

CSIRO Publishing

# FUNCTIONAL PLANT BIOLOGY

*Continuing Australian Journal of Plant Physiology*

FPB

VOLUME 29, 2002  
© CSIRO 2002

**All enquiries and manuscripts should be directed to:**

*Functional Plant Biology*  
CSIRO Publishing  
PO Box 1139 (150 Oxford St)  
Collingwood, Vic. 3066, Australia



**CSIRO**  
PUBLISHING

Telephone: +61 3 9662 7625  
Fax: +61 3 9662 7611  
Email: [publishing.fpb@csiro.au](mailto:publishing.fpb@csiro.au)

Published by CSIRO Publishing  
for CSIRO and the Australian Academy of Science

[www.publish.csiro.au/journals/fpb](http://www.publish.csiro.au/journals/fpb)

## Review:

Energy dissipation in C<sub>3</sub> plants under droughtJaume Flexas<sup>A</sup> and Hipólito Medrano

Laboratori de Fisiologia Vegetal, Departament de Biologia, Instituto Mediterráneo de Estudios Avanzados (CSIC-UIB), Universitat de les Illes Balears, Carretera de Valldemossa Km 7.5, 07071 Palma de Mallorca, Spain.

<sup>A</sup>Corresponding author; email: dbajfs4@ps.uib.es

*This paper originates from a presentation at the Light Stress satellite meeting of the 12th International Photosynthesis Congress, held at Heron Island, Queensland, Australia, August 2001*

**Abstract.** A general quantification of the relative contribution of different light energy dissipation processes to total dissipation under different drought conditions is lacking. Here we compare six studies, including enough data for such a general quantification, to build up a general pattern of the relative importance of several energy dissipation mechanisms in response to drought in C<sub>3</sub> plants. Such a general pattern apparently emerges independently of specific acclimation to drought, but largely dependent on CO<sub>2</sub> availability in the chloroplasts, which may be regulated under drought by adjustments in stomatal and mesophyll conductances. Under irrigation and saturating light, more than 50% of absorbed light is thermally dissipated, while photosynthesis dissipates 20–30% and photorespiration 10–20%. Under mild drought, the contribution of photosynthesis decreases, and that of photorespiration increases in a compensatory manner. During moderate to severe drought, the contribution of both photosynthesis and photorespiration decreases, and thermal dissipation increases up to 70–90% of the total light absorbed. The contribution of other processes, like the Mehler reaction, is shown to be very low under both irrigation and drought. Therefore, in C<sub>3</sub> plants subjected to different degrees of drought, more than 90% of the total energy absorbed by leaves is dissipated by the sum of thermal dissipation, photorespiration and photosynthesis.

## Introduction

Drought is a very common stress, generally characterized by a combination of water shortage, high temperatures and high light intensities (Cornic 1994; Lawlor 1995). Photosynthesis and photosynthetic capacity are progressively decreased under drought, initially due to drought-induced stomatal closure, which reduces CO<sub>2</sub> availability in the chloroplasts. In consequence, a lower light intensity is required to saturate photosynthesis under drought than under well-watered conditions (Cornic 1994; Lawlor 1995). Therefore, at high light intensity, more light is in excess of what can be used for photosynthesis as drought progresses. This may increase the susceptibility of drought plants to photoinhibition (Osmond 1994).

Different processes have been proposed to safely dissipate excess light energy under drought, thus avoiding severe photoinhibition and photooxidation. Osmond and co-workers proposed thirty years ago that photorespiration might serve as a safety valve to dissipate excess electron transport under the low CO<sub>2</sub> concentrations prevailing in chloroplasts during drought (Osmond and Björkman 1972; Powles and Osmond 1978; Osmond *et al.* 1980). Recently, the use of mutants with modified photorespiratory cycle served to demonstrate a photoprotective function of photorespiration (Kozaki and

Takeba 1996) and an increased photorespiration rate in plants subjected to mild drought (Wingler *et al.* 1999), thus establishing a physiological basis for Osmond's early hypothesis. However, the results of Brestic *et al.* (1995) for drought-stressed bean plants suggest that photorespiration does not protect leaves from photoinhibition. More recently, the Mehler reaction was also proposed to serve a function similar to photorespiration (Osmond and Grace 1995; Osmond *et al.* 1997). On the other hand, Demmig *et al.* (1988) were the first to show an increased xanthophyll-cycle-dependent thermal dissipation in *Nerium oleander* plants subjected to drought under high light. A number of studies have since confirmed this trend for many plant species (Cornic 1994; Lawlor 1995). Other minor pathways, such as increased cyclic electron transport around PSII (Canaani and Havaux 1990) and PSI (Katona *et al.* 1992), have been proposed to dissipate excess light during drought.

