Planta (2025) 261:124
https://doi.org/10.1007/s00425-025-04694-1

REVIEW

Here comes the sun: integration of light, temperature, and auxin during herbaceous plant grafting

Ruiduo Han[1] · Rui Lin[1] · Yanhong Zhou[1,2,3] · Hannah Rae Thomas[1]

Received: 17 January 2025 / Accepted: 8 April 2025 / Published online: 2 May 2025
© The Author(s) 2025

Abstract

Main conclusion **Light and temperature can regulate auxin production which has been recently shown to be key during graft healing, suggesting that abiotic factors may be vital variables for future graft studies.**

Abstract Grafting is an important horticultural tool used to combine advantageous plant traits. Despite its broad usage, the mechanisms that underlie graft healing remain poorly understood. Recent work has highlighted the influence of high temperature-mediated auxin flow on graft success. Light and temperature sensing utilize partially overlapping mechanisms to regulate auxin biosynthesis, signaling, and transport. In this review, we explore the sensors and transcriptional regulators that modulate auxin response, specifically emphasizing how these components regulate graft success and vascular reconnection. We also discuss areas of graft biology regulated by auxin and underexplored areas of photobiology that may be key to a better understanding of graft mechanisms. This review underscores the importance of translating genetic findings from model systems into horticultural crops to expand our knowledge of economically valuable techniques like grafting.

Keywords Grafting · Auxin · Light · Temperature · Sugars · Signaling

Introduction to plant grafting

Plant grafting is a traditional horticultural technique used to combine distinct plant parts, where the apical portion of a graft is known as the scion, and the root is the (root) stock. Humans have been grafting for thousands of years to cultivate species that are difficult to grow (Mudge et al. 2009). Traditionally, woody fruit crops have been grafted to quickly cultivate desirable varieties onto existing stocks (Bradley and Garner 2017; Wang et al. 2019). Plant architecture and yield can also be altered through this process for optimum crop production (Donadio et al. 2019; Kundariya et al. 2020). Grafting can also confer abiotic and biotic disease resistance, either using resistant rootstocks or through graft-mobile signals, likely carried through the vascular tissue (King et al. 2008). In the last 50 years, herbaceous grafting has become common in Solanaceae and Cucurbitaceae vegetable crops (Lee et al. 2010).

Until recently, the mechanisms that underpin graft junction formation remained elusive. Only in the last few decades has graft biology made strides toward understanding this unique phenomenon, largely through the introduction of Arabidopsis (*Arabidopsis thaliana*) micrografting as an accelerated laboratory model and tomato (*Solanum lycopersicum*) grafting as an applied horticultural system (Melnyk et al. 2015; Thomas et al. 2022). In commercially grafted plants, optimal temperature and light conditions determine graft success. In the early days following wounding, the

Communicated by Gerhard Leubner.

✉ Yanhong Zhou
yanhongzhou@zju.edu.cn

✉ Hannah Rae Thomas
HR.Thomas@zju.edu.cn

Ruiduo Han
hrd_avocado@nwafu.edu.cn

Rui Lin
ruilin@zju.edu.cn

[1] Department of Horticulture, Zhejiang University, Hangzhou 310058, China

[2] Yazhou Bay Science and Technology City, Hainan Institute, Zhejiang University, Sanya 572025, China

[3] Key Laboratory of Horticultural Plant Growth and Development, Agricultural and Rural Ministry of China, Zhejiang University, Hangzhou 310058, China

newly severed scion is susceptible to desiccation unless properly handled (Zeist et al. 2020). Strategies to reduce water stress include high humidity and low light until vascular connectivity is reestablished; often, plants recover in specialized structures known as acclimation chambers (Bradley and Garner 2017). These conditions differ significantly from Arabidopsis, which is capable of high graft success rates under day/night light cycles (Melnyk 2017). Recent work has shown that in addition to simple water hydraulics, light and temperature also affect graft success through more dynamic means, such as hormonal signaling. Furthermore, since many commercially grafted plants are grown in glasshouses and other regulated spaces, the ability to customize light and temperature to optimize grafting success is an attainable goal in commercial horticulture (Fig. 1A).

This review will highlight the current understanding of light and temperature perception, its effect on auxin production, and recent advancements in integrating these abiotic factors with graft success. We also discuss related research areas that intersect auxin and graft biology. Furthermore, we focus on the value of horticultural crop systems when studying grafting and note recent work that has successfully utilized molecular knowledge from Arabidopsis to elucidate the mechanisms underlying horticultural production.

