Light energy allocation at PSII under field light conditions; how much energy is lost in NPQ-associated dissipation?

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Abstract

In the field, plants are exposed to fluctuating light, where photosynthesis occurs under conditions far from a steady state. Excess energy dissipation associated with energy quenching of chlorophyll fluorescence (qE) functions as an efficient photo-protection mechanism in photosystem II. PsbS is an important regulator of qE, especially for the induction phase of qE. Beside the regulatory energy dissipation, some part of energy is lost through relaxation of excited chlorophyll molecules. To date, several models to quantify energy loss through these dissipative pathways in PSII have been proposed. In this short review, we compare and evaluate these models for PSII energy allocation when they are applied to non-steady state photosynthesis. As a case study, an investigation on energy allocation to qE-associated dissipation at PSII under non-steady state photosynthesis using PsbS-deficient rice transformants is introduced. Diurnal and seasonal changes in PSII energy allocation in rice under natural light are also presented. Future perspective of studies on PSII energy allocation is discussed.

Key words; Photosystem II, Electron transport, Energy dissipation, NPQ, PsbS

Abbreviations: α, the distribution ratio of PAR to PSII: ETR, electron transport rate, Fm (Fm’), maximum fluorescence obtained by a saturating light pulse at predawn (or during daytime under light); Fo (Fo’), minimum fluorescence obtained under the measuring light in the dark (or under light); Ft, steady-state fluorescence under light; Φ_{A}, quantum yield of energy that is inevitably dissipated in antenna during the course of energy transfer among chlorophyll molecules in PSII: Φ_{D}, quantum yield of heat dissipation in PSII antenna: Φ_{D}, quantum yield of excess energy dissipated in PSII core: $\Phi_{f,D}$, quantum yield of basic dissipation in PSII; Φ_{NPQ}, quantum yield of light inducible regulatory dissipation; Φ_{PsbS}, a part of Φ_{NPQ} induced by PsbS: Φ_{qU}, a part of Φ_{NPQ} which could be induced in the absence of PsbS: Φ_{II} , quantum yield of electron transport in PSII; NPQ, non-photochemical quenching of chlorophyll fluorescence; “NPQ”, traditional parameter of NPQ defined as (Fm-Fm’/Fm’; PAR, photosynthetically active radiation: PSI, photosystem I: PSII, photosystem II; qE, energy quenching: qI, a part of NPQ ascribed to photoinhibition: qT, a part of NPQ ascribed to state transition

Highlights

- Two models for energy allocation in PSII are compared and evaluated.
- Energy allocation to qE-associated dissipation under non-steady state photosynthesis using PsbS-deficient rice transformants is shown.
- The relevance of energy allocation models in PSII under fluctuating light is discussed.

1. Introduction

In photosystem II (PSII), under high light, only some of the absorbed light energy is used for photosynthetic electron transport, and a large portion of the energy is lost through regulatory thermal dissipation, which can be monitored as non-photochemical quenching [1,2], based on a quenching analysis of chlorophyll fluorescence [3]. The advantages of use of the quenching analysis for various aspects of photosynthesis studies had been reviewed in [4-6]. Thermal dissipation associated with non-photochemical quenching has been shown to be an essential photo-protective mechanism of PSII [for review see 4, 7-10]. However, it is not yet clear how much of the light energy absorbed in PSII is dissipated through this mechanism, since the original parameters for non-photochemical quenching, such as "NPQ" = (Fm-Fm')/Fm' [11], are not based on the quantum yield, and thus they cannot be compared quantitatively with the quantum yield of electron transport expressed as Φ_{II} = (Fm'-Ft)/Fm' [12]. Therefore, attempts have been made to simulate the dissipation associated with non-photochemical quenching on the basis of quantum yield. The first of these attempts was reported by Demmig-Adams et al. [13], and was based on the puddle model of energy transfer, in which the light energy absorbed in antennae chlorophyll is always transferred to the same reaction centers. Later, new simulations based on the lake model of energy transfer, in which the excitation energy of chlorophylls can be exchanged among reaction centers, were proposed independently by Kramer et al. [14] and Hendrickson et al. [15].

Although these models for energy allocation in PSII seem to be good tools for better understanding environmental adaptation in photosynthesis [16-18], there have been few field studies under naturally fluctuating light using these models [19].

In this review, we compare Demmig-Adams model and Hendrickson model using the same data set of chlorophyll fluorescence in PsbS-deficient transformants of rice. We also introduce an example of application of Hendrickson model to monitor diurnal and seasonal changes in energy allocation in PSII in field-grown rice.

2. Models for energy allocation in PSII

Absorbed light energy in PSII is allocated to either electron transport or other dissipative processes (Fig. 1, left panel). Since the quantum yield of energy allocation to electron transport Φ_{II} (= (Fm'-Ft)/Fm') was first proposed by Genty et al. [12], it has become widely accepted in physiological studies. In contrast, there is no clear consensus regarding the fate of the remaining absorbed light energy ($1-\Phi_{II}$). However, it can be said that the absorbed light energy that is not used for the electron transport is considered as energy loss that is dissipated mainly as heat.