Despite the fact that operation of the above-mentioned processes under drought is relatively well understood, the relative importance of each is still under debate (Niyogi 1999; Wingler *et al.* 1999; Asada 2000; Badger *et al.* 2000). The main cause for this uncertainty is possibly the fact that few studies are available in which two or more of these processes have been simultaneously evaluated. Also, studies

Abbreviations used: *g*, stomatal conductance; RWC, relative water content;  $\Psi$ , water potential.

on the effects of drought are usually performed under different light conditions, temperatures, water shortage, severity and velocity of water stress imposition — factors that may all interfere with the intensity of light stress and plant response. Nevertheless, a comparative framework is necessary to elucidate which of these mechanisms are of major importance, thus enabling better understanding of the processes that lead to plant acclimation to drought.

The percentages of absorbed light that are used in photochemistry or dissipated as heat can be estimated by chlorophyll fluorescence measurements (for extensive reviews see Krause and Weis 1991; Björkman and Demmig-Adams 1994; Schreiber *et al.* 1994, 1998). The partitioning of photochemistry into different processes is estimated through different approaches. Using a gas exchange system coupled to a mass spectrometer and allowing a leaf to photosynthesize in air containing only  $^{18}\text{O}_2$  permits measurement of rates of net  $\text{CO}_2$  uptake, gross oxygen evolution, and net oxygen uptake (Biehler and Fock 1996; Flexas *et al.* 1999b; Haupt-Herting *et al.* 2001). This may allow direct estimates of rates of photosynthesis, photorespiration, and the Mehler reaction. Alternatively, partitioning of electron transport between photosynthesis and photorespiration can be estimated from combined fluorescence and gas exchange measurements, via models in which the possible contribution of the Mehler reaction is ignored (Epron *et al.* 1995; Valentini *et al.* 1995; Flexas *et al.* 1999a).

The main objective of the present work is to establish a quantitative comparison of the importance of different light dissipation mechanisms for leaf photoprotection under drought conditions. We have pooled data from the available literature on this subject in order to determine if there is a general pattern of response of these mechanisms among  $\text{C}_3$  plants, or if the precise response of each mechanism is dependent on the experimental conditions and species studied.

### Studies analysed and their experimental conditions

Seven complete studies were available in which the effects of water stress on light energy utilization could be quantified as described, although the data needed for some of them were distributed in more than one paper, as indicated in Table 1. The species analysed differed in each study, as did the environmental conditions during imposition of drought. Up to seven different species were studied: cotton (*Gossypium hirsutum* L. cv. Alcala SJ2), French bean (*Phaseolus vulgaris* L.), wheat (*Triticum aestivum* L.), madrone (*Arbutus menziesii* Pursh.), grapevine (*Vitis vinifera* L. cv. Tempranillo and Manto Negro), tomato (*Lycopersicon esculentum* Mill. cv. Moneymaker) and watermelon (*Citrullus lanatus* (Thunb.) Matsum. & Nakai). Ambient temperature ranged from moderate (20–25°C) to high (35–40°C), depending on the studies

(Table 2). Some of the studies were performed under field conditions with high light intensities (1400–1830  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ ), whereas others were performed under controlled conditions with low (340  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ ) to moderate (850  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ ) light intensities (Table 2). It must be noted, however, that the moderate light intensity used in the studies by Biehler and Fock (1996) and Haupt-Herting and co-workers (Haupt-Herting and Fock 2000, 2002; Haupt-Herting *et al.* 2001) may be sufficient to saturate photosynthesis, even in well-irrigated plants acclimated to high light intensity under field conditions (Escalona *et al.* 1999). Therefore, only in the work by Cornic (1994) may light intensity be below that needed to saturate photosynthesis. However, since the bean plants used were grown and acclimated to these low light intensities, it is possible that, for these particular plants, 340  $\mu\text{mol photons m}^{-2} \text{s}^{-1}$  was sufficient to saturate photosynthesis.

Differences in the velocity of drought imposition between studies are remarkable, which may allow analysis of possible acclimation effects of the different dissipation pathways (Table 2). The studies with cotton, madrone and grapevine were performed under field conditions, and drought developed slowly (during 2–3 months). The studies with bean, wheat and tomato were performed in growth chambers, and drought developed more rapidly (from days to weeks). An additional study was included (Miyake and Yokota 2000) in which very severe drought was simulated within minutes by placing leaves in an atmosphere at  $\text{CO}_2$ -compensation point. If acclimation to drought plays a role in distribution of energy dissipation, it is likely we will observe a gradient of responses, from maximum acclimation in field studies to no acclimation in the study of Miyake and Yokota (2000).