Light and temperature sensing utilize overlapping regulatory mechanisms

Light and temperature regulate auxin biosynthesis

Plants are photosynthetic autotrophs, meaning they rely on energy produced through photosynthesis for growth, defense, and reproduction. In addition to acting as the fuel for photosynthesis, light is also an important signal that can transmit information regarding the constantly dynamic environment, changing seasons, and stress. Some of these environmental cues are regulated by shared genetic processes, such as light and temperature, which show significant overlap in their molecular mechanisms.

Natural light comprises a spectrum of visible, far-red, and ultraviolet wavelengths. Light quality is defined as the spectral makeup a plant is exposed to. A specific intensity of light is required for photosynthesis (Shafiq et al. 2021). In contrast, light signaling is extremely sensitive, with low-intensity light capable of triggering quality-induced changes. With the advent of light-emitting diodes (LEDs), research regarding the effect of light quality on plant growth and development has expanded. Light is perceived by photoreceptors, each with a specific spectral range. Red (R) and far-red (FR) light is detected by PHYTOCHROMES (Phys), UV RESISTANCE LOCUS 8 (UVR8) detects ultraviolet-B light (UV-B), and blue light is perceived by multiple photoreceptors, including CRYPTOCHROMES (CRYs) and PHOTOTROPINS (PHOTs; Fig. 1B). Recent work has highlighted the role of light sensors, especially downstream signaling pathways of the Phys, as integral to graft success.

Many components of the light-sensing pathway are also involved in temperature sensing. For example, PhyB is a R light sensor, but it also functions in the perception of high temperature. As a light sensor, the red:far-red (R:FR) ratio converts PhyB between two forms: the inactive form (Pr) and the active form (Pfr) (Alba et al. 2000; Clack et al. 1994). In low R:FR, CONSTITUTIVE PHOTOMORPHOGENIC 1 (COP1) and SUPPRESSOR OF PHYA − 105 (SPA1) form a complex that degrades transcription factors (TFs) such as

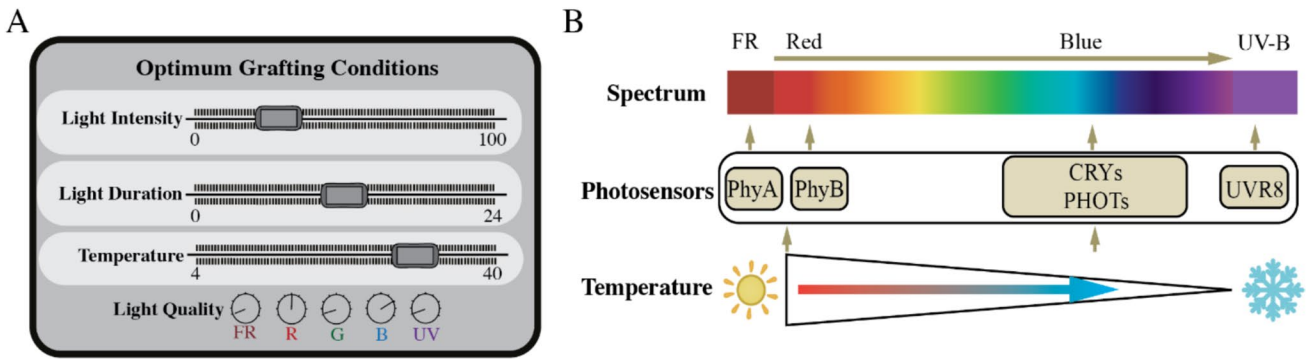

Fig. 1 Plants perceive light and temperature through overlapping sensors, which can be exploited to optimize graft success. **A** A hypothetical control board shows different controllable environmental conditions that can be adjusted to facilitate the most efficient grafting environment. Grafted plants require a specific set of environmental conditions to facilitate graft healing. Abiotic factors such as light intensity, duration, quality, and temperature can be manipulated in an artificial growth environment. To optimize graft conditions, more research is needed to understand how these conditions regulate graft healing and success. **B** A simplified schematic shows the overlap between light and temperature sensing in plants. The light spectrum contained within white light spans far-red (FR), red, orange, yellow, green, blue, and ultra-violet (UV-B). FR light is perceived by PhyA, red light by PhyB, blue light by CRYs and PHOTs, and UV-B by UVR8. Primarily high temperatures are perceived by PhyB (but also CRYs), and cold temperatures by PHOTs and CRYs (but also Phys)

