REVIEW PAPER

Role of blue and red light in stomatal dynamic behaviour

Jack S.A. Matthews*, Silvere Vialet-Chabrand# and Tracy Lawson*,
School of Life Sciences, University of Essex, Wivenhoe Park, Colchester CO4 3SQ, UK

*Correspondence: tlawson@essex.ac.uk

Received 24 October 2019; Editorial decision 10 December 2019; Accepted 19 December 2019

Editor: John Evans, Research School of Biology, Australian National University, Australia

Abstract

Plants experience changes in light intensity and quality due to variations in solar angle and shading from clouds and overlapping leaves. Stomatal opening to increasing irradiance is often an order of magnitude slower than photosynthetic responses, which can result in CO2 diffusional limitations on leaf photosynthesis, as well as unnecessary water loss when stomata continue to open after photosynthesis has reached saturation. Stomatal opening to light is driven by two distinct pathways; the ‘red’ or photosynthetic response that occurs at high fluence rates and saturates with photosynthesis, and is thought to be the main mechanism that coordinates stomatal behaviour with photosynthesis; and the guard cell-specific ‘blue’ light response that saturates at low fluence rates, and is often considered independent of photosynthesis, and important for early morning stomatal opening. Here we review the literature on these complicated signal transduction pathways and osmoregulatory processes in guard cells that are influenced by the light environment. We discuss the possibility of tuning the sensitivity and magnitude of stomatal response to blue light which potentially represents a novel target to develop ideotypes with the ‘ideal’ balance between carbon gain, evaporative cooling, and maintenance of hydraulic status that is crucial for maximizing crop performance and productivity.

Keywords: Blue light, guard cells, mesophyll, osmoregulation, photosynthesis, red light, signalling, stomata

Introduction

Stomata control the flux of CO2 into the leaf and water lost through transpiration, and are crucial in maintaining plant water status, leaf temperature, and photosynthetic rates, depending on the current needs of the plant. The surface of most leaves is effectively impermeable to water and CO2; therefore, most of the CO2 fixed and water lost by plants must pass through stomatal pores (Cowan and Troughton, 1971; Caird et al., 2007; Jones, 2013), with stomata controlling the majority of gas exchange between the leaf and external environment, despite typically occupying only a small proportion (0.3–5%) of the leaf surface (Morison, 2003). The capacity of stomata to allow CO2 into or water out of the leaf is known as stomatal conductance (gs), with stomatal behaviour leading to alterations in stomatal aperture and therefore diffusional fluxes. Stomatal aperture is governed by changes in guard cell (GC) volume and turgor pressure driven by alterations in osmotic potential (Willmer and Fricker, 1996; Blatt, 2000; Chen et al., 2012), and, along with stomatal density, determines gs. Adjustment in stomatal behaviour is driven by the external environmental (e.g. light) and internal signalling cues (see Lawson and Blatt, 2014), with responses to these signals varying between and within species (Asamama and Söber, 2011; Drake et al., 2013; McAusland et al., 2016; Matthews et al., 2018). In general, stomatal opening is triggered by increasing light or temperatures (up to an optimum), low CO2, and low vapour pressure deficit (VPD), whilst closure is driven by the reverse; decreasing light, extreme low or high temperatures, high CO2, and high VPD (Raschke, 1975; Outlaw, 2003; Ainsworth and Rogers, 2007; Bernacchi et al., 2007; Matthews and Lawson, 2019). In response to changes in these environmental cues, various ion and solute channels in GCs are...
activated via a signalling cascade, triggering the uptake or release of ions and solutes that modify the osmotic and water potential of the cell, leading to the uptake or loss of water, changes in turgor pressure, and, therefore, changes in stomatal aperture (e.g. see Willmer and Fricker, 1996; Shimazaki, 2007; Lawson and Blatt, 2014; Inoue and Kinoshita, 2017). It is well established that high gs can facilitate a higher net photosynthetic rate (A); however, this is at a greater cost of water loss, making plants more vulnerable to water stress or cavitation (depending on the species) (Naumburg and Ellsworth, 2000; Lawson et al., 2010, 2012; Matthews et al., 2017), whereas low gs can limit CO2 diffusion and photosynthetic rates by up to 20% in well-watered C3 species, negatively affecting biomass accumulation and yield (Farquhar and Sharkey, 1982; Barradas et al., 1994; Fischer et al., 1998; Lawson et al., 2010). Low stomatal aperture may also impact evaporative cooling and conservation of leaf temperature in an optimal range, which is important for maintaining photosynthetic rates and therefore has consequences for harvestable yield (Fischer et al., 1998; Fischer and Rebetzke, 2018; Matthews and Lawson, 2019). Although it is well established that a close correlation between gs and A exists, and whilst the exact signalling pathways and mechanisms that support this relationship have not been fully established, several theories have been put forward. This correlation between A and gs is understood to exist to optimize the trade-off between carbon gain and water loss (Wong et al., 1979; Mansfield et al., 1990; Buckley and Mott, 2013; Buckley, 2017), with stomata continually adjusting aperture to balance the requirement for CO2 for photosynthesis against the need to maintain leaf hydration. However, stomatal responses tend to be an order of magnitude slower than photosynthetic responses, and this leads to non-coordinated responses in gs and A (Lawson et al., 2010; McAusland et al., 2016), where lags in behaviour or slow stomatal responses will often lead to a limitation in carbon gain or unnecessary increase in water loss (see Lawson and Viale-Chabrand, 2019). It has therefore been suggested that species with more rapid gs responses to changing environmental conditions will maximize both photosynthesis and water use efficiency (WUE; Lawson et al., 2010; Raven, 2014; Lawson and Blatt, 2014; Viale-Chabrand et al., 2017a; Lawson and Viale-Chabrand, 2019) and that manipulation of stomatal kinetics could be a novel approach to enhancing CO2 uptake, maintaining optimal leaf temperature for carbon assimilation, and improving water use in important crops (Lawson et al., 2010; Faralli et al., 2019), particularly given the predicted changes to the climate (Matthews and Lawson, 2019).

As photosynthesis and stomata do not respond with the same rapidity to changes in light intensity and spectral quality (Shimazaki et al., 2007), short-term fluctuations in light lead to temporal and spatial disconnections between stomatal behaviour and photosynthesis (e.g. Kirschbaum et al. 1988; Viale-Chabrand et al., 2017b; Lawson et al., 2018; Matthews et al., 2018). Although it is possible to detect these spatial and temporal differences in gs and A (e.g. using imaging approaches; see McAusland et al., 2013; Viale-Chabrand and Lawson, 2019), there are still major gaps in our understanding of the impact of this variation on plant carbon gain or WUE and how such patterns relate to differences in light perception, signal transduction, and stomatal behaviour. Therefore, manipulation of stomatal behaviour and/or the mechanisms that coordinate stomatal response to light quality and intensity could provide potential targets for increasing photosynthesis, WUE, and overall plant productivity in the field. However, in order to succeed, more evidence on the mechanisms and signalling pathways associated with stomatal dynamics in response to light quantity and quality is required, as well as an understanding of the influence of the mesophyll and the hierarchy of stomatal responses.

**Diurnal changes in light quality and intensity impact stomatal behaviour**

Plants experience light in a range of intensities and spectral properties, largely due to passing clouds, changes in canopy cover, and self-shading from overlapping leaves, and this produces unpredictable fluctuations in spectral distribution (see Fig. 1) that impact stomatal behaviour, carbon gain, and the diurnal course of WUE (Pearcy, 1990; Chazdon and Pearcy, 1991; Kaiser and Kappen, 2000, 2001; Viale-Chabrand et al., 2016; Matthews et al., 2017, 2018). The path of the sun across the sky affects both the quantity and quality of the light available to plants at any given location. At dawn and towards dusk as solar angle diminishes, sunlight negotiates an increasingly long path through the atmosphere, enhancing atmospheric light absorption and scattering, thus depleting shorter wavelengths of light (Kendrick and Kronenberg, 1994). Furthermore, the contribution of direct radiation relative to diffuse radiation declines, often leading to a pronounced peak in blue light (Urban et al., 2007), changing the light quality and therefore plant response. Changes in the quality of light throughout the day may impact stomatal dynamic response and diurnal behaviour, and therefore affect photosynthetic efficiency and water use through gs, enforced diffusional constraints on A.

![Fig. 1. Diurnal variation in total irradiance (black) and spectral composition: 360–450 nm (purple), 450–500 nm (blue), 500–570 nm (green), 570–591 nm (yellow), 591–610 nm (orange), and 610–760 nm (red) (A); including changes in the ratio of blue:red light, highlighting peaks in blue light at the beginning and end of the day (B). Purple, yellow, and orange lines almost entirely overlap. Spectral measurements were performed using a microspectrometer C12880MA attached to a C13016 circuit and calibrated to provide PAR intensity.](image-url)
Stomatal response to light

Stomata in C₃ and C₄ species open in response to increasing light intensity whilst closure is brought about by reductions in intensity, whereas CAM (crassulacean acid metabolism) species display the opposite response, with stomata opening in darkness for nocturnal CO₂ uptake and closing in the light (Cockburn, 1983). The stomatal opening responses to light can be divided into two distinct pathways: termed the red or mesophyll/photosynthetic response (herein, termed the red light response) and the GC-specific blue light response (Zeiger, 1983; Assmann and Shimazaki, 1999; Roelfsema and Hedrich, 2005; Shimazaki et al., 2007; Doi et al., 2015; Inoue and Kinoshita, 2017). The red light response occurs at high fluence rates and saturates at similar intensities to photosynthesis, and many studies have suggested that response is the primary mechanism linking stomatal behaviour with photosynthetic rates and that it is responsible for the close correlation between A and gₛ (Wong et al., 1979; Ball and Berry, 1982). The blue light stomatal response occurs and is saturated at a low fluence rate (~5–10 μmol m⁻² s⁻¹; Shimazaki et al., 2007), is GC specific, and is thought to be independent of mesophyll photosynthesis. Blue-light-initiated responses are not exclusive to stomata, and other responses are initiated by this signal that are important for optimal performance; including phototropism (see Christie and Briggs, 2001; Briggs and Christie, 2002; Christie, 2007), photomorphogenesis (Lin and Shalitin, 2003), flowering and circadian clock function (Banerjee and Batschauer, 2005), and the directional movement of chloroplasts in the mesophyll and GC complexes (see Haupt, 1999; Banaś et al., 2012).

