	0·0001	—	—	—	—	—	—
		66	8	0·0003	—	—	—	—	—	—
Rhynchosia pyramidalis	6–30	8–10	8	0·0000	11	< 1	0·0008	—	—	—
		74	4	0·0001	12	1	0·0091	—	—	—
					13	2	0·0107	—	—	—
					15	4	0·0066	—	—	—
					11	< 1f	0·0101	—	—	—
					12	1f	0·1930	—	—	—
					13	2f	0·1904	—	—	—
					15	4f	0·4642g	—	—	—

Seed age (age) is the number of days (or years = Y) between seed collection and a respiration measurement. Moisture content (Moist.) is expressed as a percentage on a dry-mass basis. Imbibition time (Imb. time) is the number of days between wetting of dry seeds and a respiration measurement. Seeds per sample is the total number of seeds within a syringe used for each measurement. Letters after numbers indicate: f, seeds were filed to initiate germination (see Methods); g, seeds were germinating.