In a simple simulation, the amount of absorbed light is linearly correlated with the intensity of illuminated light that is commonly expressed as photosynthetically active radiation (PAR) (Fig. 1 right panel). Absorbed light energy in PSII is generally given as PAR x α, where α is the distribution ratio of PAR to PSII. When the ratio of energy lost by reflection and transmission is assumed to be 0.16, and the absorbed light energy is assumed to be equally distributed between PSII and PSI, α is 0.42 [20, 21], the energy flux to electron transport is calculated as Φ_{II} x PAR x α. Similarly, the flux of energy dissipation is calculated as (1 - Φ_{II}) x PAR x α. Under high light conditions, when the energy flux of electron transport is saturated, the energy flux of dissipation is the main path of energy flow.

Demmig-Adams et al. [13] described for the first time an energy allocation model for the portion of energy that is not used for electron transport, i.e. 1- Φ_{II}. They used a traditional puddle model to separate total dissipation into two parts: energy that is dissipated in antenna, *Dissipation* (*D*) (= Fo'/Fm'), and energy that reaches the reaction center but is not used for electron transport, *Excess* (*E*) (= (Ft-Fo')/Fm'). In this review we express *D* as Φ_D and *E* as Φ_E to clarify that both parameters represent the quantum yields of the corresponding dissipation, which can be compared with Φ_{II} on the same unit basis.

Kramer et al. [14] and Hendrickson et al. [15] presented new models for energy allocation in PSII using a lake model of energy transfer. These models categorize total dissipation (1- Φ_{II}) according to its origin, i.e., the basal intrinsic non-radiative decay of excited chlorophyll (Φ_{NO} in the Kramer model or $\Phi_{f,D}$ in the Hendrickson model) and light-dependent regulative dissipation (Φ_{NPQ}), including the energy dissipation that is associated with the xanthophyll cycle qE. The two models are not identical, but give very similar results. Kasajima et al. [22] showed that both models are different expressions of the same model. The model of Hendrickson et al. seems to be more practical for measurements under natural light, since this model did not require the measurement of Fo', which is difficult to be measured , especially under field conditions; in this model, $\Phi_{f,D}$ is calculated as Ft/Fm and Φ_{NPQ} as $1-(\Phi_{II} + \Phi_{f,D})$.

3. Energy allocation at PSII in PsbS-deficient rice under semi-steady state conditions

Energy dissipation associated with non-photochemical quenching can be further divided into subcategories because non-photochemical quenching has been assumed to be induced by at least three independent factors: qE (energy quenching), qT (non-photochemical quenching associated with a state transition) and qI (non-photochemical quenching associated with photoinhibition), as deduced from three phasic relaxation kinetics in the dark [23-25], although molecular basis for qT and qI has not been clear. The most well-studied mechanism is qE, which depends on the presence of the PsbS polypeptide [26], and is associated with the xanthophyll cycle [27]. Dissipation associated with qE is considered to be an important regulatory process for photoprotection of PSII because it is triggered by the light-induced generation of a pH gradient across thylakoid membranes [for review see 4, 8]. Extensive studies have shown that PsbS and the xanthophyll cycle are especially important for the induction phase of qE under a dark-light transition, while under steady-state photosynthesis, qE is maintained without PsbS or the xanthophyll cycle [10, 28-35]

In a study with rice transformants, in which *psbS* genes were silenced by RNAi, Ishida et al. applied these models for energy allocation in PSII to quantify PsbS-dependent energy dissipation [36]. They first compared energy allocation calculated with both models in wild type and PsbS-deficient lines in a semi-steady state under illumination with fixed light intensities. In the following paragraphs, their results were summarized.

In the Demmig-Adams model, the deficiency in PsbSs resulted in a decrease in Φ_D and an increase in Φ_E, while Φ_{II} did not change. Therefore, energy dissipated by a PsbS-dependent process was categorized as dissipation in antenna (Φ_D). Without PsbS, Φ_D was constant over a wide range of light intensities. This light-independent portion of Φ_D was designated Φ_A, which might represent energy that is inevitably dissipated in antenna during the course of energy transfer among chlorophyll molecules. The decrease in Φ_D in PsbS-deficient lines increased energy input to PSII centers, which, however, was not used for electron transport but rather increased energy dissipation in or around PSII centers (Φ_E).

Using the same data set of chlorophyll fluorescence, Ishida et al. re-calculated energy allocation in PSII by the model of Hendrickson et al. In the Hendrickson model, the deficiency of PsbS resulted in a decrease in Φ_{NPQ} and an increase in $\Phi_{f,D}$. Therefore, PsbS-dependent dissipation was categorized as light-inducible energy dissipation (Φ_{NPQ}). No change was found in Φ_{II}, as in the Demmig-Adams model above. The enhanced flow of energy that was not used for electron transport or dissipated with light-inducible mechanisms stimulated energy discharge as the basal decay of excited chlorophyll molecules.

The energy allocation calculated for both models under saturating light shown in [36] is summarized in Table 1. The data shown here represent energy flow in the induction phase at the dark-light transition. Under steady-state conditions, $\Phi_{f,D}$ and

Φ_{NPQ} in PsbS-deficient lines were much closer to those in the wild type lines (unpublished data). In this table, Φ_{PsbS} is defined as Φ_{NPQ} -associated dissipation induced by PsbS, and Φ_{qU} represents the remaining Φ_{NPQ} -associated dissipation, which could be induced in the absence of PsbS. The former was comprised mainly of rapidly forming qE and the latter was probably comprised of dissipation associated with slowly forming qE [31, 37-39] and that associated with photoinhibition (see below).