Although most research on the spectral aspect of stomatal behaviour and photosynthesis focuses on red and blue light, there is also evidence that green light plays a vital role in physiological responses to the environment. It has been reported that plants may use green wavelengths as a crucial signal to determine short-term dynamic responses and long-term developmental acclimation, enabling optimization of resource use efficiency and photosynthesis to available irradiance (Smith et al., 2017). Furthermore, green light has been shown to inhibit blue-light-induced stomatal opening (Talbott et al., 2006; Aasamaa and Aphalo, 2016), potentially to prevent excessive leaf water loss in shade environments when photosynthetic potential is low (Talbott et al., 2006). In this review, we focus on light-stimulated stomatal behaviour and specifically on the red- (photosynthetic) and blue-light-driven responses, what is understood about the different signalling pathways involved, and GC metabolism that facilitates these responses. We further explore how a better understanding of stomatal response to irradiance, and the influence of the mesophyll on these responses, could provide novel targets for the development of plants with improved photosynthetic carbon gain and WUE.

Red light response of stomata

The red-light-driven opening response of stomata resembles the carbon assimilation response to increasing light intensity (Sharkey and Raschke, 1981) and is eliminated by inhibitors of photosynthetic electron transport, including 3-(3,4-dichlorophenyl)-1,1-dimethylurea (DCMU), indicating that it is photosynthesis dependent (e.g. Kuiper, 1964; Sharkey and Raschke, 1981; Tominaga et al., 2001; Olsen and Junttila, 2002; Messinger et al., 2006), suggesting that chlorophyll could be the receptor (Assmann and Shimazaki, 1999; Zeiger et al., 2002). This red light response is considered the primary mechanism linking stomatal behaviour with mesophyll demands for CO₂, although the exact location of the red light signal has not been fully elucidated. There are suggestions that it occurs in the chloroplast, either in the GCs themselves (Zeiger, 1983; Olsen et al., 2002) or in the mesophyll, and that a signal is transferred from the mesophyll to the GCs (Lawson et al., 2014). Several studies have suggested that GCs do not directly sense red light, and instead respond to the supply of CO₂ in the mesophyll (see Fig. 2), coupling A and gₛ via Cᵢ (Mott, 1988; Roelfsema et al., 2002), although other signalling mechanisms have been suggested (discussed below). Mesophyll consumption of CO₂ driven by increasing photosynthetic irradiance reduces [CO₂] in the intercellular air spaces to which stomata respond by opening; low light reduces this consumption and increases the concentration, closing stomata (Mott, 1988). Support for a Cᵢ-driven response, independent of a specific GC response, was provided by Roelfsema et al. (2001, 2002), who showed that a beam of blue light, but not red, induced changes in membrane potential in the GCs, altering K⁺ transport across the plasma membrane (Roelfsema et al., 2001). Only when [CO₂] around the GCs was altered under red light did these authors report that cells were hyperpolarized in CO₂-free air, and switched to being depolarized when [CO₂] was increased to 700 μmol mol⁻¹, extruding K⁺ which resulted in stomatal closure (Roelfsema et al., 2002). From these studies, it was concluded that GCs do not respond to red light directly but to the indirect changes in Cᵢ (Roelfsema et al., 2002). This is further supported by studies that have shown that stomatal opening to red light is mediated in part by a component of the low CO₂ signalling network (HT1; HIGH LEAF TEMPERATURE1; Hashimoto et al., 2006; Matrosova et al., 2015), and that high [CO₂] activates the release of Cl⁻ ions from GCs via S-type anion channels (such as SLAC1; Yamamoto et al., 2016; Kusumi et al., 2017; Zhang et al., 2018), inducing stomatal closure (Fig. 2). Further studies showed that stomata of Vicia faba (Roelfsema et al., 2006) and wheat (Karlsson, 1986a) did not respond to red light when treated with the carotenoid inhibitor norflurazon (which results in al-bino leaves that lack functional green chloroplasts). Similarly, no response was observed in the white area of variegated Hedera helix (Aphalo and Sanchez, 1986) and Chlorophytum comosum (that do contain photosynthetically active chloroplasts in the GC) when illuminated with red light (Roelfsema et al., 2006). However, in both cases, stomata still responded to blue light, [CO₂], and abscisic acid (ABA); therefore, it was concluded that photosynthetically active mesophyll is required for stomatal red light responses via changes in Cᵢ. However, many other studies have suggested that stomatal aperture responses to Cᵢ are too small to account for the changes observed in gₛ in response to light (Raschke, 1975; Sharkey and Raschke, 1981; Farquhar and Sharkey, 1982), and it has been reported that under red light, gₛ increases even when Cᵢ is held constant (Messinger et al., 2006; Lawson et al., 2008; Wang and Song, 2008), questioning Cᵢ as the
main driver of $A$ and $g_s$ coordination. Furthermore, studies on transgenic plants in which expression levels of several enzymes associated with electron transport or the Calvin cycle were manipulated leading to reduced photosynthetic rates demonstrated that stomata opened in response to light regardless of the higher $C_i$ values observed (von Caemmerer et al., 2004; Baroli et al., 2008; Lawson et al., 2008). The lack of coordination between $A$ and $g_s$ in these transgenic plants raised questions concerning the mechanism(s) that links these two processes. Other studies have suggested that an as yet unidentified signal originating in the mesophyll is potentially sensed by GCs activating a stomatal response. Wang and Song (2008) demonstrated that stomata on the abaxial leaf surface opened more widely when the leaf was irradiated from the adaxial rather than the abaxial side with $C_i$.
kept constant, supporting the idea of a direct mesophyll signal influencing stomatal behaviour. Lee and Bowling (1992, 1995) were the first to propose an aqueous metabolic signal, with potential candidates such as ribulose bisphosphate (RuBP), ATP, NADPH, malate, and sugar (Hedrich and Marten, 1993; Hedrich et al., 1994; Zeiger and Zhu, 1998; Tominaga et al., 2001; Lee et al., 2008; Fujita et al., 2013, 2019). However, further research has suggested a gaseous vapour phase ion (Mott et al., 2008; Sibbernse and Mott, 2010; Mott and Peak, 2013), sucrose metabolism (Lu et al., 1997; Outlaw, 2003; Kang et al., 2007), and even GC photosynthesis itself (Lawson et al., 2003, 2014, 2018; Lawson, 2009).

**Guard cell osmoregulation in response to red light**

The red light response (as with all stomatal responses) requires changes in osmotic potential in the GC, driven by the accumulation or loss of ions such as K⁺ and/or sugar accumulation (see reviews by Shimazaki et al. 2007; Lawson, 2009) to change water flux and therefore pore width (Fig. 2). Early research demonstrated that potassium accumulation is the result of red light activation of the plasma membrane proton pump (Serrano et al., 1988; Olsen et al., 2002), with ATP supplied by photophosphorylation in the GC chloroplasts (Shimazaki and Zeiger, 1985, Tominaga et al. 2001), although subsequent patch-clamp experiments could not replicate red light activation of the proton pump (Taylor and Assmann, 2001). Sugars as well as K⁺ have also been reported to accumulate in response to red-light-induced stomatal opening (Talbott and Zeiger, 1998; Olsen et al., 2002), provided either by starch breakdown (Outlaw and Manchester, 1979), import from the mesophyll (e.g. Lu et al., 1995), or directly through GC photosynthesis (see review by Lawson, 2009). Although early studies on red-light-induced stomatal opening in epidermal peels reported high GC sucrose concentrations (Talbott and Zeiger, 1993), suggesting that the supply must be from GC photosynthetic carbon assimilation (Talbott and Zeiger, 1998), other studies have reported that GC photosynthesis is insufficient to produce sucrose required for osmoregulation (e.g. Outlaw, 1989; Reckmann et al., 1990). This led to the hypothesis that apoplastic sucrose fixed in the mesophyll cells travels to the GCs via the transpiration stream (Lu et al., 1995; Kang et al., 2007), where it can be imported into the GCs via sucrose-mediated H⁺ symporter mechanism(s) (Daloso et al., 2016) and act to open or close stomata or replace GC carbon stores (Lu et al., 1997; Kelly et al., 2013). Apoplastic sucrose accumulation at the GC has been proposed to initiate stomatal closure and provide a mechanism to coordinate A and gₛ (Lu et al., 1997; Ewert et al., 2000; Outlaw and De Vlieghere–He, 2001). Outlaw and colleagues suggested that when mesophyll cells produce more sugar than can be loaded into the phloem, any excess will be carried to the GCs to reduce stomatal aperture (see Outlaw, 2003). This is supported by the numerous studies that have demonstrated sugar import into GCs (Reddy and Das 1986; Rittle et al., 1999; Stadler et al., 2003; Weise et al., 2008; Bates et al., 2012; Daloso et al., 2015; Antunes et al., 2017). Kelly et al. (2013) suggested that sucrose arriving at the GC is cleaved in the apoplasts to produce glucose and fructose that is then sensed by hexokinase, which signals stomatal closure response. Although this mechanism may explain the reported decrease in A and gₛ often observed over longer time scales and toward the end of the day (Violet-Chabrand et al., 2017b; Matthews et al., 2018), it cannot explain the short-term coordination of A and gₛ.