ELONGATED HYPOCOTYL 5 (HY5) (Leivar and Monte 2014). PHYTOCHROME-INTERACTING FACTORs (PIFs), important transcription factors (TFs) involved in photo-sensing, accumulate in the nucleus, which triggers shade avoidance syndrome (SAS)-related gene expression, such as auxin biosynthesis (Leivar and Monte 2014; Franklin et al. 2011; Lee et al. 2021). During high R:FR, light-activated PhyB moves to the nucleus to interfere with the COP1-SPA1 complex (Sheerin et al. 2015; Chen et al. 2015; Lu et al. 2015; Podolec and Ulm 2018). With the COP1-SPA1 complex nonfunctional, TFs such as HY5 can accumulate and activate light-responsive gene expression, while *PIF* activity and accumulation are significantly decreased (Al-Sady et al. 2006; Park et al. 2012). In this way, PhyB regulates transcriptional activity through sensitive light sensing (Fig. 2A).

PhyB also acts as a thermo-sensor, where PhyB is switched into the Pr inactive form during heat stress, and the Pfr half-life is increased during colder temperatures (Jung et al. 2016; Legris et al. 2016). During heat, with PhyB inactive, PIFs can accumulate and regulate gene expression (Fig. 2A). A central component during both high-temperature and low R:FR response is *PIF4* (Lorrain et al., 2008; Koini et al. 2009) PIF4 accumulation allows for increased hypocotyl elongation, whereas HY5 acts as an antagonist (Bellstaedt et al. 2019; Delker et al. 2014; Gangappa and Kumar 2017). Research has previously shown that *PIF4* and *PIF5* respond to light and temperature (Nozue et al. 2011) and that PIF4, 5, and 7 regulate auxin biosynthesis by directly binding to the promoters of key auxin biosynthesis genes such as *TRYPTOPHAN AMINOTRANSFERASE of ARABIDOPSIS 1 (TAA1)* and *YUCCA 8 (YUC8)* (Franklin et al. 2011; Hornitschek et al. 2012; Li et al. 2012; Sun et al. 2012).

The indole-3-pyruvic acid (IPA) pathway is tightly linked to light processes as tryptophan is synthesized in the chloroplast (Radwanski and Last 1995). During auxin biosynthesis, TAA1/TRANSPORT INHIBITOR RESPONSE 2 (TIR2) converts tryptophan into IPA (Tao et al. 2008). The expression level of *TAA1/TIR2* is elevated under high temperatures, in part due to regulation by PIF4 (Franklin et al. 2011; Yamada et al. 2009). IPA is then converted into the primary active auxin molecule, indole-3-acetic acid (IAA), by YUCs (Mashiguchi et al. 2011). *YUC8*, which is induced during low R:FR and high temperature to promote auxin synthesis, is also positively regulated by PIF4, 5, and 7 (Fiorucci et al. 2020; Franklin et al. 2011; Hornitschek et al. 2012; Sun et al. 2012). PIF4 also downregulates IAA conjugation by inducing *YUC8* expression and repressing transcription of the *UDP-glycosyltransferase, UGT76 F1*, which can act on IPA to form conjugated IPA-Glc (Chen et al. 2020).

Temperature-regulated auxin affects graft formation

Grafting to alleviate abiotic stress is an ongoing field, yet a basic understanding of how these environmental conditions affect graft success remains poorly understood. Tomato