There was a considerable difference between the two models regarding the values that represented the PsbS- dependent portion of dissipation (Φ_{PsbS}). The low Φ_{PsbS} in the Demmig-Adams model might be due to the underestimation of Φ_D; in this study, Φ_D was calculated by using theoretically calculated F_o' according to Oxborough and Baker [40], however, it is generally known that illumination with high light is likely to increase the F_o' value [41]. Therefore, the true Fo' might be higher than the calculated value, and as a consequence Φ_D calculated as Fo'/Fm' could be an underestimation.

When the new parameters of energy dissipation Φ_{NPQ} are compared with the traditional parameter “NPQ” (= (Fm-Fm')/Fm'), there is not a correlation between the two parameters which can be expressed by a simple equation even when Fo and Fm values are known. In contrast, Ishida et al. [36] showed that Φ_D is closely associated with “NPQ” (=(Fm-Fm'/Fm'), if the Fo' value is calculated according to [40]:

$$\text{“NPQ”} = (Fm-Fo)/Fo \times \Phi_D/(1-\Phi_D) - 1$$

Therefore, when the energy dissipation associated with the traditional parameter “NPQ” (= (Fm-Fm')/Fm') is incorporated into the quantification model of energy allocation, Φ_D can be a better parameter than Φ_{NPQ}. In this sense, the term “Φ_{NPQ}" is rather confusing.

Although photoinhibition of PSII is not a main topic of this review, it is interesting to consider how we can incorporate photoinhibition-associated dissipation in the energy allocation models at PSII. In the Demmig-Adams model, the intrinsic association of Φ_D and “NPQ” (= (Fm-Fm’)/Fm’) shown above indicated that photoinhibition-associated dissipation was included in Φ_D, since photoinhibition is a factor that induced “NPQ” (= (Fm-Fm’)/Fm’). We can divide total Φ_D into qE and qI based on recovery kinetics in the dark of “NPQ” (= (Fm-Fm’)/Fm’). In the Hendrickson model, it is not clear whether photoinhibition-associated dissipation should be categorized as Φ_{NPQ} or $\Phi_{f,D}$. Kasajima et al. [22] allocated photoinhibition-associated dissipation as Φ_{NPQ}, based on the dark recovery kinetics of “NPQ” (= (Fm-Fm’)/Fm’). However, this simulation is valid only when dissipation associated with photoinhibition is categorized predominantly as Φ_{NPQ}. At present we cannot integrate energy loss associated with photoinhibtion into Hendrickson model properly.

4. Energy allocation at PSII in rice grown under field light conditions

Although the new models for energy allocation in PSII seem to be a good tool for deeper understanding of environmental adaptation of photosynthesis, field studies using these models are limited. Demmig-Adams et al. examined diurnal changes in energy allocation in PSII, showing considerable differences between species and also between sun and shade conditions within a same species [13]. Most of recent studies in fields have used the lake models. One of excellent examples has been shown by Locsciale et al. [42], in which they integrated gas exchange analysis with PSII energy allocation model.

Ishida et al. tried to quantify diurnal energy usage and dissipation in PSII in field-grown rice using Hendrickson's lake model, where absorbed light energy in PSII was allocated to either electron transport Φ_{II}, basal dissipation $\Phi_{f,D}$ or regulative dissipation Φ_{NPQ} [43]. In the following paragraphs, we summarize the data described in [43].

The diurnal changes in energy allocation in PSII were estimated in rice plants cultivated in Kyoto, Japan, from July to September, which corresponds to the rapidly growing period and ripening period. The integrated energy flow in PSII over an entire day and its seasonal (developmental) changes were analyzed quantitatively. From predawn to after sunset, Ft was measured under ambient light in the field, and Fm' induced by a saturating light pulse was measured every 5 min. For this measurement, a monitoring PAM fluorometer designed for field use [44] was employed.

Significant fluctuations of PAR occurred within a day. Φ_{II} responded very rapidly to the sudden fluctuation of PAR, and this change in Φ_{II} was accompanied by an inverse change in Φ_{NPQ}. In contrast, $\Phi_{f,D}$ did not fluctuate.

Upright (vertical) leaves received more sunlight in the ripening phase (September) than in the growing phase (July and August), since the altitude of the sun at noon in Kyoto decreased from 75° to 55°, but a large portion of the absorbed light energy seems to be dissipated as heat via an NPQ-associated mechanism. Further analysis indicated that energy flux to electron transport was saturated under a lower light intensity in September than in July-August, and that the saturating energy flux of the electron flow was also lower in September. As a result, the integrated daily flux of electron transport was smaller in September, even though the integrated light energy absorption in PSII was higher (Fig. 2, calculated from the data set in [43]).

When crop photosynthesis is considered, it had been simulated that upright leaves accept a lower intensity of light, but the leaf area that can accept direct sunlight is greater when the sun is high above the horizon, [45], as in the case of upright leaves in July-August presented here. Under this low light in July-August, the efficiency of energy usage (Φ_{II}) is greater than under saturating light in September, where a large portion of the absorbed energy is lost by heat dissipation. As a consequence, it can be concluded from the energy allocation data described in Fig. 2 that efficient photosynthesis under non-saturating light is dominant in July-August, while that wasteful photosynthesis under saturating light is dominant in September. Therefore, even from the limited data based on a single-point measurement adopted in this study, we can assume that "non-light saturated photosynthesis" in the growing phase results in the efficient use of light energy, which might maximize total flux to photosynthetic electron transport in the whole plant. This is in accordance with an old theoretical simulation, in which upright leaves could show a maximum rate of canopy photosynthesis when the angle of incidence is small [46].