**Role of guard cell chloroplasts in the red light response**

It has recently been shown that Arabidopsis mutants with reduced GC chlorophyll content have reduced gₛ, implying that GC photosynthesis is crucial for energetics and stomatal movements (Fig. 2; Azoulay–Shemer et al., 2015). Support for the involvement of GC electron transport in light-induced stomatal opening also comes from work on the ‘crumpled leaf’ mutants, which lack GC chloroplasts. These mutants exhibited reduced levels of GC ATP and stomatal aperture in response to white light (Wang et al., 2014). Wang et al. (2014) also demonstrated that lower ATP levels were observed in epidermal peels incubated (for 2 h in light) in isolation from the mesophyll, compared with peels that had been collected from intact leaf material after the incubation period. These findings strongly suggest that both GCs and mesophyll cells provide ATP for stomatal opening. Therefore, as there is currently no evidence for ATP import into GCs, mesophyll cells are likely to indirectly supply ATP by providing sugars that are utilized by the mitochondria (Wang et al., 2014). The ATP from GC electron transport provides additional energy to that produced by glycolysis and mitochondrial respiration (Vavasseur and Raghavendra, 2005; Daloso et al., 2016), which can be directly used for proton pumping or other metabolic processes involved in osmoregulation for stomatal opening in response to red (and blue) light (see Daloso et al., 2015, 2016; Santellia and Lawson 2016; Santellia and Lunn, 2017). Interestingly, a recent report has demonstrated that red-light-induced plasma membrane H⁺-ATPase phosphorylation correlated with stomatal opening (Yamauchi et al., 2016; Ando and Kinoshita, 2018), a process previously thought to be blue light dependent and under the control of the photoreceptor protein kinases, phototropins (see below). Using knockout mutants of one of the major isoforms of plasma membrane H⁺-ATPase in GCs, ath1-9, Ando and Kinoshita (2018) revealed that red-light-dependent stomatal opening was delayed in whole leaves. An immunohistochemical technique to detect phosphorylation demonstrated that DCMU inhibited plasma membrane H⁺-ATPase phosphorylation and red-light-induced stomatal opening. However, the lack of this response in isolated epidermal peels further suggests that mesophyll photosynthesis is required for the red light response. Furthermore, the authors did not rule out that GC chloroplasts might have the potential to induce partial phosphorylation of the plasma membrane H⁺-ATPase, and that this could be the underlying cause for interspecific differences in red light sensitivity in GCs. Moreover, as electron transport in the GCs and mesophyll chloroplasts is essentially the same, this could be involved in the regulation and coordination of A and gₛ responses (Sharkey and Raschke, 1981; Lawson et al., 2002, 2014; Messinger et al., 2006; Lawson, 2009). Interestingly, the redox state of the
chloroplastic plastoquinone pool (QA) has been put forward as a signal that coordinates red light stomatal responses with the mesophyll at a range of light intensities (Busch, 2014). This was experimentally tested using tobacco mutants with reduced expression levels of PSII subunit S (PsbS), which directly effects the redox state of QA and results in a strong correlation with $g_s$ when measured under a range of light intensities (Glowacka et al., 2018). As tobacco generally lacks the GC-specific blue light response (see below), this enabled a direct relationship, driven by the red or photosynthetic light response, between QA and $g_s$ to be evaluated.

**Stomatal blue light response**

Blue-light-induced stomatal opening has been demonstrated in isolated epidermal peels and GC protoplasts (Zeiger and Helper, 1977); therefore, all the components required for this response are located in the GCs themselves (Kinoshita and Shimazaki, 2002; Ueno et al., 2005; Hayashi et al., 2011) and, unlike the red light response, does not require the involvement of a mesophyll signal (Ando and Kinoshita, 2018). Blue-light-induced stomatal opening is saturated at a low (~5–10 μmol m$^{-2}$ s$^{-1}$) fluence rate, too low to drive photosynthetic carbon gain, and is 20 times more effective at opening stomata than red light (Hsiao et al., 1973; Karlsson, 1986b; Sharkey and Ogawa, 1987; Briggs, 2005). It has been proposed that the stomatal blue light response is important for morning pore opening to facilitate photosynthetic carbon gain early in the diel period, when the irradiance spectrum is enriched in blue wavelengths (Fig. 1; Zeiger, 1984). Additionally, this response could be important in rapid stomatal responses to sun flecks (Iino et al., 1985) to maximize opportunistic periods of photosynthesis (Pearcy, 2007), particularly in understory environments (Chazdon, 1988; Chazdon and Pearcy, 1991).

Although the stomatal blue light response has been reported as not requiring photosynthesis, it has been demonstrated that two kinases found in the GC blue light signalling pathway—CBC1 and CBC2 (CONVERGENCE OF BLUE LIGHT AND CO$_2$)—are actually involved in linking blue light responses (via phototropins) to low CO$_2$ concentrations in the GC (Hiyama et al., 2017; see Fig. 2). This therefore suggests that photosynthesis is indirectly involved, and in fact it has been shown previously that the sensitivity and magnitude of the stomatal response to blue light depend on the intensity of background red light (Karlsson, 1986a). In their review on light regulation of stomatal movement, Shimazaki et al. (2007) reported an increased $g_s$ rate when blue light was applied to a background of red light compared with red alone, and virtually no response to weak blue light was observed when red light was absent, although this response could be species specific. Figure 3 highlights the impact of blue light on the magnitude of $g_s$ compared with red light alone, demonstrating the independent and synergistic behaviour of the two distinct signals.

**Blue light signalling pathway**

The stomatal blue light response is mediated by blue light photoreceptor protein kinases, known as phototropins (phot1 and phot2; Kinoshita and Shimazaki, 2001; Doi et al., 2004; Christie et al., 2007). Under blue light, phototropins within the GCs are activated via autophosphorylation and initiate a signalling cascade that eventually results in stomatal opening (see Fig. 2; Kinoshita and Shimazaki, 2001; Christie et al., 2007; Shimazaki et al., 2007; Inoue and Kinoshita, 2017). The protein kinase BLUE LIGHT SIGNALLING 1 (BLUS1) is directly phosphorylated by the activated phototropins (Takemiya et al., 2013), and has been shown to indirectly transmit a signal to a type 1 protein phosphatase (PP1) and regulatory subunit PRSL1 (Takemiya et al., 2006, 2013; Takemiya and Shimazaki, 2016). This blue-light-driven BLUS1 signal activates plasma membrane H$^+$-ATPase in GCs via phosphorylation of a penultimate C-terminal residue (Thr) and through subsequent binding of a 14-3-3 protein (Shimazaki et al., 2007; Hayashi et al., 2011). Further recent research on the stomatal blue light pathway has identified that a Raf-like (receptor kinase involved in cell cycle regulation) protein kinase, BLUE LIGHT-DEPENDENT H$^+$-ATPase PHOSPHORYLATION (BHP), binds to BLUS1 and forms a signalling complex with phototropins to mediate phosphorylation of plasma membrane H$^+$-ATPase (Hayashi et al., 2017). However, these authors suggest that BHP does not directly phosphorylate the penultimate Thr of membrane H$^+$-ATPase, and another as yet unidentified signalling kinase may exist that directly controls phosphorylation of H$^+$-ATPase, and stomatal opening in the blue light signalling cascade (Hayashi et al., 2017; Inoue and Kinoshita, 2017). Blue-light-activated plasma membrane H$^+$-ATPase initiates hyperpolarization of the membrane and drives H$^+$ transport out of the GC (Shimazaki et al., 2007). This hyperpolarization further activates inward-rectifying K$^+$ channels, resulting in the influx and accumulation of K$^+$ in the cytosol (Lebady et al., 2008; Inoue and Kinoshita, 2017). Transport and accumulation of K$^+$ and the counter-ions Cl$^-$ and malate$^{2-}$ into the vacuole occurs via K$^+/H^+$ exchangers (NHX1 and NHX2), chloride channel c (CLCc), and chloride channel malate transporters (ALMT9) (Jossier et al., 2010; De Angeli et al., 2013; Andrés et al., 2014), which decrease GC water potential, increasing water uptake and turgor pressure, and ultimately leads to stomatal opening (Fig. 2; Inoue et al., 2010; Eisenach and De Angeli, 2017; Inoue and Kinoshita, 2017; Jezek and Blatt, 2017). It has been suggested that among the phototropin-mediated responses, BLUS1 defines signalling specificity of stomatal opening and, as BLUS1 expression is only found in the GC and not in mesophyll cells, is not involved in other important phototropin-mediated responses (including phototropism and chloroplast movement) (Takemiya et al., 2013; Inoue and Kinoshita, 2017), and therefore could be an unexploited target for manipulating stomatal behaviour.

In addition, the phototropin-dependent blue light signalling cascade and activation of plasma membrane H$^+$-ATPase has been reported to be involved in carbon metabolism in GCs. A study by Horrer et al. (2016) revealed a novel pathway of starch degradation involving synergistic activities of β-amylase 1 (BAM1) and α-amylase 3 (AMY3) in GCs. This is in contrast to the mesophyll starch metabolism in which BAM3 is the major isoform and BAM1 has limited involvement (see Horrer et al., 2016). Using the phot1/phot2 double mutant and
the BLUS1 mutant, these authors showed that the blue light signalling pathway was required for starch breakdown, in order to produce maltose which is subsequently turned into malate (which acts as a counter-ion for K⁺ uptake) through glycolysis and the activity of phosphoenolpyruvate carboxylase (PEPc; Horrer et al., 2016). This is in agreement with other studies that have suggested a role for malate and PEPc in GC osmoregulation (Daloso et al., 2016; Santelia and Lunn, 2017). This novel starch degradation pathway proposed by Horrer et al. (2016) highlighted that BAM1 and AMY3 are redox regulated (unlike the starch degradation enzymes associated with mesophyll activity), and suggested that GC electron transport could provide the reduced environment that would support BAM1 and AMY3 activation as well as providing a further connection between increasing light and stomatal opening, and a possible link between red and blue light responses (see below).

**Energetic and ATP supply for the guard cell blue light response**

The fact that the blue light response has been observed in isolated tissues (Kinoshita and Shimazaki, 2002; Ueno et al., 2005; Hayashi et al., 2011) and occurs at low fluence rates has led to the suggestion that the ATP required for the proton pumps is most probably provided by GC mitochondria (Shimazaki et al., 2007). This is supported by the lower number of chloroplasts (although this is species specific; Lawson et al., 2003) and high concentration of mitochondria reported for GCs (Parvathi and Raghavendra, 1995; Willmer and Fricker, 1996) along with high rates of respiration (Allaway and Setterfield, 1972; Shimazaki et al., 1983). Furthermore, when respiration was repressed with the inhibitors oligomycin and KCN or low [O₂], ATP levels were greatly reduced in GCs (Shimazaki et al., 1983; Gautier et al., 1991), blue-light-dependent proton pumping was reduced (Mawson, 1993), and stomatal opening was inhibited (Schwartz and Zeiger, 1984). These findings demonstrate that the signalling pathways and at least some of the energetics for osmoregulation lie within the GCs themselves. In addition, photosynthetic electron transport within GC chloroplasts has been proposed to directly provide ATP for blue-light-induced H⁺ transport via plasma membrane H⁺-ATPase (Suetsugu et al., 2014). In their experiment, Suetsugu et al. (2014) showed that red light enhanced blue-light-dependent H⁺ pumping in protoplasts, and that this was eliminated by DCMU, and in intact leaves DCMU inhibited both red and blue light stomatal opening. From this work, they concluded that ATP and/or reducing equivalents from GC electron transport is involved in fuelling blue-light-dependent stomatal opening.