### Definition of drought levels for comparison of different studies

Comparing results from different studies is complex, because it requires establishment of categories that allow comparison between similar degrees of water stress. The

**Table 1. References in which data from the different studies analysed were published (see 'References' for further details)**

A number is assigned to each group of references belonging to a single study, which is used in the following tables for identification

Number	Reference(s)
1	Björkman and Schäfer 1989; Björkman and Demmig-Adams 1994
2	Cornic 1994
3	Biehler and Fock 1996
4	Osmond <i>et al.</i> 1997; Badger <i>et al.</i> 2000
5	Flexas <i>et al.</i> 1999a, b; Flexas 2000; Flexas <i>et al.</i> 2002a, b
6	Haupt-Herting and Fock 2000, 2002; Haupt-Herting <i>et al.</i> 2001
7	Miyake and Yokota 2000

**Table 2. Experimental conditions in the studies analysed**

Species analysed are shown, as well as a qualitative description of conditions used for plant growth. Maximum average temperature [T (°C)] and photosynthetically-active photon flux densities [PPFD ( $\mu\text{mol m}^{-2} \text{s}^{-1}$ )] are also provided, together with a qualitative characterization of the velocity of drought imposition (D.I.)

Reference number	Species	Growth conditions	T	PPFD	D.I.
1	Cotton	Field High light	33–40	1830	Slow
2	Bean	Growth chamber Weak light	22	340	Rapid
3	Wheat	Growth chamber Moderate light High humidity (70%)	23	850	Medium (14 d)
4	Madrone	Field High light High temperature	?	Full sunlight	Slow
5	Grapevine	Field High light Low humidity (40–60%)	35–40	1400	Slow
6	Tomato	Growth chamber Moderate light High humidity (70%)	23	850	Rapid (8 d)
7	Watermelon	Growth chamber High light Low humidity (40%)	35	1000	Simulated by lowering CO <sub>2</sub>

fact that water stress is not always assessed using standard parameters complicates such a classification. The most common parameters used to assess the severity of drought are leaf water potential ( $\Psi$ ) and relative water content (RWC) (Cornic 1994; Lawlor 1995). However, we have recently found (Flexas and Medrano 2002; Flexas *et al.* 2002b; Medrano *et al.* 2002a) that light-saturated stomatal conductance ( $g$ ) is a more adequate basis for comparison of the effects of drought among different species and environmental conditions. Basically, the dependence of metabolic processes on  $\Psi$  or RWC is extremely variable among species and experimental conditions, because drought alters metabolic processes through signals other than decreased water availability in cells such as ABA synthesis or decreased hydraulic conductivity. Since  $g$  integrates all these components, the dependence of different photosynthetic parameters on  $g$  is largely constant (Flexas and Medrano 2002; Flexas *et al.* 2002b; Medrano *et al.* 2002a).

However, because the studies reviewed here based their estimations of water stress on different parameters, it has been necessary to establish criteria for definition of the following categories for comparison: control plants, mildly-stressed plants, moderately-stressed plants, and severely-stressed plants. None of the studies presented information about RWC, so a classification based on this parameter was not attempted. Leaf  $\Psi$  was given in four of six studies (Table 3), but it was clearly a poor basis for

comparison. For instance, in isohydric species such as grapevines (Flexas 2000) the leaf  $\Psi$  remains quite unaffected, even when plants are severely stressed (Table 3). In several other species, by contrast, leaf  $\Psi$  markedly decreases as stress becomes more severe, but to a different extent

**Table 3. Leaf water potential ( $\Psi$ ) and light-saturated stomatal conductance ( $g$ ) under different stress levels**  
nd, not determined

Reference	Stress level	$\Psi$ (MPa)	$g$ ( $\text{mmol m}^{-2} \text{s}^{-1}$ )
1	Control	–1.0	185
	Moderate	–1.9	105
	Severe	–2.8	25
2	Control	nd	nd
	Mild	nd	nd
	Moderate	nd	nd
3	Control	nd	nd
	Severe	nd	nd
	Severe	nd	nd
4	Control	–0.7	nd
	Severe	–2.6	nd
	Severe	–2.6	nd
5	Control	nd	nd
	Control	–1.2	350
	Mild	–1.1	125
6	Moderate	–1.1	70
	Severe	–1.2	20
	Severe	–1.2	20
6	Control	–0.6	160
	Severe	–1.8	20