5. Why is $\Phi_{f,D}$ constant?

Dissipation associated with Φ_{NPQ} functions as if to minimize $\Phi_{f,D}$ with changes in Φ_{II} over the short term, diurnally, and seasonally. It has been reported that, under steady-state photosynthesis in plants, $\Phi_{f,D}$ remains at a minimum value regardless of the light intensity [14,15,36]. Interestingly, $\Phi_{f,D}$ also remained at a constant low level in the field study [43], in which PAR fluctuated rapidly, and thus photosynthesis was far from a steady state.

In contrast, $\Phi_{f,D}$ can transiently increase when dark-adapted leaves are illuminated (data not shown). In addition, high $\Phi_{f,D}$ has often been observed in photosynthetic mutants; $\Phi_{f,D}$ in the dark or under low light was high in some high-Fo mutants with impaired PSII [47-50], and a light-induced increase in $\Phi_{f,D}$ was found in PsbS-defective mutants [36]. Does the constantly low $\Phi_{f,D}$ have any physiological relevance? In most cases, a high $\Phi_{f,D}$ might be associated with abnormal PSII or photoinhibited PSII. The origin of $\Phi_{f,D}$ has been suggested to be the constitutive deactivation of excited chlorophyll through thermal and radiative dissipation [14, 15]. Probably, a main fraction of $\Phi_{f,D}$ might be associated with a harmful deactivation pathway that leads to the generation of active oxygen species such as singlet oxygen. In this sense, constantly low $\Phi_{f,D}$ might be beneficial for plant wholesomeness, and the thermal dissipation associated with Φ_{NPQ} could be one of the mechanisms to minimize $\Phi_{f,D}$.

6. Perspective

We show here that energy allocation models at PSII can be powerful tools for understanding the physiological relevance of regulatory mechanisms in PSII. However, the molecular significance of each quantum yield parameter is still unclear. In the

Demmig-Adams model, the terms “antenna” and “core” are used without a clear definition, and therefore the molecular basis that induce Φ_D and Φ_E is not clear. In the Kramer and Hendrickson models, while the theoretical basis for Φ_{NPQ} and $\Phi_{f,D}$ seems to be clearer, the molecular basis for $\Phi_{f,D}$ and Φ_{NPQ} is still not clear. For example, the parameter Φ_{NPQ} is not intrinsically associated with the traditional parameter “NPQ” (=(Fm-Fm’)/Fm’), especially in non-steady state photosynthesis. Before these models can be widely recognized as useful tools, extensive experimental and theoretical studies are needed.

Detailed analyses of fluorescence induction kinetics and spectrophotometry with high time resolution using improved measuring systems are direct ways to clarify mechanisms for thermal dissipation. Alternative promising approach seems to be studies with photosynthetic mutants. At present, we estimate daily changes in energy allocation in PSII by Hendrickson model in psbS-deficient rice under natural light conditions. The preliminary results show that integrated Φ_{NPQ} over an entire day is significantly higher in the psb-S deficient rice than in the wild type, although steady state Φ_{NPQ} is in the same level in both genotypes. These results show an importance of PsbS under fluctuating light conditions. This might be a molecular basis of the declined growth of npq4 (psbS-deficinet) mutant of Arabidopsis [51, 52]. To obtain more insights for molecular identities of the quantum yield parameters, we need to use other photosynthesis mutants.

Field use of energy allocation analysis in PSII using chlorophyll fluorescence shown here can be a convenient nondestructive way to know diurnal and seasonal changes in photosynthetic potential in PSII. For further improvements of this type of analysis to get more generalized vision for canopy photosynthesis, multipoint measurements and simultaneous measurements of PSII yield and CO_2 fixation should be considered.

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References

[1] C.A. Wraight, A.R. Crofts, Energy-dependent quenching of chlorophyll a fluorescence in isolated chloroplasts, Eur. J. Biochem. 17 (1970) 319 –327.

[2] J.-M. Briantais, C. Vemotte, M. Picaud, G.H. Krause, A quantitative study of the slow decline of chlorophyll a fluorescence in isolated chloroplasts, Biochim. Biophys. Acta 548 (1979) 128 –138.

[3] U. Schreiber, U. Schliwa, W. Bilger, Continuous recording of photochemical and non-photochemical chlorophyll quenching with a new type of modulation fluorometer. Photosynth. Res. 10 (1986) 51-62.

[4] P. Horton, A.V. Ruban, R.G. Walters, Regulation of light harvesting in green plants. Annu. Rev. Plant Physiol. Plant Mol. Biol. 47 (1996) 655–684.

[5] K.K. Niyogi, Photoprotection revisited: Genetic and molecular approaches. Annu. Rev. Plant Physiol. Plant Mol. Biol. 50 (1999) 333-359.

[6] K. Maxwell, G.N. Johnson, Chlorophyll fluorescence *a* practical guide. J. Exp. Bot. 345 (2000) 659-668.