**Impact of green light on stomatal behaviour and photosynthesis**

Although the red and blue regions of the spectrum are considered the main drivers of photosynthesis and stomatal behaviour in higher plants, it is important to consider the influence of other spectral qualities of light, including green light. Green light (~500–560 nm) has been reported to inhibit blue-light-induced stomatal opening across a number of plant species (Talbott et al., 2002), but seems to depend almost entirely on the light environment experienced by the plant during growth (Wang et al., 2011; Aasamaa and Aphalo, 2016). Stomatal responses to green light were observed in plants grown under conditions reproducing an understory environment (Aasamaa and Aphalo, 2016), and the magnitude of green-light-driven stomatal responses decreased over the course of the day (Talbott et al., 2006). Although the receptors and exact mechanism of stomatal response to green light have not been identified, green light is known to deactivate the blue light cryptochrome photoreceptors via removal of the signal that suppresses ABA production in GCs, promoting a decrease in stomatal aperture.
(Bouly et al., 2007). Green light has also been shown to contribute to photosynthesis, often at a more efficient rate than red or blue light due to non-photosynthetic absorption of blue light by carotenoids (McCree, 1972), and particularly in strong white light (Terashima et al., 2009), and therefore can impact photosynthetic-dependent stomatal opening (Lanoue et al., 2018). Moreover, Wang et al. (2011) observed a green-light-driven stomatal response in sunflower leaves, which, similarly to the stomatal response to red light, was photosynthesis-dependent as it was partly eliminated when DCMU was applied (Wang et al., 2011). This implicates the potential existence of a green light receptor, with cryptochromes suggested as being involved in this DCMU-independent fraction (Lawson et al., 2018), and that any further signal transduction is photosynthetically dependent, although further work would be required to elucidate a green light photoreceptor. It has been suggested that a possible role of the green light reversal effect on blue-light-driven stomatal opening could be the prevention of excessive leaf water loss through stomata under (green light-rich) vegetational shade, where, within a crop or other terrestrial plant canopy, photosynthetic potential is greatly reduced (Talbott et al., 2006; Aasamaa and Aphalo, 2016).

**Species specificity to blue light**

Interestingly stomatal responses to blue light are not universal, with fern species of Polypodiopsida (Doi et al., 2015) and Adiantum capillus-veneris (Doi et al., 2006), along with several species from the family Solanaceae, exhibiting a lack of stomatal response to blue light. The facultative CAM plant Mesembryanthemum crystallinum loses its stomatal blue light response when the plant shifts from C₃ metabolism to CAM (Tallman et al., 1997). In some species, including the gymnosperm Cycas revoluta and the ferns Equisetum hyemale and Psilotum nudum, blue light is essential for stomata to open (Doi et al., 2015), suggesting that these differences may be due to various evolutionary pressures, whilst the loss of a stomatal blue light response in Polypodiopsida may be the result of adaptation to understory canopy environments (Doi et al., 2015). Moreover, it should be kept in mind that growth conditions, such as drought and high temperatures that may alter the water status of the plant, may impact stomatal sensitivity to blue and red light (Wang et al., 2011; Aasamaa and Aphalo, 2016; Lanoue et al., 2018). This is because plants will balance the need to maintain leaf turgor and/or maximize carbon gain and evaporative cooling (Lawson and Blatt, 2014), and finding the balance between these factors is ultimately dependent on species specificity to the growth environment (Lawson and Vialet-Chabrand, 2019). Although several species have been reported to respond to blue light (e.g. Arabidopsis thaliana and Vicia faba; Table 1), those that do not respond or exhibit a diminished or slow response have generally not been reported (e.g. Nicotiana tabacum; Loreto et al., 2009). This is further complicated by the different protocols used to assess red and blue light responses, making comparison almost impossible, with different red and blue light intensities, ratios, and even durations being applied. Furthermore, the photosynthesis dependence of stomatal response to blue light is species specific (see Wang et al., 2011; Dumont et al., 2013; Suetsugu et al., 2014), with some species known to respond to blue light even in the dark (with no red light background) (Dumont et al. 2013), whilst in others stomatal blue light is only apparent on a background of red light. This has significance, as it is presumed that the response of stomata to blue light does not necessarily require photosynthesis, and that different species may use different sources of energy for blue-light-induced stomatal opening (Daloso et al., 2016; McLachlan et al., 2016; Santelia and Lunn, 2017). Given the evidence that indicates that species specificity of stomatal response to blue light exists, there is a need for standardization of the protocols to be able to accurately compare the biological importance of the stomatal blue light response between and within species.

**Impact of red and blue light on dynamic stomatal response**

Although some species do not exhibit a stomatal blue light response (Doi et al., 2006), little is known about diversity in the magnitude and/or speed of these responses. This is especially important when considering the impact stomatal behaviour to blue light might have on carbon uptake and water use in major crop species (see Wang et al., 2011; Dumont et al., 2013; Suetsugu et al., 2014). The fact that stomata in some species open to blue light even when photosynthesis is already saturated with red light (Shimazaki et al., 2007) means that gs may be higher than required to achieve maximum CO₂ diffusion for photosynthesis, and therefore WUE is greatly reduced. Conversely, the impact of stomatal opening response to blue light on carbon uptake depends on the degree of diffusional limitation of gs for photosynthesis. Figure 4 illustrates the influence of blue light on gs over the diurnal period, and how this greatly affects WUE even when photosynthesis is saturated. From this, we can infer that reducing stomatal sensitivity to blue light may potentially be beneficial for optimizing crop resource use, whereby photosynthetic rates are maintained whilst using water more efficiently. However, this may only be beneficial under certain environmental conditions, as reduced gs could lead to increased leaf temperature, which, depending on the species and environment, could be detrimental to photosynthetic rates (Matthews and Lawson, 2019) and overall plant productivity. Here we show, via thermal imaging, the impact of blue-light-dependent gs response on leaf cooling, and how the addition of ~10% of blue light facilities greater leaf evaporative cooling even when light intensity is held constant (Fig. 5). Decreasing water loss during early stages of growth in crops such as wheat would facilitate greater water availability later in the season, that in turn could facilitate sustained photosynthetic rates through the grain-filling period when water is a major limiting factor, potentially increasing overall grain yield (Acreche and Slaper, 2009; Carmo-Silva et al., 2017; Kaiser et al., 2018; Lanoue et al., 2018).

Generally, the mechanisms behind the speed of stomatal response refer to short-term responses (seconds to minutes), and are not necessarily sufficient to explain diurnal behaviour of A
| Species                     | Response | Publications                          | Measurement | Intensity of RL (µ mol m⁻² s⁻¹) | Intensity of BL (µ mol m⁻² s⁻¹) | Duration of BL (min) | Approximate increase (%) |
|----------------------------|----------|---------------------------------------|-------------|----------------------------------|----------------------------------|----------------------|--------------------------|
| **Model plants**           |          |                                       |             |                                  |                                  |                      |                          |
| Arabidopsis thaliana       | Yes      | Talbott et al. (2002)                 | a           | 0                                | 5                               | 90                   | –                        |
|                            |          | Takerniya et al. (2013)               | gsw         | 80/600                           | 5                               | 20                   | 75                       |
|                            |          | Suetsugu et al. (2014)                | gsw         | 60/240/600                       | 5                               | 10                   | 50                       |
|                            |          | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 5                    | 30                       |
| Nicotiana glauca           | Yes      | Talbott et al. (2002)                 | a           | 0                                | 5                               |                       | 90 –                     |
| Nicotiana tabacum          | Yes      | Talbott et al. (2002)                 | a           | 0                                | 5                               |                       | 90 –                     |
|                            | No       | Loreto et al. (2009)                  | gsw         | 210                             | 90                              | 30                   | 0                        |
| **Crops**                  |          |                                       |             |                                  |                                  |                      |                          |
| Triticum aestivum          | Yes      | Karlsson et al. (1983)                | E           | 0                                | 20/50/100                       | 120                  | –                        |
|                            |          | Karlsson (1986a)                      | gsw         | 460                             | 25                              | 2                    | 30                       |
| Oryza sativa               | Yes      | Shimazaki et al. (2007)               | gsw         | 600                             | 5                               | 20                   | 33                       |
| Helianthus annuus          | Yes      | Wang et al. (2011)                    | gsw         | 0                               | 250                             | 30                   | 100                      |
| Hordeum vulgare            | Yes      | Talbott et al. (2002)                 | a           | 0                                | 5                               |                       | 90 –                     |
| Vicia faba                 | Yes      | Lurie (1978)                          | a           | 0                                | 20                              | 150                  | 33                       |
|                            |          | Ogawa (1981)                          | E           | –                               | –                               | 30                   | 50                       |
|                            |          | Assmann et al. (1985)                 | gsw         | 525                             | 260                             | 0.83                 | 19                       |
|                            |          | Gorton et al. (1993)                  | gsw         | 0                               | 150                             | 90                   | 50                       |
|                            |          | Frechilla et al. (2000, 2004)         | a           | 120                             | 10                              | 90                   | 50                       |
|                            |          | Talbott et al. (2002)                 | a           | 0                                | 5                               |                       | 90 –                     |
|                            |          | Takerniya et al. (2006)               | a           | 150                             | 10                              | 150                  | 50                       |
| Pisum sativum              | Yes      | Talbott et al. (2002)                 | a           | 0                                | 5                               |                       | 90 –                     |
| Lactuca sativa             | Yes      | Clavijo-Herrera et al. (2018)         | gsw         | 270                             | 19                              |                       | 50                       |
| Allium cepa                |          | Ogawa (1981)                          | E           | –                               | –                               | 30                   | 50                       |
|                            |          | Talbott et al. (2002)                 | a           | 0                                | 5                               |                       | 90 –                     |
| **Trees**                  |          |                                       |             |                                  |                                  |                      |                          |
| Nothofagus alpina (Popp. and Endl.) | Yes | Aasama and Aphalo (2016)             | gsw         | 250                             | 15                              | 6                    | 50                       |
| Betula pendula Roth        | No       | Aasama and Aphalo (2016)              | gsw         | 250                             | 15                              | 6                    | 0                        |
| Populus deltoides × Populus nigra | Yes | Dumont et al. (2013)               | gsw         | 0                               | 30                              | 40                   | 165                      |
| Platanus orientalis        | No       | Loreto et al. (2009)                  | gsw         | 210                             | 90                              | 30                   | 0                        |
| Ginkgo biloba              | Yes      | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 30                       |
| **Ferns**                  |          |                                       |             |                                  |                                  |                      |                          |
| Dicksonia linearis         | No       | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 0                        |
| Angiopteris lygodifolia    | Yes      | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 15                       |
| Botrychium tenatum         | Yes      | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 43                       |
| Equisetum hyemale          | Yes      | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 300                      |
| Psilotum nudum             | Yes      | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 1150                     |
| Lepisorus thunbergianus    | No       | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 0                        |
| Theleptis acuminata        | No       | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 0                        |
| Osmunda japonica           | No       | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 0                        |
| Alsophila mertensiana      | No       | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 0                        |
| Lycopodites                |          |                                       |             |                                  |                                  |                      |                          |
| Selaginella moellendorffii | Yes      | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 50                       |
| Selaginella uncinata       | Yes      | Doi et al. (2015)                     | gsw         | 600                             | 5                               | 60                   | 80                       |
| Others                     |          |                                       |             |                                  |                                  |                      |                          |
| Commelina communis         | Yes      | Iino et al. (1985)                    | gsw         | 500                             | 25                              | 90                   | 54                       |
|                            |          | Assmann (1988)                        | gsw         | 263                             | 100                             | 15                   | 36                       |
|                            |          | Assmann (1992)                        | gsw         | 700/1500                        | 200                             | 43                   | 60                       |
|                            |          | Lascève et al. (1993)                 | gsw         | 105                             | 65                              | 1                    | 115                      |
|                            |          | Talbott et al. (2002)                 | a           | 0                               | 5                               |                       | 90 –                     |
| Paphiopedilum hamsonianum  | Yes      | Zeiger et al. (1983)                  | a           | 0                               | 10                              | 120                  | 300                      |
|                            |          | Assmann (1988)                        | gsw         | 263                             | 100                             | 15                   | 33                       |
| Mesembryanthemum crystallinum | Yes | Mawson and Zaugg (1994)             | a           | 0                               | 300                             | 110                  | 43                       |
|                            |          | Talman et al. (1997)                  | a           | 350                             | 15                              | 240                  | –                        |
and gᵢ. Even in constant light conditions, decreases in gᵢ are often seen towards the end of the day (Matthews et al., 2018), and it has been suggested that mechanisms such as increases in the amount of sucrose from photosynthesis mediate this response, and that sucrose content and metabolism play a major role in the long term coordination of A and gᵢ (Lawson et al., 2014). This sugar accumulation at high photosynthetic rates associated with high photosynthetically active radiation (PAR) conveys long-term photosynthetic feedback on gᵢ over the course of the day (Lu et al., 1995, 1997; Outlaw, 2003; Kang et al., 2007; Kelly et al., 2013), which may theoretically alter stomatal dynamic behaviour over the diurnal period (Matthews et al., 2017). It should also be noted that toward the end of the day, some species exhibit a slower gᵢ response to changes in light intensity, with a slow closing response resulting in continued high gᵢ, leading to substantial water loss and reduced WUE over the diurnal period (Blom-Zandstra et al., 1995; Lawson and Blatt, 2014).