depending on the species and environmental conditions (Table 3). Therefore, whenever data on light-saturated  $g$  were available (Flexas 2000; Haupt-Herting and Fock 2000, 2002; Haupt-Herting *et al.* 2001) or calculable from information given in the original papers (Björkman and Schäfer 1989; Björkman and Demmig-Adams 1994), they were used as the basis for classification of different degrees of stress. This was made according to the rules proposed by Flexas and Medrano (2002). Control plants are considered to be those with  $g > 150 \text{ mmol m}^{-2} \text{ s}^{-1}$ ; mildly-stressed plants with  $g$  between 100 and  $150 \text{ mmol m}^{-2} \text{ s}^{-1}$ ; moderately-stressed plants with  $g$  between 50 and  $100 \text{ mmol m}^{-2} \text{ s}^{-1}$ ; severely-stressed plants with  $g < 50 \text{ mmol m}^{-2} \text{ s}^{-1}$ . For two studies where  $g$  was not available (Biehler and Fock 1996; Osmond *et al.* 1997; Badger *et al.* 2000) only control plants (those well-irrigated plants originally defined as controls by the authors) and severely-stressed plants (those presenting almost null photosynthesis, therefore undoubtedly having  $g < 50 \text{ mmol m}^{-2} \text{ s}^{-1}$ ) were considered. The study by Cornic (1994) was made along a continuous gradient of drought conditions (from control to severe stress). Although  $g$  was not available, it was possible to estimate conditions of control, mild stress, moderate stress and severe stress by referring the values of light- and  $\text{CO}_2$ -saturated photosynthesis given in the original reference to their most probable  $g$  values, according to the relationship defined by Medrano *et al.* (2002a) for a broad range of  $\text{C}_3$  species. Finally, the work of Miyake and Yokota (2000), in which plants were artificially forced to operate at  $\text{CO}_2$ -compensation point, was used to compare with severely-stressed plants only, which also presumably operate at  $\text{CO}_2$ -compensation point (Flexas *et al.* 2002b). Table 3 contains treatments analysed for each reference, with their corresponding leaf  $\Psi$  and light-saturated  $g$  values.

#### **Thermal dissipation accounts for most energy dissipation under saturating light, and becomes increasingly important during moderate to severe drought**

It is clear that under conditions of saturating irradiance, most absorbed light (about 60%) is dissipated thermally, even in the absence of water stress (Table 4). However, under moderate to severe drought this proportion is increased, accounting for as much as 90% of total energy dissipation in some of the analysed studies. Therefore, thermal dissipation may be the most important mechanism for excess energy dissipation in  $\text{C}_3$  plants under drought.

Most of this thermal dissipation may be related to a xanthophyll-dependent dissipation mechanism in light-acclimated plants, with only a minor proportion due to photoinactivation of photosystem units (Björkman and Demmig-Adams 1994; Niyogi 1999). In some of the studies reviewed here, the incidence of drought-induced photosystem photoinactivation was analysed by means of chlorophyll fluorescence analyses in dark-adapted leaves. Drought

did not enhance photoinactivation in any of these studies (Björkman and Schäfer 1989; Cornic 1994; Flexas *et al.* 1998, 1999a, b, 2002a, b; Haupt-Herting and Fock 2000). In contrast, an excellent correlation was demonstrated between the extent of thermal dissipation and the de-epoxidation state of the xanthophyll cycle in one of the studies (Flexas 2000; Medrano *et al.* 2002b).

#### **Electron transport to oxygen is essential for dissipation of excess energy during mild drought**

Even when heat dissipation is the most important pathway for energy dissipation under saturating light, leaf photochemistry accounts for as much as 38% in well-watered plants, of which about 22% is due to photosynthesis and 16% to photorespiration (Table 4). This should not be regarded as a minor amount of energy, since it could represent up to  $800 \mu\text{mol photons m}^{-2} \text{ s}^{-1}$  on clear days. Since drought leads to stomatal closure, thus decreasing  $\text{CO}_2$  availability in the chloroplasts, it has been proposed that increased electron transport to oxygen through both photorespiration and the Mehler reaction may increase, serving as a safe means of excess energy dissipation during drought (Osmond and Grace 1995). Increased electron transport to oxygen during drought has been reported in different studies (Cornic *et al.* 1989; Renou *et al.* 1990; Tourneux and Peltier 1995; Flexas *et al.* 1998, 1999a, b, 2002b; Wingler *et al.* 1999).

The results of the present survey show that under mild drought (i.e.  $g > 150 \text{ mmol m}^{-2} \text{ s}^{-1}$ ) thermal dissipation remains unaffected, the contribution of photosynthesis decreases, and that of photorespiration increases in a compensatory manner. Thus, under mild drought, increased photorespiration should be enough to dissipate excess light as photosynthesis decreases. From moderate to severe drought (i.e.  $g < 150 \text{ mmol m}^{-2} \text{ s}^{-1}$ ), however, contributions of both photosynthesis and photorespiration decrease, possibly as a consequence of decreased ribulose-1,5-bisphosphate availability and/or decreased Rubisco activity (Lawlor 1995). The contribution of photorespiration decreases to a lesser degree than that of photosynthesis, although a part of the estimated photorespiratory contribution could be due to photosynthetic reduction or re-assimilation of  $\text{CO}_2$  evolved from photorespiration (Haupt-Herting and Fock 2000, 2002; Haupt-Herting *et al.* 2001). Notice that only one study (Haupt-Herting and Fock 2000, 2002; Haupt-Herting *et al.* 2001) takes into account the re-assimilation of  $\text{CO}_2$  evolved from photorespiration into the estimation of total photosynthesis. However, and even when these authors showed that re-assimilation increased as drought progressed, the estimated percentages of dissipation do not differ significantly from those of the studies in which re-assimilation was not considered.