[7] K. Asada, The water-water cycle in chloroplasts: scavenging of active oxygens and dissipation of excess photons. Annu. Rev. Plant Physiol. Plant Mol. Biol. 50 (1999) 601-639.

[8] P. Müller, X.-P. Li, K.K. Niyogi, Non-photochemical quenching. A response to excess light energy. Plant Physiol. 125 (2001) 1558–1566..

[9] S. Takahashi, M. Badger, Photoprotection in plants: a new light on photosystem II damage. Trends Plant Sci. 16 (2011) 53-60.

[10] A.V. Ruban, M.P. Johnson, C.D.P. Duffy, The photoprotective molecular switch in the photosystem II antenna. Biochim. Biophys. Acta 1817 (2012) 167-181.

[11] W. Bilger, O. Björkman, Role of the xanthophyll cycle in photoprotection elucidated by measurements of light-induced absorbance changes, fluorescence and photosynthesis in leaves of *Hedera canariensis*, Photosynth. Res. 25 (1990) 173–185.

[12] B. Genty, B, J-M. Briantais, N.R. Baker, The relationship between the quantum yield of photosynthetic electron transport and quenching of chlorophyll fluorescence. Biochim. Biophys. Acta 990 (1989) 87-92.

[13] B. Demmig-Adams, W.W. Adams III, D.H. Baker, B.A. Logan, D.R. Bowling, A.S. Verhoeven, Using chlorophyll fluorescence to assess the fraction of absorbed light allocation to thermal dissipation of excess excitation. Physiol. Plant. 98 (1996) 253-264.

[14] D.M. Kramer, G. Johnson, O. Kiirats, G.E. Edwards, New fluorescence parameters for the determination of Q_A redox state and excitation energy fluxes. Photosynth. Res. 79 (2004) 209-218.

[15] L. Hendrickson, R.T. Furbank, R.T., W.S. Chow, A simple alternative approach to assessing the fate of absorbed light energy using chlorophyll fluorescence. Photosynth. Res. 82 (2004) 73-81.

[16] F. Busch, N.P.A. Hüner, I. Ensminger, Biochemical constrains limit the potential of the photochemical reflectance index as a predictor of effective quantum efficiency of photosynthesis during the winter spring transition in Jack pine seedlings, Funct.

Plant Biol. 36 (2009) 1016–1026.

[17] Y. Zhou, J. Zhou, L. Huang, X. Ding, K. Shi, J. Yu, Grafting of Cucumis sativus onto Cucurbia ficifolia leads to improved plant growth, increased light utilization and reduced accumulation of reactive oxygen species in chilled plants. J. Plant Res. 122 (2009) 529-540.

[18] J. Yamazaki, K. Kamata, E. Maruta, Seasonal changes in the excess energy dissipation from photosystem II antennae in overwintering evergreen broad-leaved trees Quercus myrsinaefolia and Machilus thunbergii. J. Photochem. Photobiol. B. 104 (2011) 348-356.

[19] P. Losciale, W.S. Chow, L.C. Grappadelli, Modulating the light environment with peach ‘asymmetric orchard’: effects on gas exchange performances, photoprotection, and photoinhibition. J. Exp. Bot. 61 (2010) 1177-1192.

[20] M. Melis, M. Spangfort, B. Andersson, Light-absorption and electron transport balance between photosystem II and photosystem I in spinach chloroplasts. Photochem. Photobiol. 45 (1987) 129-136.

[21] E. Weis, D. Lechtenberg, Fluorescence analysis during steady state photosynthesis. Philos. Trans. R. Soc. B: Biol. Sci. 323 (1989) 253-268.

[22] I. Kasajima, K. Takahara, M. Kawai-Yamada, U. Uchimiya, Estimation of the relative sizes of rate constants for chlorophyll de-excitation processes through comparison of inverse fluorescence intensities. Plant Cell Physiol. 50 (2009) 1600-1616.

[23] W.P. Quick, M. Stitt, An examination of factors contributing to non-photochemical

quenching of chlorophyll fluorescence in barley leaves. Biochim. Biophys. Acta 977 (1989) 287-296.

[24] G.H. Krause, E. Weis, Chlorophyll fluorescence and photosynthesis: the basics, Annu. Rev. Plant Physiol. Plant Mol. Biol. 42 (1991) 313–349.

[25] J.F. Allen, J. Forsberg, Molecular recognition in thylakoid structure and function, Trends Plant Sci. 6 (2001) 317–326.

[26] X.-P. Li, O. Björkman, C. Shih, A.R. Grossman, M. Rosenquist, S. Jansson, K.K. Niyogi, A pigment-binding protein essential for regulation of photosynthetic light harvesting. Nature 403 (2000) 391-395.

[27] B. Demmig-Adams, Carotenoids and photoprotection in plants: a role for the xanthophyll zeaxanthin. Biochim. Biophys. Acta 1020 (1990) 1-24.

[28] D. Rees, A. Young, G. Noctor, G. Britton, P. Horton, Enhancement of the ΔpH-dependent dissipation of excitation energy in spinach chloroplasts by light-activation; correlation with the synthesis of zeaxanthin, FEBS Lett. 256 (1989) 85–90.