It has already been reported that in wheat the magnitude or sensitivity of stomata to blue light is enhanced under a ‘strong’ red light background (Karlsson, 1986a), with this behaviour potentially being dose dependent where the intensity and even the duration of the background red light determines the extent to which stomata respond to a blue light signal (Ogawa et al., 1978; Zeiger, 1984; Iino et al., 1985; Karlsson, 1986a; Sharkey and Ogawa, 1987; Assmann, 1988). Furthermore, as the stomatal response to blue light is GC specific, it may be suggested that the speed of the gᵢ response is increased under blue light. It can therefore be proposed that altering the sensitivity and diurnal behaviour of the gᵢ blue light response, via either breeding techniques or genetic modification, could lead to a reduction in the limitation of A by gᵢ, and the slow decrease in A and gᵢ through the day may be prevented. This paves the way for potential improvements in photosynthetic carbon assimilation over the diurnal period (Vialet-Chabrand et al., 2017b; Matthews et al., 2017, 2018), whilst positively influencing WUE and plant productivity. As most studies have been carried out under ‘ideal’

---

### Table 1. Continued

| Species                  | Response | Publications                           | Measurement | Intensity of RL (μ mol m⁻² s⁻¹) | Intensity of BL (μ mol m⁻² s⁻¹) | Duration of BL (min) | Approximate increase (%) |
|--------------------------|----------|----------------------------------------|-------------|-------------------------------|-------------------------------|-----------------------|--------------------------|
| Xanthium pennsylvanicum  | Yes      | Mansfield and Meidner (1966)            | gₛₛ         | –                              | –                             | 240                   | 700                      |
| Tradescantia pallida    | Yes      | Ballard et al. (2019)                   | a           | 50                             | 50                            | –                     | –                       |
| Musa acuminate cv. Grand Nain AAA | Yes | Zait et al. (2017)                   | gₛₛ         | 1080                          | 120                           | 120                   | 40                       |
| Festuca arundinacea     | Yes      | Bariliot et al. (2010)                 | gₛₛ         | 277                           | 60                            | –                     | 100                      |
| Cycas revoluta          | Yes      | Doi et al. (2015)                      | gₛₛ         | 600                           | 5                             | 60                    | 3900                     |
| Chamaecyparis obtusa    | Yes      | Doi et al. (2015)                      | gₛₛ         | 600                           | 5                             | 60                    | 50                       |
| Gnetum spp.             | Yes      | Doi et al. (2015)                      | gₛₛ         | 600                           | 5                             | 60                    | 120                      |
| Zamia furfuracea        | Yes      | Doi et al. (2015)                      | gₛₛ         | 600                           | 5                             | 60                    | 100                      |
| Phragmites longifolium  | Yes      | Zeiger et al. (1985)                   | gₛₛ         | 65                            | 85                            | 15                    | 30                       |
| Paphiopedilum insignae  | Yes      | Zeiger et al. (1985)                   | gₛₛ         | 68                            | 82                            | 15                    | 20                       |

The presence or absence of the response was reported for each species as well as the experimental protocol used. Measurements are reported as stomatal aperture (a), stomatal conductance (gₛₛ), or transpiration (E).
well-watered conditions, there is little information describing the influence of drought, water status, or temperature on the temporal response of g (Lawson and Blatt, 2014; Haworth et al., 2018). As a consequence, it is currently unknown how manipulation of stomatal sensitivity to blue and red light may impact plant fitness and productivity. However, as the frequency and intensity of periods of drought are set to increase globally in the near future, water availability and its transport from roots to stomata will be a major limiting factor for crop and terrestrial ecosystems moving forward. As such, improving our understanding of the dynamic response of stomata to different spectra of light and how manipulation of the stomatal sensitivity will impact spatial and temporal stomatal responses (Matthews et al., 2017; Vialet-Chabrand et al., 2017a; Lawson and Vialet-Chabrand, 2019; Vialet-Chabrand and Lawson, 2019), remains an unexploited avenue in which to improve plant performance and crop productivity.

Summary and future perspectives

Stomatal research over the past few decades has revealed a complicated network of osmoregulatory and signalling pathways in GCs (e.g. Lawson, 2009; Daloso et al., 2016; Inoue and Kinoshita, 2017) that are species specific and influenced by the growth environment. Although significant progress has been made over the past few decades, understanding of stomatal responses to various environmental signals (including irradiance) and substantial advances in GC metabolism and osmoregulatory pathways, many gaps remain regarding the integration and hierarchy of these diverse processes and the extent to which each contributes to stomatal function. As we have illustrated throughout this review, there is extensive evidence for both mitochondrial and photosynthetic electron transport ATP supply for the H⁺-ATPase, ion channel activation, and stomatal opening in response to both blue and red illumination. However, the extent to which each energetic supply contributes to stomatal movements has yet to be fully elucidated. It is also apparent that manipulating one pathway may result in the up-regulation of an alternative pathway to compensate, increasing the difficulty and complexity for determining the involvement and extent of each. The role and participation of GC electron transport and photosynthetic processes remain continuing subjects of debate, despite the fact that chloroplasts are a key feature of most GCs. Furthermore, the extent to which mesophyll signals play a role and the origins and nature of these signals need clarification. Therefore, understanding the mechanisms and signal transduction pathways that operate in GCs and the influence of mesophyll photosynthesis on these processes (and subsequent stomatal responses) is essential if we are to fully exploit the relationship between A and g, in order to improve gaseous fluxes, maximize CO₂ uptake, and optimize water use in fluctuating environments given the predicted changes in climate (Matthews and Lawson, 2019).

We are all fully aware that global demand for food is growing, and, due to a growing world population, it has been estimated that a >50% increase in major crop yield is required by 2050 (Long et al., 2015). This situation is further exacerbated by predicted increases in atmospheric temperature and heat wave frequency experienced by crop and terrestrial ecosystems (Perkins et al., 2012). These variable changes in climate conditions are often linked to changes in precipitation patterns, water availability (Stéfanon et al., 2014), and drought (Urban et al., 2017), and are set to aggravate crop losses and increase the agricultural water supply requirements by an estimated 17% (Pennis, 2008). When water is limiting, plants close stomata to avoid excessive water loss, even during periods of high light when mesophyll demands for CO₂ are high, and long-term damage to photosynthetic machinery may be induced (Berry and Björkman, 1980). In crop and forest ecosystems this reduces transpiration

![Fig. 5. Theoretical representation of the impact of blue-light-driven changes on stomatal conductance (gₛ) on leaf temperature and evaporative cooling. Representative courses of gₛ under a step increase in red light and a further addition of blue light (the total light intensity remains constant) (A). Arabidopsis thaliana plants subjected to these light conditions are shown, highlighting the change in leaf temperature driven by changes in gₛ (A). Representative rice (Oryza sativa) plants exposed to 30 min of 100% red light and a 90:10 ratio of red to blue light, demonstrating the difference in leaf temperature (°C) and therefore gₛ in a major crop variety (B).](image-url)
but at a cost of reduced evaporative cooling (Ainsworth and Rogers, 2007; Bernacchi et al., 2007; Lambersma et al., 2011; Hussain et al., 2013; Tricker et al., 2018), which greatly impacts biochemical and metabolic mechanisms of photosynthesis (Perdomo et al., 2017); including but not limited to the activity of the temperature-sensitive enzyme Ribisco activase and ATP synthesis (Tezara et al., 1999; Galmés et al., 2007). Finding the ‘ideal’ balance between carbon gain, evaporative cooling, and maintenance of hydraulic status is crucial for maximizing crop performance and productivity, whether it be for field- or greenhouse-grown crops.

As mentioned above, blue light induces a GC-specific stomatal response that enables stomatal opening, increasing the magnitude of $g_s$. Given the importance of the blue light response of $g_s$ for evaporative cooling and carbon gain in major crop species (see above), manipulation of this blue-light-triggered response represents a novel and generally unexploited target as a strategy to increase carbon uptake and/or maintenance of optimal leaf temperature, and conversely for crop water use. An analogous approach has recently been used by Papantasiou et al. (2019) who expressed the synthetic light-gated K$^+$ channel BLINK1 specifically in the GCs to enhance solute fluxes, and produced plants with stomata that opened and closed more rapidly, resulting in greater WUE and biomass.