Even when the importance of photorespiration as an effective dissipation mechanism is mainly restricted to mild

drought situations it must not be neglected, since mild drought is the more common situation in nature. For instance, accumulation of high light and vapour-pressure deficit at midday leads to temporary water deficits in leaves, which can be considered situations of mild drought in the sense that they partially reduce *g* to mild drought levels. Photorespiration may play an important role in the dynamic regulation of energy dissipation during short periods in which photosynthesis is restricted. Indeed, photorespiration has been shown to increase in the afternoon, especially in drought-stressed plants (Valentini *et al.* 1995; Flexas *et al.* 1999a).

In contrast, the Mehler reaction seems to contribute very little to energy dissipation. Its highest estimated contribution was 9% of total absorbed energy in severely-stressed wheat (Biehler and Fock 1996). It is likely, however, that this value was overestimated. This is due to the fact that in this study, the rate of the Mehler reaction was calculated as the difference between the rates of gross oxygen uptake and photorespiration. Since the latter was estimated by analysing <sup>14</sup>C-content in glycolate after a short period of feeding the leaf with <sup>14</sup>CO<sub>2</sub>, it is likely that any small loss of glycolate during the extraction and analysis procedure leads to

underestimated photorespiration rates and, thus, overestimated Mehler reaction. As discussed by Haupt-Herting and Fock (2002), photoreduction in the Mehler reaction is a non-enzymatic process that depends mainly on electron transport rate, so its overall contribution is likely to decrease as electron transport rate decreases during drought. As discussed recently (Flexas *et al.* 2002b), the small contribution of the Mehler reaction to total dissipation, found in all the studies in which the oxygen isotope technique was used for its measurement, supports the validity of estimations made by the gas exchange–fluorescence modelling approach in other studies, in which the contribution of the Mehler reaction is assumed to be low or null.

#### Energy dissipation under drought seems largely dependent on CO<sub>2</sub> availability in the chloroplasts, rather than on leaf water potential

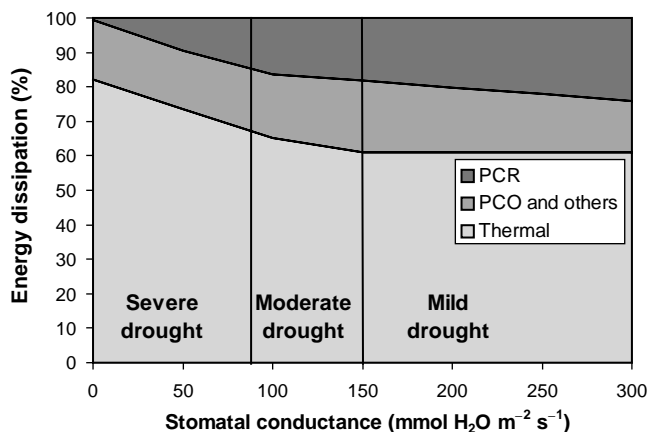
For any given degree of drought, the percentage partitioning of dissipation between photosynthesis, photorespiration and thermal dissipation was similar for the different studies. Even the small differences observed between studies presented no clear effect of velocity of drought imposition or any other environmental condition. Since the different

**Table 4. Literature survey on the effects of drought on dissipation of energy absorbed by leaves as heat (Thermal), photosynthesis (PCR), photorespiration (PCO), Mehler reaction (Mehler) and other processes (Other)**

‘Other’ may include chlorophyll fluorescence, nitrate reduction, zeaxanthin formation, and export out of chloroplasts via the malate–oxaloacetate translocator. The results are expressed as % of total absorbed light for each process. Photorespiration and Mehler are considered together in some of the references, and separately in others. Mehler reaction and ‘Others’ are considered together in one report