[29] G. Noctor, D. Rees, A. Young, P. Horton, The relationship between zeaxanthin, energy-dependent quenching of chlorophyll fluorescence and the transthylakoid pH-gradient in isolated chloroplasts, Biochim. Biophys. Acta 1057 (1991) 320–330.

[30] P. Horton, A.V. Ruban, Regulation of photosystem II, Photosynth. Res. 34 (1992) 375–385.

[31] A.V. Ruban, A.J. Young, P. Horton, Induction of nonphotochemical energy dissipation and absorbance changes in leaves. Evidence for changes in the state of

the light-harvesting system of photosystem II in vivo, Plant Physiol. 102 (1993) 741–750.

[32] A.V. Ruban, P. Horton, The xanthophyll cycle modulates the kinetics of nonphotochemical energy dissipation in isolated light-harvesting complexes, intact chloroplasts, and leaves of spinach, Plant Physiol. 119 (1999) 531–542.

[33] M.P. Johnson, A.V. Ruban, Arabidopsis plants lacking PsbS protein possess photoprotective energy dissipation. Plant J. 61 (2010) 283-289.

[34] M.P Johnson, A.V. Ruban, Restoration of rapidly reversible photoprotective energy dissipation in the absence of PsbS protein by enhanced delta pH. J. Biol. Chem. 286 (2011) 19973-19981.

[35] Z. Chen, D.R. Gallie, Violaxanthin de-epoxidase is rate-limiting for non-photochemical quenching under subsaturating light or during chilling in Arabidopsis. Plant Physiol. Biochem. 58 (2012) 66-82.

[36] S. Ishida, K. Morita, M. Kishine, A. Takabayashi, R. Murakami, S. Takeda, K. Shimamoto, F. Sato, T. Endo, Allocation of absorbed light energy in Photosystem II to thermal dissipations in the presence or absence of PsbS subunits of rice. Plant Cell Physiol. 52 (2011) 1822-1831.

[37] G. Oquist, W.S. Chow, J.M. Anderson, Photoinhibition of photosynthesis represents a mechanism for the long-term regulation of photosystem II, Planta 186 (1992) 450–460.

[38] A.V. Ruban, P. Horton, An investigation of the sustained component of nonphotochemical quenching of chlorophyll fluorescence in isolated chloroplasts

and leaves of spinach, Plant Physiol. 108 (1995) 721–726.

[39] M. Nilkens, E. Kress, P. Lambrev, Y. Miloslavina, M. Müller, A.R. Holzwarth, P. Jahns, Identification of a slowly inducible zeaxanthin-dependent component of non-photochemical quenching of chlorophyll fluorescence generated under steady-state conditions in Arabidopsis, Biochim. Biophys. Acta 1797 (2010) 466–475.

[40] K. Oxborough, N.R. Baker, Resolving chlorophyll a fluorescence images of photosynthetic efficiency into photochemical and non-photochemical components-calculation of *qP* and *Fv'/Fm'* without measuring *Fo'*. Photosynth. Res. 54 (1997) 135-142.

[41] A.V. Ruban, E.H. Murchie, Assessing the photoprotective effectiveness of non-photochemical chlorophyll fluorescence quenching: a new approach. Biochim. Biophys. Acta 1817 (2012) 977-982

[42] P. Losciale, W.S. Chow, L.C. Grappadelli, Modulating the light environment with peach 'asymmetric orchard': effects on gas exchange performances, photoprotection, and photoinhibition. J. Exp. Bot. 61 (2010) 1177-1192.

[43] S. Ishida, N. Uebayashi, Y. Tazoe, M. Ikeuchi, K. Homma, F. Sato, T. Endo, Diurnal and developmental changes in energy allocation of absorbed light at PSII in field-grown rice. *Plant Cell Physiol.* In press (2013) DOI: 10.1093/pcp/pct169.

[44] A. Porcar-Castell, E. Pfündel, J.F.J. Korhonen, E. Juurola, A new monitoring PAM fluorometer (*MONI-PAM*) to study the short- and long-term acclimation of photosystem II in field conditions. Photosynth. Res. 96 (2008) 173-179.

[45] W.G. Duncan, Leaf angle, leaf area and canopy photosynthesis. Crop Sci. 11 (1971) 482-485.

[46] S. Kuroiwa, Theoretical analysis of light factor and photosynthesis in plant communities (3) Total photosynthesis of a foliage under parallel light in comparison with that under isotropic light condition. J. Agric. Meteorol. 24 (1968) 75-90.

[47] K. Schult, K. Meierhoff, S. Paradies, T. Töller, P. Wolff, P. Westhoff, The nuclear-encoded factor HCF173 is involved in the initiation of translation of the psbA mRNA in *Arabidopsis thaliana*. Plant Cell 19 (2007) 1329-1346.

[48] R. Murakami, K. Ifuku, A. Takabayashi, T. Shikanai, T. Endo, F. Sato, Characterization of an *Arabidopsis thaliana* mutant with impaired *psb*O, one of two genes encoding extrinsic 33-kDa proteins in photosystem II. FEBS Lett. 523 (2002) 138-142.

[49] M. Kishine, A. Takabayashi, Y. Munekage, T. Shikanai, T. Endo, F. Sato, Ribosomal RNA processing and an RNase R family member in chloroplasts of Arabidopsis. Plant Mol. Biol. 55 (2004) 595-606.