Understanding the mechanisms behind the blue- and red-light-driven responses of stomata would potentially enable greater control of the synchronicity between $A$ and $g_s$ under dynamic light conditions, and therefore optimize the relationship between water use and carbon gain (Lawson and Blatt, 2014). Enabling a blue light $g_s$ response in species in which it is absent increases the potential for cultivating crop ideotypes for specific climate conditions. In fact, it has already been demonstrated that blue light, even at low intensities, reduced stomatal oscillations often seen under drought conditions (Zait et al., 2017; Ballard et al., 2019). These oscillations, the cyclic opening and closing of stomata, are presumed to initiate from hydraulic mismatch between water supply and transpiration rate, and therefore it is suggested that a blue-light-driven $g_s$ response helps recover this synchronicity and improve plant performance under drought (Zait et al., 2017). On the other hand, reducing stomatal sensitivity to blue light could provide a route to producing plants with reduced levels of $g_s$, potentially enhancing water saving at crucial stages during plant development (e.g., grain filling). However, this could be to the detriment of CO$_2$ diffusion and leaf cooling, but could provide ideotypes for specific growth environments.

Given the increase in alternative growth spaces (e.g., vertical farming) for ‘indoor’ crops, a new generation of smart LED lighting allowing for more precise control of light quality and quantity has recently become available. Several recent studies have demonstrated the importance of blue light for vegetable crop growers, as a way of improving WUE and even overall yield (Clavijo-Herrera et al., 2018; Kaiser et al., 2018; Lanoue et al., 2018; Pennisi et al., 2019). For example, a recent study demonstrated that different ratios of red and blue light optimized growth, yield, and WUE in basil (Pennisi et al., 2019). This is interesting, as the optimal ratios for these targets were shown to be different from the ratios of light spectra observed in nature, whether it is direct or diffuse light (Urban et al., 2007). This highlights the potential to engineer the tapestry of plant pigments to utilize more of the sunlight’s spectrum, to maximize light absorption, and to overcome light saturation of the downstream photosynthetic processes (Long et al., 2015; Song et al., 2017), as it is already known that more than half of the energy in the solar spectrum is not utilized by the plant (Zhu et al., 2008).

In this review, we have focused on stomatal response to different light qualities. Emphasis is placed on the response of $g_s$ to blue and red spectra of light, and how plants use these independent light responses to enforce GC movement to maximize plant performance. With recent research highlighting the importance of the rapidity of $g_s$ responses to light for plant water status and photosynthetic carbon gain, we emphasize the need to re-assess the role of stomatal behaviour under blue and red light between and within species, as a means to understand the importance of stomata in crop performance. Additionally, with increases in global temperature and water demand for agrcultural practices predicted to intensify crop losses in the future, we outline the potential of manipulating stomatal response to light quality to maximize drought tolerance, water-saving strategies, and yield.

Acknowledgements

JSAM was supported through a BBSRC IWYP programme (grant no: BB/S005080/1) awarded to TL. SV-C was supported by BBSRC Transforming India’s Green Revolution by Research and Empowerment for Sustainable food Supplies (BB/P027970/2) award to TL and led by the University of Cambridge.

References

Aasamaa K, Ahalo PJ. 2016. Effect of vegetational shade and its components on stomatal responses to red, blue and green light in two deciduous tree species with different shade tolerance. Environmental and Experimental Botany 121, 94–101.
Aasamaa K, Sõber A. 2011. Responses of stomatal conductance to simultaneous changes in two environmental factors. Tree Physiology 31, 855–864.
Acreche MM, Slater GA. 2009. Grain weight, radiation interception and use efficiency as affected by sink-strength in Mediterranean wheats released from 1940 to 2005. Field Crops Research 110, 98–105.
Ainsworth EA, Rogers A. 2007. The response of photosynthesis and stomatal conductance to rising [CO$_2$]: mechanisms and environmental interactions. Plant, Cell & Environment 30, 258–270.
Allaway WG, Setterfield G. 1972. Ultrastructural observations on guard cells of Vicia faba and Allium porrum. Canadian Journal of Botany 50, 1405–1413.
Ando E, Kinoshita T. 2018. Red light-induced phosphorylation of plasma membrane H+-ATPase in stomatal guard cells. Plant Physiology 178, 838–849.
Andrés Z, Pérez-Hormaeche J, Leidi EO, et al. 2014. Control of vacuolar dynamics and regulation of stomatal aperture by tonoplast potassium uptake. Proceedings of the National Academy of Sciences, USA 111, E1806–E1814.
Antunes WC, de Menezes Daloso D, Pinheiro DP, Williams TCR, Loureiro ME. 2017. Guard cell-specific down-regulation of the sucrose transporter SUT1 leads to improved water use efficiency and reveals the interplay between carbohydrate metabolism and K$^+$ accumulation in the regulation of stomatal opening. Environmental and Experimental Botany 135, 73–85.
Aphalo PJ, Sánchez RA. 1986. Stomatal responses to light and drought stress in variegated leaves of *Hedera helix*. Plant Physiology 81, 768–773.

Assmann SM. 1988. Enhancement of the stomatal response to blue light by red light, reduced intercellular concentrations of CO₂, and low vapor pressure differences. Plant Physiology 87, 226–231.

Assmann SM. 1992. Effects of light quantity and quality during development on the morphology and stomatal physiology of *Commerella communis*. Oecologia 92, 188–195.

Assmann SM, Shimazaki K. 1990. The multisensory guard cell. Stomatal responses to blue light and abscisic acid. Plant Physiology 119, 809–816.

Assmann SM, Simoncini L, Schroeder JI. 1985. Blue light activates electrogenic ion pumping in guard cell protoplasts of *Vicia faba*. Nature 318, 285.

Azoulay-Shemer T, Palomares A, Bagheri A, Israelsson-Nordstrom M, Engineer CB, Bargmann BO, Stephan AB, Schroeder JI. 2015. Guard cell photosynthesis is critical for stomatal turgor production, yet does not directly mediate CO₂- and ABA-induced stomatal closing. The Plant Journal 83, 567–581.

Bail JT, Berry JA. 1982. Cl/Ca ratio: a basis for predicting stomatal control of photosynthesis. Carnegie Institution of Washington Yearbook 81, 98–92.

Ballard T, Peak D, Mott K. 2019. Blue and red light effects on stomatal oscillations. Functional Plant Biology 46, 146–151.

Banáš AK, Anwaral C, Labuz J, Sztalteman O, Gabryš H. 2012. Blue light signalling in chloroplast movements. Journal of Experimental Botany 63, 1559–1574.

Banerjee R, Batschauer A. 2005. Stomatal responses to blue light and abscisic acid. Plant Physiology 139, 109–116.

Baroli I, Price GD, Badger MR, von Caemmerer S. 2008. The contribution of transpiration to environmental factors. Plant, Cell & Environment 31, 2353–2371.

Barillot R, Frak E, Combes D, Durand JL, Escobar-Gutiérrez AJ. 2012. A comparative study of the effects of ozone on stomatal responses to environmental parameters. Plant, Cell & Environment 37, 517–523.

Barillot R, Frak E, Combes D, Durand JL, Escobar-Gutiérrez AJ. 2012. Effects of ozone on stomatal responses to environmental parameters. Plant, Cell & Environment 35, 517–523.

Barlow DR, Jones AM. 2001. Blue light sensing in higher plants. Journal of Biological Chemistry 276, 11457–11460.

Clavijo-Herrera J, van Santen E, Gómez C. 2018. Growth, water-use efficiency, stomatal conductance, and nitrogen uptake of two lettuce cultivars grown under different percentages of blue and red light. Horticulture 4, 16.

Cockburn W. 1983. Stomatal mechanism as the basis of the evolution of CAM and C₄ photosynthesis. Plant, Cell & Environment 6, 275–279.

Cowan IR, Troughton JH. 1971. The relative role of stoma in transpiration and assimilation. Planta 97, 325–336.

Daloso DM, Antunes WC, Pinheiro DP, Waquim JP, Araújo WU, Loureiro ME, Fernie AR, Williams TC. 2015. Tobacco guard cells fix CO₂ by both Rubisco and PEPCase while sucrose acts as a substrate during light-induced stomatal opening. Plant, Cell & Environment 38, 2353–2371.

Daloso DM, Dos Anjos L, Fernie AR. 2016. Roles of sucrose in guard cell regulation. New Phytologist 211, 809–818.

Daloso DM, Dos Anjos L, Fernie AR. 2016. Roles of sucrose in guard cell regulation. New Phytologist 211, 809–818.

De Angelis A, Zhang J, Meyer S, Martinoia E. 2013. AATLMT9 is a malate-activated vacuolar chloride channel required for stomatal opening in Arabidopsis. Nature Communications 4, 1804.

Doi M, Kitagawa Y, Shimazaki K. 2015. Stomatal blue light response is present in early vascular plants. Plant Physiology 169, 1205–1213.

Doi M, Shigenaga A, Emi T, Ninomiya T, Shimazaki K. 2006. Stomatal responses to blue light and abscisic acid. Plant Physiology 143, 188–195.

Eisenach C, De Angelis A. 2017. Ion transport at the vacuole during stomatal movements. Plant Physiology 174, 520–530.

Ewert MS, Outlaw WH, Zhang S, Aghoram K, Riddle KA, Outlaw B. 2000. Accumulation of an apoplastic solute in the guard-cell wall is sufficient to exert a significant effect on transpiration in *Vicia faba* leaflets. Plant, Cell & Environment 23, 195–203.

Faralli M, Matthews JSA, Lawson T. 2019. Exploiting natural variation and genetic manipulation of stomatal conductance for crop improvement. Current opinion in Plant Biology 49, 1–7.

Farquhar GD, Sharkey TD. 1982. Stomatal conductance and photosynthesis. Annual Review of Plant Physiology 33, 317–345.

Fischer RA, Rebetzke GJ. 2018. Indirect selection for potential yield in early-generation, spaced plantings of wheat and other small-grain cereals: a review. Crop and Pasture Science 69, 439–459.

Fischer RA, Rees D, Sayre KD, Lu Z-M, Condon AG, Saavedra AL. 1998. Wheat yield progress associated with higher stomatal conductance and photosynthetic rate, and cooler canopies. Crop Science 38, 1467–1475.