Reference number	Species	Drought	Thermal	PCR	PCO	Mehler	PCO plus Mehler	Mehler plus Other	Other
1	Cotton	Control	54	26	—	—	20	—	0
2	Bean	Control	55	35	—	—	10	—	0
3	Wheat	Control	65	22	8	—	—	5	—
4	Madrone	Control	60	12	—	—	28	—	0
5	Grape	Control	64	24	9	< 1	—	—	3
6	Tomato	Control	72	15	6.2	3.5	—	—	3
<b>Average — control</b>			<b>62</b>	<b>22</b>			<b>16<sup>A</sup></b>		
2	Bean	Mild	55	30	—	—	15	—	0
4	Madrone	Mild	60	5	—	—	35	—	0
5	Grape	Mild	64	21	12	< 1	—	—	3
<b>Average — mild drought</b>			<b>60</b>	<b>18</b>			<b>22<sup>A</sup></b>		
1	Cotton	Moderate	70	16	—	—	14	—	0
2	Bean	Moderate	60	18	—	—	22	—	0
5	Grape	Moderate	75	10	10	3	—	—	2
<b>Average — moderate drought</b>			<b>68</b>	<b>15</b>			<b>17<sup>A</sup></b>		
1	Cotton	Severe	82	9	—	—	9	—	0
2	Bean	Severe	75	6	—	—	19	—	0
3	Wheat	Severe	70	10	5	9	—	—	6
4	Madrone	Severe	75	2	—	—	23	—	0
5	Grape	Severe	92	2	3	3	—	—	0
6	Tomato	Severe	91	4	3.5	0.5	—	—	1
7	Watermelon	Comp. point	72	5	13	5	—	—	5
<b>Average — severe drought</b>			<b>78</b>	<b>5</b>			<b>17<sup>A</sup></b>		

<sup>A</sup>Average for PCO, Mehler and other.



**Fig. 1.** Idealized response of percentage contribution of thermal dissipation, photorespiration and CO<sub>2</sub> assimilation in C<sub>3</sub> plants to drought-induced decreases of stomatal conductance. Vertical lines indicate approximate separations between mild, moderate and severe drought.

degrees of drought were described here on the basis of  $g$ , these observations strongly suggest that decreasing CO<sub>2</sub> availability in the chloroplasts triggers most of the drought-induced changes in dissipation pathways. By contrast, no clear relationship was observed between leaf  $\Psi$  and energy dissipation partitioning (not shown, but compare Tables 3 and 4). A dominant role for CO<sub>2</sub> availability in regulation of energy dissipation pathways is also supported by the remarkable similarity observed between the results of Miyake and Yokota (2000) and those of the other studies. The first analysed well-irrigated plants forced to operate at the CO<sub>2</sub>-compensation point, but their results resembled those of severely water-stressed plants, even though  $g$  was probably large in Miyake and Yokota's study due to lowering of atmospheric CO<sub>2</sub>, and very low in drought-stressed plants. Also, similar variations of photochemical and non-photochemical dissipation pathways were obtained when progressive stomatal closure was forced within 1 h in grape leaves (Flexas *et al.* 2002a). According to the average results obtained from this literature survey, an idealized response of different dissipation processes through an entire gradient of  $g$  can be established (Fig. 1).

### Concluding remarks

This analysis shows that, in C<sub>3</sub> plants under saturating light, most of the absorbed energy is funnelled through three different processes: photosynthesis, photorespiration, and thermal dissipation. Of the three, thermal dissipation accounts for more than half of total energy dissipation even in irrigated plants, whereas the contribution of the Mehler reaction and other physiological process is very low at any given degree of stress.

The greatest contribution of photorespiration to total dissipation occurs under mild drought stress. Under moderate

to severe drought stress, the contributions of photosynthesis and photorespiration diminish, and that of thermal dissipation raises progressively, accounting for 80–90% of total energy dissipation in severely-stressed leaves.

Interestingly, the variation of the relative importance of each of these processes to total energy dissipation seems largely independent of the species studied and specific acclimation to drought. In contrast, the available evidence indicates that such a variation depends mainly on CO<sub>2</sub> availability in the chloroplasts and not on leaf water status, so that regulation in response to drought may be driven by adjustments in stomatal and mesophyll conductances to CO<sub>2</sub> diffusion.

### Acknowledgments

We are indebted to Dr Miquel Ribas-Carbó for helpful discussions during manuscript preparation.