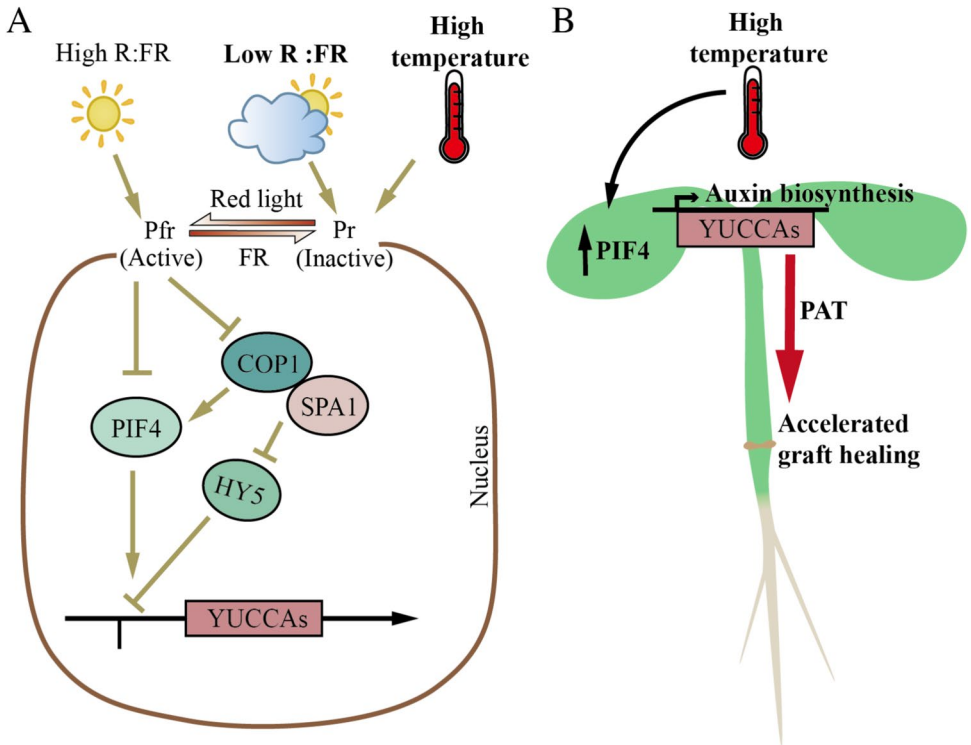

Fig. 2 PhyB regulates PIF4-mediated auxin accumulation during grafting. **A** During high R:FR light exposure, PhyB transitions into the active Pfr form and translocates to the nucleus, interfering with the COP1-SPA1 complex. With COP1-SPA1 deactivated, HY5 accumulates, while PIF4 is deactivated. This regulatory network represses SAS during light exposure. In contrast, low R:FR and high temperatures induce the inactivation of PhyB. In this state, the COP1-SPA1 complex represses HY5 and induces *PIF4*. PIF4 promotes auxin biosynthesis via *YUC* genes. **B** In grafted plants grown under high temperatures, PIF4-mediated auxin biosynthesis undergoes vascular and polar auxin transport (PAT) to the graft junction accelerating healing

seedlings typically recover around 23–25 ℃ following grafting. (Zeist et al. 2020). Elevated temperatures (30 ℃ or above) have been shown to increase reactive oxygen species (ROS) and antioxidant activity (Muneer et al. 2016). While most physiological studies advise against elevated graft recovery temperatures for vegetables, recent work has shown that elevated temperature in the first few days following grafting can accelerate callus production and vascular connectivity (Bartusch et al. 2020; Meng et al. 2025; Serivichyaswat et al. 2022).

In Arabidopsis, high-temperature perception in the cotyledons requires PIF4-mediated auxin production to accelerate vascular connections; *pif4* mutants, while capable of grafting, lose high-temperature thermo-responsiveness (Serivichyaswat et al. 2022). This temperature-mediated process was further shown to be due to increased scion auxin via YUC2, 5, 8, and 9. Mutations to *YUC*s did not affect graft success, although these mutants did show delayed vascular connectivity, but a complete blockage to auxin responses using the *INDOLE-3-ACETIC ACID 12* (*IAA12;* also known as *BONDELOS (BDL))* dominant negative mutant showed a significant reduction in graft success at all temperatures (Serivichyaswat et al. 2022). Similarly, elevated temperatures (35℃) in tomato led to SlPhyB- and SlPIF4-dependent accelerated vascular connectivity (Meng et al. 2025). Furthermore, SlPIF4 was shown to directly bind to *SlYUC4, 10, 11*, and *13*. Thus, elevated temperatures in both tomato and Arabidopsis affect vascular connectivity by increasing scion auxin content (Fig. 2B). However, thermo-sensor mutants are still graft-capable, suggesting that other forms of auxin, such as shoot apical meristem-derived, may be sufficient for graft success. It is also important to note that while these experiments have shown that high temperatures can increase vascular connectivity, they fail to demonstrate whether elevated temperatures show any long-term benefits for grafted crops. Indeed, the tradeoff between increased auxin content in the initial days after grafting and heat stress may not pay off. Instead, it is of future interest to test if increased auxin flux in the scion can rescue graft failures such as incompatible partners.