[50] N.P. Schultes, R.J.H. Sawers, T.P. Brutnell, R.W. Kruger, Maize high chlorophyll fluorescent 60 mutation is caused by an Ac disruption of the gene encoding the chloroplast ribosomal small subunit protein 17. Plant J. 21 (2000) 317-327.

[51] C. Külheim, J. Agren, S. Jansson, Rapid regulation of light harvesting and plant fitness in the field. Science 297 (2002) 91-93.

[52] C. Külheim, S. Jansson, What leads to reduced fitness in non-photochemical

quenching mutant? Physiol. Plant 125 (2005) 202-211.

Legends for figures

Fig. 1 Energy allocation at PSII. The light energy absorbed at PSII was either used for photosynthetic electron transport or dissipated as heat and chlorophyll fluorescence. The relation between the use and loss of ~~the~~ energy is shown on the basis of quantum yields (left panel) and energy flux (right panel). The energy flux to electron transport (electron transport rate, ETR) is calculated as Φ_{II} x PAR x α. Similarly, the flux of energy dissipation is calculated as (1 - Φ_{II}) x PAR x α.

Fig. 2 Seasonal changes in allocation of the energy flux in PSII calculated from the data set in 2010 shown in Table 1 of [43]. Data are the averages of integrated values over an entire day during the corresponding seasons. In July/August, leaves accept sub-saturating light over a wide area, and use most of the accepted energy for electron transport. In September, leaves accept saturating light over a limited area, and lose a large proportion of the energy as heat.

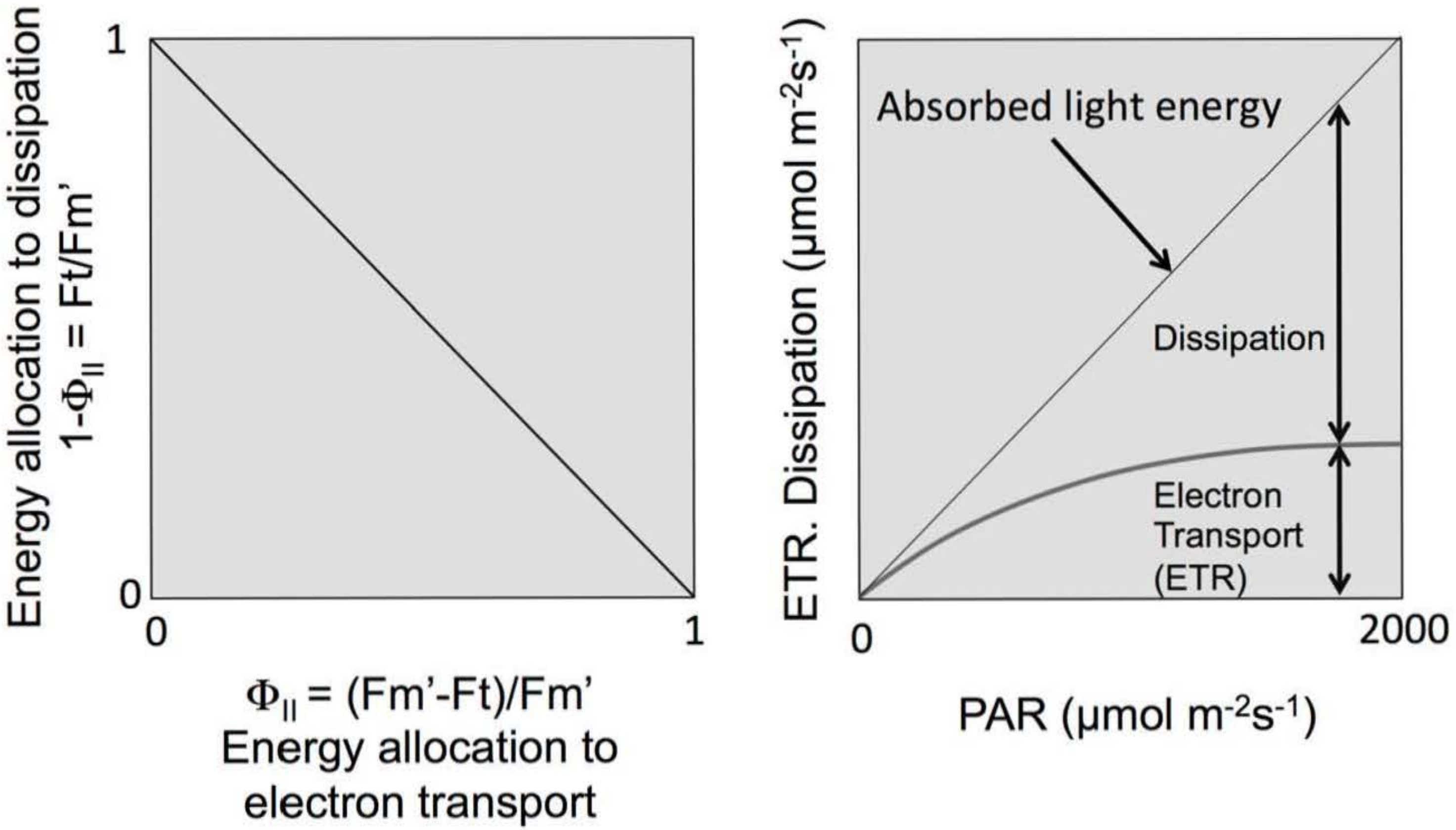
Energy allocation to dissipation
1-Φ_II = Ft/Fm'
1
0
0
1
Φ_II = (Fm'-Ft)/Fm'
Energy allocation to electron transport
ETR. Dissipation (μmol m-2s-1)
Absorbed light energy
Dissipation
Electron Transport (ETR)
0
2000
PAR (μmol m-2s-1)