### References

- Asada K (2000) The water–water cycle as alternative photon and electron sinks. *Philosophical Transactions of the Royal Society of London Series B* **355**, 1419–1431.
- Badger MR, von Caemmerer S, Ruuska S, Nakano H (2000) Electron flow to oxygen in higher plants and algae: rates and control of direct photoreduction (Mehler reaction) and rubisco oxygenase. *Philosophical Transactions of the Royal Society of London Series B* **355**, 1433–1446.
- Biehler K, Fock H (1996) Evidence for the contribution of the Mehler-peroxidase reaction in dissipating excess electrons in drought-stressed wheat. *Plant Physiology* **112**, 265–272.
- Björkman O, Demmig-Adams B (1994) Regulation of photosynthetic light energy capture, conversion, and dissipation in leaves of higher plants. In 'Ecophysiology of photosynthesis'. (Eds E-D Schulze and MM Caldwell) pp. 17–47. (Springer-Verlag: Berlin)
- Björkman O, Schäfer C (1989) A gas exchange-fluorescence analysis of photosynthetic performance of a cotton crop under high-irradiance stress. *Philosophical Transactions of the Royal Society of London Series B* **323**, 309–311.
- Brestic M, Cornic G, Fryer MJ, Baker NR (1995) Does photorespiration protect the photosynthetic apparatus in French bean leaves from photoinhibition during drought stress? *Planta* **196**, 450–457.
- Canaani O, Havaux M (1990) Evidence for a biological role in photosynthesis for cytochrome b-559, a component of photosystem II reaction center. *Proceedings of the National Academy of Sciences USA* **87**, 9295–9299.
- Cornic G (1994) Drought stress and high light effects on leaf photosynthesis. In 'Photoinhibition of photosynthesis. From molecular mechanisms to the field'. (Eds NR Baker and JR Bowyer) pp. 297–313. (BIOS Scientific: Oxford)
- Cornic G, Le Gouallec JL, Briantais J-M, Hodges M (1989) Effect of dehydration and high light on photosynthesis of two C<sub>3</sub> plants (*Phaseolus vulgaris* L. and *Elastostema repens* (hour.) Hall f.). *Planta* **177**, 84–90.
- Demmig B, Winter K, Krüger A, Czygan FC (1988) Zeaxanthin and the heat dissipation of excess light energy in *Nerium oleander* exposed to a combination of high light and water stress. *Plant Physiology* **87**, 17–24.