The intersection of auxin signaling and grafting

Auxin is a critical phytohormone involved in many developmental processes, including lateral and adventitious root formation (Bhalerao et al. 2002; Della et al. 2013; Fattorini et al. 2017), cell growth and division (Spartz et al. 2012), tissue and organ patterning (Friml et al. 2003; Mattsson et al. 1999) and tropic responses (Friml et al. 2002). Since it was first discovered, auxin has been intrinsically linked to light signaling. Francis and Charles Darwin determined that some "influence," later known to be auxin, was moving from the growing tip to control phototropism, a process now known as polar auxin transport (PAT). Auxin is now known to be an integral downstream player in light and temperature signaling.

Auxin signaling requires a complex regulatory network. In general, high auxin levels inhibit the repressive action of AUXIN/INDOLE-3-ACETIC ACID (AUX/IAAs), which bind and inhibit the action of AUXIN RESPONSE FACTORS (ARFs). Auxin binds to the auxin receptor, $SCF^{TIR1/ARBs}$, which then binds to AUX/IAAs; the bound AUX/IAAs are then ubiquitinated for proteolysis (Fig. 3) (Dharmasiri et al. 2005). Free from repression, ARFs can act as both promoters and repressors of auxin-responsive genes (Roosjen et al. 2018). For example, ARFs can promote the expression of auxin transporters, other *AUX/IAAs*, and even *SMALL AUXIN UP-REGULATED RNA (SAURs*), which are rapid auxin-responsive genes (Knauss et al. 2003). ARF7 and 19 promote the expression of *SAUR19*, which has been shown to induce cell elongation critical for tropism via cell wall acidification (Chae et al. 2012; Spartz et al. 2014; Wang et al. 2020). Components of the auxin receptor, *TRANSPORT INHIBITOR RESPONSE 1 (TIR1)* and *AUXIN SIGNALING F-BOX PROTEINs (AFBs)*, are further regulated by light. Expression of *TIR1* and *AFB2* increase in the dark due to the reduced presence of *miR393*, which represses the receptor genes to control auxin sensitivity (Navarro et al. 2006; Pucciariello et al. 2018). In addition to their role in auxin biosynthesis, PIFs can also interact with ARFs (Jia et al. 2020).

While auxin is historically associated with apical biosynthesis, many tissues can synthesize the key hormone (Ljung et al. 2001). Young leaves, cotyledons, and the shoot apical meristem are all sources of auxin, which is then transported basipetal to regulate a multitude of processes, including grafting (Procko et al. 2014). Numerous commercially grafted species show an auxin-related transcriptional response, such as *Torreya grandis* (Yuan et al. 2017), pecan (*Carya illinoinensis)* (Mo et al. 2023), Chinese hickory *(Carya cathayensis)* (Mei et al. 2024; Qiu et al. 2016), tomato (Xie et al. 2019), Norway spruce (*Picea abies*) (Feng et al. 2024), grape (*Vitis vinifera)* (Assunção et al. 2019), and apple (*Malus robusta)* (Li et al. 2016). Despite the prevalence of auxin-related genes differentially expressed following horticultural grafting, auxin-related graft mechanisms have largely been explored in Arabidopsis only.

Auxin transport

Auxin can utilize the phloem or plasmodesmata to migrate (Cambridge and Morris 1996; Gao et al. 2020), but significant amounts of auxin translocate cell-to-cell via transporters during PAT. Auxin is actively pumped out of cells