Fig. 1

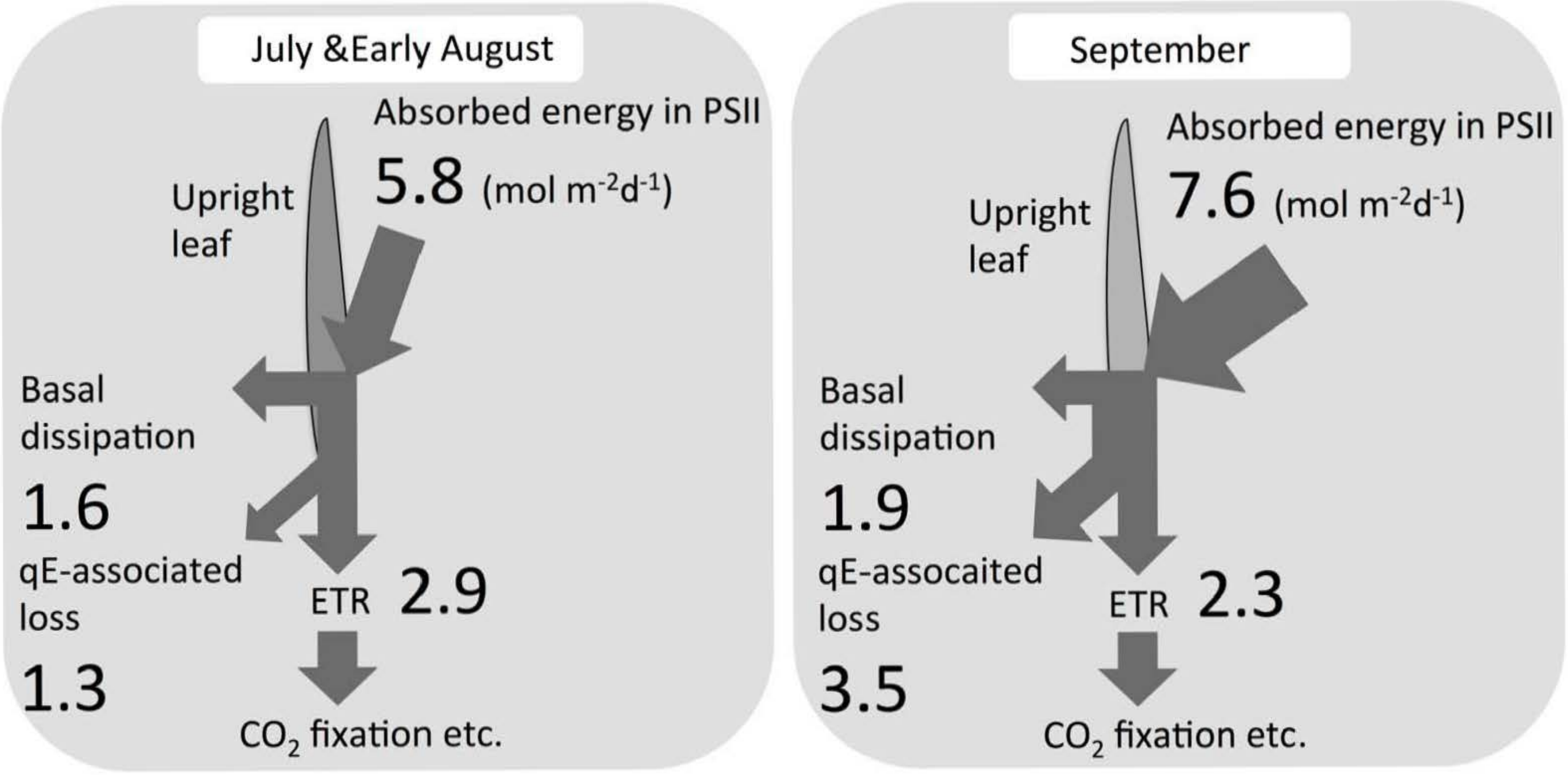
July &Early August
Absorbed energy in PSII
5.8 (mol m-2d-1)
Upright leaf
Basal dissipation
1.6
qE-associated loss
1.3
ETR 2.9
CO2 fixation etc.
September
Absorbed energy in PSII
7.6 (mol m-2d-1)
Upright leaf
Basal dissipation
1.9
qE-assocaited loss
3.5
ETR 2.3
CO2 fixation etc.

Fig. 2

Table 1 Estimated energy allocation under saturating light in rice plants grown in a growth chamber

Estimation with Demmig-Adams model

Φ_{II}				10%
Total dissipation	Φ_{E}			30%
	Φ_{D}	Φ_{NPQ}	Φ_{PsbS}	30%
			Φ_{qU}	little
		Φ_{A}		30%

Estimation with Hendrickson model

Φ_{II}			10%
Total dissipation	$\Phi_{f,D}$		30%
	Φ_{NPQ}	Φ_{PsbS}	50%
		Φ_{qU}	10%