- Epron D, Godard D, Cornic G, Genty B (1995) Limitation of net CO<sub>2</sub> assimilation rate by internal resistances to CO<sub>2</sub> transfer in the leaves of two tree species (*Fagus sylvatica* L. and *Castanea sativa* Mill.). *Plant, Cell and Environment* **18**, 43–51.
- Escalona JM, Flexas J, Medrano H (1999) Stomatal and non-stomatal limitations of photosynthesis under water stress in field-grown grapevines. *Australian Journal of Plant Physiology* **26**, 421–433.
- Flexas J (2000) 'Regulation of the photosynthetic processes in response to drought in leaves of *Vitis vinifera* L.' PhD thesis, University of the Balearic Islands, Spain.
- Flexas J, Medrano H (2002) Drought-inhibition of photosynthesis in C<sub>3</sub> plants: stomatal and non-stomatal limitations revisited. *Annals of Botany* **89**, 183–189.
- Flexas J, Escalona JM, Medrano H (1998) Down-regulation of photosynthesis by drought under field conditions in grapevine leaves. *Australian Journal of Plant Physiology* **25**, 893–900.
- Flexas J, Escalona JM, Medrano H (1999a) Water stress induces different photosynthesis and electron transport rate regulation in grapevine. *Plant, Cell and Environment* **22**, 39–48.
- Flexas J, Badger M, Chow WS, Medrano H, Osmond CB (1999b) Analysis of the relative increase in photosynthetic O<sub>2</sub> uptake when photosynthesis in grapevine leaves is inhibited following low night temperatures and/or water stress. *Plant Physiology* **121**, 675–684.
- Flexas J, Escalona JM, Evain S, Gulías J, Moya I, Osmond CB, Medrano H (2002a) Steady-state chlorophyll fluorescence (Fs) measurements as a tool to follow variations of net CO<sub>2</sub> assimilation and stomatal conductance during water-stress in C<sub>3</sub> plants. *Physiologia Plantarum* **114**, 231–240.
- Flexas J, Bota J, Escalona JM, Sampol B, Medrano H (2002b) Effects of drought on photosynthesis in grapevines under field conditions: an evaluation of stomatal and mesophyll limitations. *Functional Plant Biology* **29**, 461–471.
- Haupt-Herting S, Fock H (2000) Exchange of oxygen and its role in energy dissipation during drought stress in tomato plants. *Physiologia Plantarum* **110**, 489–495.
- Haupt-Herting S, Fock H (2002) Oxygen exchange in relation to carbon assimilation in water-stressed leaves during photosynthesis. *Annals of Botany* **89**, 851–859.
- Haupt-Herting S, Klug K, Fock H (2001) A new approach to measure gross CO<sub>2</sub> fluxes in leaves. Gross CO<sub>2</sub> assimilation, photorespiration, and mitochondrial respiration in the light in tomato under drought stress. *Plant Physiology* **126**, 388–396.
- Katona E, Neimais S, Schönknecht G, Heber U (1992) Photosystem I-dependent cyclic electron transport is important in controlling photosystem II activity in leaves under water stress. *Photosynthesis Research* **34**, 449–469.
- Kozaki A, Takeba G (1996) Photorespiration protects C<sub>3</sub> plants from photooxidation. *Nature* **384**, 557–560.
- Krause GH, Weis E (1991) Chlorophyll fluorescence and photosynthesis: the basics. *Annual Review of Plant Physiology and Plant Molecular Biology* **42**, 313–349.
- Lawlor DW (1995) The effects of water deficit on photosynthesis. In 'Environment and plant metabolism. Flexibility and acclimation'. (Ed. N Smirnov) pp. 129–160. (BIOS Scientific: Oxford)
- Medrano H, Escalona JM, Bota J, Gulías J, Flexas J (2002a) Regulation of photosynthesis of C<sub>3</sub> plants in response to progressive drought: stomatal conductance as a reference parameter. *Annals of Botany* **89**, 895–905.
- Medrano H, Bota J, Abadía A, Sampol B, Escalona JM, Flexas J (2002b) Effects of drought on light-energy dissipation mechanisms in high-light-acclimated, field-grown grapevines. *Functional Plant Biology* **29**, 1197–1207.
- Miyake C, Yokota A (2000) Determination of the rate of photoreduction of O<sub>2</sub> in the water–water cycle in watermelon leaves and enhancement of the rate by limitation of photosynthesis. *Plant and Cell Physiology* **41**, 335–343.
- Niyogi KK (1999) Photoprotection revisited: genetic and molecular approaches. *Annual Review of Plant Physiology and Plant Molecular Biology* **50**, 333–359.
- Osmond CB (1994) What is photoinhibition? Some insights from comparisons of shade and sun plants. In 'Photoinhibition of photosynthesis. From molecular mechanisms to the field'. (Eds NR Baker and JR Bowyer) pp. 1–24. (BIOS Scientific: Oxford)
- Osmond CB, Björkman O (1972) Simultaneous measurements of oxygen effects on net photosynthesis and glycolate metabolism in C<sub>3</sub> and C<sub>4</sub> species of *Atriplex*. *Carnegie Institution of Washington Yearbook* **71**, 141–148.
- Osmond CB, Grace SC (1995) Perspectives on photoinhibition and photorespiration in the field: quintessential inefficiencies of the light and dark reactions of photosynthesis. *Journal of Experimental Botany* **46**, 1415–1422.
- Osmond CB, Winter K, Powles SB (1980) Adaptive significance of carbon dioxide recycling during photosynthesis in water stressed plants. In 'Adaptation of plants to water and high temperature stress'. (Eds NC Turner and PJ Kramer) pp. 137–154. (Wiley Interscience: New York)
- Osmond CB, Maxwell K, Björkman O, Badger M, Leegood R (1997) Too many photons: photorespiration, photoinhibition and photooxidation. *Trends in Plant Science* **4**, 119–121.
- Powles SB, Osmond CB (1978) Inhibition of the capacity and efficiency of photosynthesis in bean leaflets illuminated in a CO<sub>2</sub>-free atmosphere at low oxygen: a possible role for photorespiration. *Australian Journal of Plant Physiology* **5**, 619–629.
- Renou J-L, Gerbaud A, Just D, André M (1990) Differing substomatal and chloroplastic concentrations in water-stressed wheat. *Planta* **182**, 415–419.
- Schreiber U, Bilger W, Neubauer C (1994) Chlorophyll fluorescence as a noninvasive indicator for rapid assessment of *in vivo* photosynthesis. In 'Ecophysiology of photosynthesis'. (Eds E-D Schulze and MM Caldwell) pp. 49–70. (Springer-Verlag: Berlin)
- Schreiber U, Bilger W, Hormann H, Neubauer C (1998) Chlorophyll fluorescence as a diagnostic tool: basics and some aspects of practical relevance. In 'Photosynthesis. A comprehensive treatise'. (Ed. AS Raghavendra) pp 320–336. (Cambridge University Press: Cambridge)
- Tourneau C, Peltier G (1995) Effect of water deficit on photosynthetic oxygen exchange measured using <sup>18</sup>O<sub>2</sub> and mass spectrometry in *Solanum tuberosum* L. leaf discs. *Planta* **195**, 570–577.
- Valentini R, Epron D, De Angelis P, Matteucci G, Dreyer E (1995) *In situ* estimation of net CO<sub>2</sub> assimilation, photosynthetic electron flow and photorespiration in Turkey oak (*Quercus cerris* L.) leaves: diurnal cycles under different levels of water supply. *Plant, Cell and Environment* **18**, 631–640.
- Wingler A, Quick WP, Bungard RA, Bailey KJ, Lea PJ, Leegood RC (1999) The role of photorespiration during drought stress: an analysis utilising barley mutants with reduced activities of photorespiratory enzymes. *Plant, Cell and Environment* **22**, 361–373.

Manuscript received 30 January 2002, accepted 29 April 2002