Frehilla S, Taltbod LD, Bogomolni RA, Zeiger E. 2000. Reversal of blue light-stimulated stomatal opening by green light. Plant & Cell Physiology 41, 171–176.
Frechilla S, Talbott LD, Zeiger E. 2004. The blue light-specific response of Vicia faba stomata accelerates to growth environment. Plant & Cell Physiology 45, 1709–1714.

Fujita T, Noguchi K, Takaki H, Terashima I. 2019. Confirmation of mesophyll signals controlling stomatal responses by a newly devised trans-planting method. Functional Plant Biology, 46, 467–481.

Fujita T, Noguchi K, Terashima I. 2013. Apoplastic mesophyll signals induce rapid stomatal responses to CO2 in Commelina communis. New Phytologist 199, 395–406.

Galmés J, Medrano H, Flexas J. 2007. Photosynthetic limitations in response to water stress and recovery in Mediterranean plants with different growth forms. New Phytologist 175, 81–93.

Gautier H, Vavasseur A, Gans P, Lascève G. 1991. Relationship between respiration and photosynthesis in guard cell and mesophyll cell protoplasts of Commelina communis L. Plant Physiology 95, 636–641.

Głowacka K, Kromdijk J, Kucera K, Xie J, Cavanagh AP, Leonelli L, Leakey ADB, Ort DR, Niyogi KK, Long SP. 2018. Photosystem II Subunit S overexpression increases the efficiency of water use in a field-grown crop. Nature Communications 9, 568.

Gorton HL, Williams WE, Assmann SM. 1993. Circadian rhythms in stomatal responsiveness to red and blue light. Plant Physiology 103, 399–406.

Hashimoto M, Negi J, Young J, Israelsson M, Schroeder JI, Iba K. 2006. Arabidopsis HT1 kinase controls stomatal movements in response to CO2. Nature Cell Biology 8, 43–53.

Haupt W. 1999. Chloroplast movement: from phenomenology to molecular biology. In: Esser K, Kaderje JW, Lütge U, Runge M, eds. Progress in botany, Vol. 60. Berlin, Heidelberg: Springer. 3–36.

Haworth M, Scutt CP, Douthe C, Marino G, Gomes MTG, Loreto F, Flexas J, Centritto M. 2018. Allocation of the epidermis to stomata cells and its integration for stomatal dynamics. Plant Physiology 174, 857–868.

Hayashi M, Inoue S, Takahashi K, Kinoshita T. 2011. Immunohistochemical detection of blue light-induced phosphorylation of the plasma membrane H+-ATPase in stomatal guard cells. Plant & Cell Physiology 52, 1238–1248.

Hayashi M, Inoue SI, Ueno Y, Kinoshita T. 2017. A Raf-like kinase bZIP mediates blue light-dependent stomatal opening. Scientific Reports 7, 45586.

Hedrich R, Marten I. 1993. Malate-induced feedback regulation of plasma membrane anion channels could provide a CO2 sensor to guard cells. The EMBO Journal 12, 897–901.

Hedrich R, Marten I, Lohse G, Dietrich P, Winter H, Lohaus G, Heldt HW. 1994. Malate sensitive anion channels enable guard cells to sense changes in the ambient CO2 concentration. The Plant Journal 6, 741–748.

Hiyama A, Takemiya A, Munemasa S, Okuma E, Sugiyama N, Tada Y, Murata Y, Shimazaki KI. 2017. Blue light and CO2 signals converge to regulate light-induced stomatal opening. Nature Communications 8, 1284.

Horrer D, Flütsch S, Pazmino D, Matthews JS, Thalmann M, Negro A, Leonhardt N, Lawson T, Santelise D. 2016. Blue light induces a distinct starch degradation pathway in guard cells for stomatal opening. Current Biology 26, 362–370.

Hsiao TC, Allaway WG. 1973. Action spectra for guard cell Rb uptake and stomatal opening in Vicia faba. Plant Physiology 51, 82–88.

Hussain MZ, Vanloocke A, Siebers MH, Ruiz-Vera UM, Coder Markelz RJ, Leakey AD, Ort DR, Bernacchi CJ. 2013. Future carbon dioxide concentration decreases canopy evapotranspiration and soil water depletion by field-grown maize. Global Change Biology 19, 1572–1584.

Iino M, Ogawa T, Zeiger E. 1985. Kinetic properties of the blue-light response of stomata. Proceedings of the National Academy of Sciences, USA 82, 8019–8023.

Inoue SI, Kinoshita T. 2017. Blue light regulation of stomatal opening and the plasma membrane H+-ATPase. Plant Physiology 174, 531–538.

Inoue S, Takemaya A, Shimazaki K. 2010. Phototropin signaling and stomatal opening as a model case. Current Opinion in Plant Biology 13, 587–593.

Jezek M, Blatt MR. 2017. The membrane transport system of the guard cell and its integration for stomatal dynamics. Plant Physiology 174, 487–519.

Jones HG. 2013. Plants and microclimate: a quantitative approach to environmental plant physiology. Cambridge: Cambridge University Press.

Jossior M, Kroniewicz L, Dalmas F, Le Thiec D, Ephritikhine G, Thome S, Barbier-Brygoo H, Vavasseur A, Filleur S, Leonhardt N. 2010. The Arabidopsis vacuolar anion transporter, ATO1C, is involved in the regulation of stomatal movements and contributes to salt tolerance. The Plant Journal 64, 563–576.

Kaiser E, Morales A, Harbinson J. 2018. Fluctuating light takes crop photosynthesis on a rollercoaster ride. Plant Physiology 176, 977–989.

Kaiser H, Kappen L. 2000. In situ observation of stomatal movements and gas exchange of Aegopodium podagraria L. in the understory. Journal of Experimental Botany 51, 1741–1749.

Kaiser H, Kappen L. 2001. Stomatal oscillations at small apertures: indications for a fundamental insufficiency of stomatal feedback-control inherent in the stomatal turgor mechanism. Journal of Experimental Botany 52, 1203–1213.

Kang Y, Outlaw WH Jr, Andersen PC, Fiore GB. 2007. Guard-cell apoplastic sucrose concentration—a link between leaf photosynthesis and stomatal aperture size in the apoplastic phloem loader Vicia faba L. Plant, Cell & Environment 30, 551–558.

Karlsson PE. 1986a. Blue light regulation of stomata in wheat seedlings. 1. Influence of red background illumination and initial conductance level. Physiologia Plantarum 66, 202–206.

Karlsson PE. 1986b. Blue light regulation of stomata in wheat seedlings. 2. Action spectrum and search for action dichroism. Physiologia Plantarum 66, 207–210.

Karlsson PE, Höglund HO, Klockare R. 1983. Blue light induces stomatal transpiration in wheat seedlings with chlorophyll deficiency caused by SAN 9789. Physiologia Plantarum 57, 417–421.

Kelly G, Mosheilon M, David-Schwartz R, Halperin O, Wallach R, Attia Z, Belusov E, Granot D. 2013. Hexokinase mediates stomatal closure. The Plant Journal 75, 977–988.

Kendrick RE, Kronenberg GHM. 1994. Photomorphogenesis in plants. Dordrecht, The Netherlands: Kluwer Academic.

Kinoshita T, Shimazaki K. 2001. Analysis of the phosphorylation level in guard-cell plasma membrane H+-ATPase in response to fusicoccin. Plant & Cell Physiology 42, 424–432.

Kinoshita T, Shimazaki K. 2002. Biochemical evidence for the requirement of 14-3-3 protein binding in activation of the guard-cell plasma membrane H+-ATPase by blue light. Plant & Cell Physiology 43, 1359–1365.

Kirshbaum MUF, Gross LJ, Pearcy RW. 1988. Observed and modelled stomatal responses to dynamic light environments in the shade plant Alocasia macrorrhiza. Plant, Cell & Environment 11, 111–121.

Kuiper PJ. 1964. Dependence upon wavelength of stomatal movement in epidermal tissue of Sanecoc odoris. Plant Physiology 39, 952–955.

Kusumi K, Hashimura A, Yamamoto Y, Negi J, Iba K. 2017. Contribution of the S-type anion channel SLAC1 to stomatal control and its dependence on developmental stage in rice. Plant & Cell Physiology 58, 2085–2094.

Lammertsma EI, de Boer HJ, Dekker SC, Dilcher DL, Lotter AF, Karlsson PE. 2017. Contribution of blue light responses to dynamic light environments in the shade plant Vicia faba L. in the understorey. Journal of Experimental Botany 68, 3609–3619.
Lawson T, Morison JIL. 2004. Stomatal function and physiology. In: Hernsley A, Poole I, eds. The evolution of plant physiology. Amsterdam: Elsevier, 217–244.

Lawson T, Oxborough K, Morison JI, Baker NR. 2002. Responses of photosynthetic electron transport in stomatal guard cells and mesophyll cells in intact leaves to light, CO2, and humidity. Plant Physiology 128, 52–62.

Lawson T, Oxborough K, Morison JI, Baker NR. 2003. The responses of guard and mesophyll cell photosynthesis to CO2, O2, light, and water stress in a range of species are similar. Journal of Experimental Botany 54, 1743–1752.

Lawson T, Simkin AJ, Kelly G, Granot D. 2014. Mesophyll photosynthesis and guard cell metabolism impacts on stomatal behaviour. New Phytologist 203, 1064–1081.

Lawson T, Terashima I, Fujita T, Wang Y. 2018. Coordination between photosynthesis and stomatal behavior. In: Adams WW III, Terashima I, eds. The leaf: a platform for performing photosynthesis. Cham: Springer, 141–161.

Lawson T, Viallet-Chabrard S. 2019. Speedy stomata, photosynthesis and plant water use efficiency. New Phytologist 221, 93–98.

Lawson T, von Caemmerer S, Baroli I. 2010. Photosynthesis and stomatal behaviour. In: Lützé U, Beyschlag W, Büdel B, Francis D, eds. Progress in botany, Vol. 72. Berlin Heidelberg: Springer, 265–304.

Lebaudy A, Vavasseur A, Hosy E, Dreyer I, Leonhardt N, Thibaud JB, Véry AA, Simonneau T, Sentenac H. 2008. Plant adaptation to fluctuating environment and biomass production are strongly dependent on guard cell potassium channels. Proceedings of the National Academy of Sciences, USA 105, 5271–5276.

Lee J, Bowling DJF. 1992. Effect of the mesophyll on stomatal opening in Commelina communis. Journal of Experimental Botany 43, 951–967.

Lee JS, Bowling DJF. 1995. Influence of the mesophyll on stomatal opening. Functional Plant Biology 22, 357–363.

Lee M, Choi Y, Burla B, Kim YY, Jeon B, Maeshima M, Yoo JY, Martinhoia E, Lee Y. 2008. The ABC transporter ATABC14 is a malate importer and modulates stomatal response to CO2. Nature Cell Biology 10, 1217–1223.

Lin C, Shalitin D. 2003. Cryptochrome structure and signal transduction. Annual Review of Plant Biology 54, 469–496.

Long SP, Marshall-Colon A, Zhu XG. 2015. Measuring the global food demand of the future by engineering crop photosynthesis and yield potential. Cell 161, 56–66.

Loreto F, Tsonev T, Centritto M. 2009. The impact of blue light on leaf mesophyll conductance. Journal of Experimental Botany 60, 2283–2290.

Lu P, Outlaw WH Jr, Smith BG, Freed GA. 1997. A new mechanism for the regulation of stomatal aperture size in intact leaves (accumulation of mesophyll-derived sucrose in the guard-cell wall of Vicia faba). Plant Physiology 114, 109–118.

Lu P, Zhang SQ, Outlaw WH Jr, Riddle KA. 1995. Sucrose: a solute that accumulates in the guard-cell apoplast and guard-cell symplast of open stomata. FEBS Letters 362, 183–184.

Lurie S. 1978. The effect of wavelength of light on stomatal opening. Planta 140, 245–249.

Mansfield TA, Meidner H. 1966. Stomatal opening in light of different wavelengths: effects of blue light independent of carbon dioxide concentration. Journal of Experimental Botany 17, 510–521.

Mansfield TA, Hetherington AM, Atkinson CJ. 1990. Some current aspects of stomatal physiology. Annual Review of Plant Physiology and Plant Molecular Biology 41, 55–75.

Matrosova A, Bogireddi H, Mateo-Peñas A, Hashimoto-Sugimoto M, Iba K, Schroder JI, Israelsson-Nordstrom M. 2015. The HT1 protein kinase is essential for red light-induced stomatal opening and genetically interacts with OST1 in red light and CO2-induced stomatal movement responses. New Phytologist 206, 1126–1137.

Matthews JSA, Lawson T. 2019. Climate change and stomatal physiology. Annual Plant Reviews Online 2, apr0667.

Matthews JSA, Viallet-Chabrard SRM, Lawson T. 2017. Diurnal variation in gas exchange: the balance between carbon fixation and water loss. Plant Physiology 174, 614–623.

Matthews JSA, Viallet-Chabrard S, Lawson T. 2018. Acclimation to fluctuating light impacts the rapidity of response and diurnal rhythm of stomatal conductance. Plant Physiology 176, 1939–1951.

Mawson BT. 1993. Regulation of blue-light-induced proton pumping by Vicia faba L. guard-cell protoplasts: energetic contributions by chloroplast and mitochondrial activities. Planta 181, 293–301.

Mawson BT, Zaugg MW. 1994. Modulation of light-dependent stomatal opening in isolated epidermis following induction of crassulacean acid metabolism in Mesembryanthemum crystallinum L. Journal of Plant Physiology 144, 740–746.

McAusland L, Davey PA, Kanwal N, Baker NR, Lawson T. 2013. A novel system for spatial and temporal imaging of intrinsic plant water use efficiency. Journal of Experimental Botany 64, 4993–5007.

McAusland L, Viallet-Chabrard S, Davey P, Baker NR, Brendel O, Lawson T. 2016. Effects of kinetics of light-induced stomatal responses on photosynthesis and water-use efficiency. New Phytologist 211, 1209–1220.

McCree KJ. 1972. The action spectrum, absorbance and quantum yield of photosynthesis in crop plants. Agricultural Meteorology 9, 191–216.

McLachlan DH, Lan J, Geilfuss CM, et al. 2016. The breakdown of stored tracyglycerols is required during light-induced stomatal opening. Current Biology 26, 707–712.

Messinger SM, Buckley TN, Mott KA. 2006. Evidence for involvement of photosynthetic processes in the stomatal response to CO2. Plant Physiology 140, 771–778.

Morison JIL. 2003. Plant water use, stomatal control. In: Stewart BA, Howell T, eds. Encyclopedia of water science. Boca Raton, FL: CRC Press, 680–685.

Mott KA. 1988. Do stomata respond to CO2 concentrations other than intercellular? Plant Physiology 86, 200–203.

Mott KA, Peak D. 2013. Testing a vapour-phase model of stomatal responses to humidity. Plant, Cell & Environment 36, 936–944.

Mott KA, Sibbernse ED, Shope JC. 2008. The role of the mesophyll in stomatal responses to light and CO2. Plant, Cell & Environment 31, 1299–1306.

Naumburg E, Ellisworth DS. 2000. Photosynthetic sunfleck utilization potential of understory saplings growing under elevated CO2 in FACE. Oecologia 122, 163–174.

Ogawa T. 1981. Blue light response of stomata with starch-containing (Vicia faba) and starch-deficient (Allium cepa) guard cells under background illumination with red light. Plant Science Letters 22, 103–108.

Ogawa T, Ishikawa H, Shimada K, Shibata K. 1978. Synergistic action of red and blue light and action spectra for malate formation in guard cells of Vicia faba L. Journal of Plant Physiology 115, 61–65.

Olesen JE, Junttila O. 2002. Far red end-of-day treatment restores wild type-like plant length in hybrid aspen overexpressing phytochrome A. Physiologia Plantarum 115, 448–457.

Olesen RL, Pratt RB, Gump P, Kemper A, Tallman G. 2002. Red light activates a chloroplast-dependent ion uptake mechanism for stomatal opening under reduced CO2 concentrations in Vicia spp. New Phytologist 153, 497–508.

Outlaw WH Jr. 1989. Critical examination of the quantitative evidence for and against photosynthetic CO2 fixation by guard cells. Physiologia Plantarum 77, 275–281.

Outlaw WH. 2003. Integration of cellular and physiological functions of guard cells. CRC Critical Reviews in Plant Science 22, 503–529.

Outlaw WH Jr, De Vlieghere-He X. 2001. Transpiration rate. An important factor controlling the sucrose content of the guard cell apoplast of broad bean. Plant Physiology 126, 1716–1724.

Outlaw WH, Manchester J. 1979. Guard cell starch concentration quantitatively related to stomatal aperture. Plant Physiology 64, 79–82.

Papanatsiou M, Petersen J, Henderson L, Wang Y, Christie JM, Blatt MR. 2019. Optogenetic manipulation of stomatal kinetics improves carbon assimilation, water use, and growth. Science 363, 1456–1459.

Parvathi K, Raghavendra AS. 1995. Bioenergetic processes in guard cells related to stomatal function. Physiologia Plantarum 77, 145–156.

Pennisi E. 2008. The blue revolution, drop by drop, gene by gene. Science 320, 171–173.
Vialet-Chabrand S, Matthews JSA, Brendel O, Blatt MR, Wang Y, Hills A, Griffiths H, Rogers S, Lawson T. 2016. Modelling water use efficiency in a dynamic environment: an example using Arabidopsis thaliana. Plant Science 251, 65–74.

Vialet-Chabrand SRM, Matthews JSA, McAusland L, Blatt MR, Griffiths H, Lawson T. 2017a. Temporal dynamics of stomatal behavior: modeling and implications for photosynthesis and water use. Plant Physiology 174, 603–613.

von Caemmerer S, Lawson T, Oxborough K, Baker NR, Andrews TJ, Raines CA. 2004. Stomatal conductance does not correlate with photosynthetic capacity in transgenic tobacco with reduced amounts of Rubisco. Journal of Experimental Botany 55, 1157–1166.

Wang P, Song CP. 2008. Guard-cell signalling for hydrogen peroxide and abscisic acid. New Phytologist 178, 703–718.

Wang Y, Hills A, Blatt MR. 2014. Systems analysis of guard cell membrane transport for enhanced stomatal dynamics and water use efficiency. Plant Physiology 164, 1593–1599.

Wang Y, Noguchi K, Terashima I. 2011. Photosynthesis-dependent and independent responses of stomata to blue, red and green monochromatic light: differences between the normally oriented and inverted leaves of sunflower. Plant & Cell Physiology 52, 479–489.

Weise A, Lalonde S, Kühn C, Frommer WB, Ward JM. 2008. Introns control expression of sucrose transporter LeSUT1 in trichomes, companion cells and in guard cells. Plant Molecular Biology 68, 251–262.

Willmer C, Fricker M. 1996. Stomata. Dordrecht: Springer Science & Business Media.

Wong SC, Cowan IR, Farquhar GD. 1979. Stomatal conductance correlates with photosynthetic capacity. Nature 282, 424–426.

Yamauchi S, Takemiya A, Sakamoto T, Kurata T, Tsutsuji T, Kinoshita T, Shimazaki K. 2016. The plasma membrane H+-ATPase AHA1 plays a major role in stomatal opening in response to blue light. Plant Physiology 171, 2731–2743.

Yamamoto Y, Negi J, Wang C, Isogai Y, Schroeder Jr, Iba K. 2016. The transmembrane region of guard cell SLAC1 channels perceives CO2 signals via an ABA-independent pathway in Arabidopsis. The Plant Cell 28, 557–567.

Zait Y, Shapiro O, Schwartz A. 2017. The effect of blue light on stomatal oscillations and leaf turgor pressure in banana leaves. Plant, Cell & Environment 40, 1143–1152.

Zeiger E. 1983. The biology of stomatal guard cells. Annual Review of Plant Physiology 34, 441–474.

Zeiger E. 1984. Blue light and stomatal function. In: Senger H, ed. Blue light effects in biological systems. New York/Tokyo: Springer-Verlag, 484–494.

Zeiger E, Hepler PK. 1977. Light and stomatal function: blue light stimulates swelling of guard cell protoplasts. Science 196, 887–889.

Zeiger E, Taibd FF, Frechilla S, Srivastava A, Zhu J. 2002. The guard cell chloroplast: a perspective for the twenty-first century. New Phytologist 153, 415–424.

Zeiger E, Zhu J. 1998. Role of zeaxanthin in blue light photoreception and the modulation of light–CO2 interactions in guard cells. Journal of Experimental Botany 49, 433–442.

Zhang J, Wang N, Miao Y, Hauser F, McCammon JA, Rappel WJ, Schroeder Jr, 2018. Identification of SLAC1 anion channel residues required for CO2/bicarbonate sensing and regulation of stomatal movements. Proceedings of the National Academy of Sciences, USA 115, 11129–11137.

Zhu XG, Long SP, Ort DR. 2008. What is the maximum efficiency with which photosynthesis can convert solar energy into biomass? Current Opinion in Biotechnology 19, 153–159.