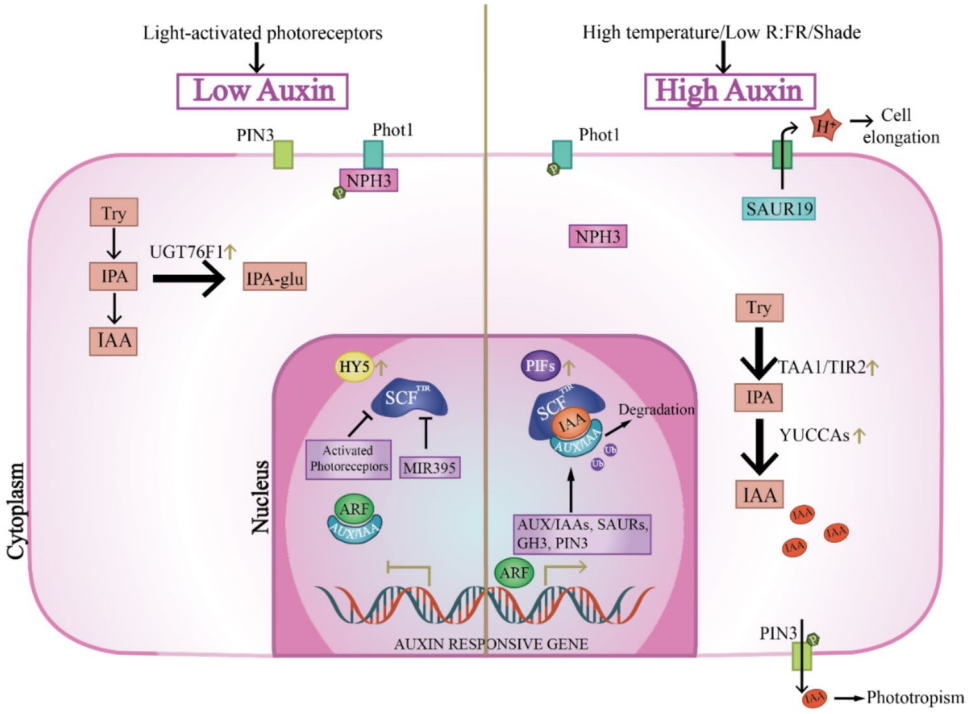

Fig. 3 High auxin levels lead to transcription of auxin-responsive genes. During periods of high light, photoreceptors are activated and move into the nucleus. HY5 accumulates to activate light-responsive genes, and the auxin receptor ($SCF^{TIR1/ARBs}$) is repressed by activated photoreceptors and *miR395*. ARFs are bound by AUX/IAAs, which repress their transcriptional activity. Meanwhile, in the cytoplasm, auxin intermediates are channeled into conjugated forms through reversible reactions. All of these conditions lead to low levels of auxin. During low light or elevated temperature, photoreceptors are deactivated, which allows PIF levels to accumulate in the nucleus. PIFs activate the transcription of auxin biosynthesis genes such as *TAA1/TIR2* and *YUCs*, which lead to increased active IAA levels. Elevated auxin levels then enter the nucleus, binding to $SCF^{TIR1/ARBs}$ and AUX/IAAs. Through this process, AUX/IAAs are marked for ubiquitin-mediated degradation. ARFs are then free to bind to auxin-responsive genes to activate or repress transcription. One type of gene which is stimulated by ARFs are *SAURs*. *SAUR19*, for example, is expressed during high auxin conditions, leads to cell wall acidification and elongation. Another change that happens in low light is the reallocation of PIN transporters. During high blue light, activated PHOT1 binds to NPH3 at the membrane, but during low light, NPH3 is released into the cytoplasm, leading to PIN3 endocytosis and relocation to the shaded side of the cell. With PIN3 located away from the light source, a local auxin maximum accumulates, leading to phototropism

by PIN-FORMED (PIN) or B-type ATP-binding cassette (ABCB) family exporters (Gälweiler et al. 1998; Geisler et al. 2005). PIN exporters have polar localization that determines the direction of auxin transport. For example, the well-studied PIN1 localizes to the basal end of auxin-transporting cells, leading to strong downward auxin mobility (Gälweiler et al. 1998). Auxin is actively imported by the AUXIN1/LIKE-AUX1 (AUX1/LAX) importers (Yang et al. 2006). (Fig. 4A). Light has a strong effect on the localization of PIN proteins. The most well-studied example is the localization of PIN3 during hypocotyl hook formation, leading to the reorientation of PIN3 from basipetal to lateral transport, asymmetrically directing auxin to the dark side of the plant hypocotyl (Ding et al. 2011) (Žádníkova et al. 2010).

Arabidopsis auxin efflux proteins PIN1 and ABCB1 accumulate in the scion after grafting, likely acting to pump auxin across the graft site and inducing vascular reconnections (Melnyk et al. 2018; Wang et al. 2014) Similarly, in pea (*Pisum sativum*), wounding triggers reorientation of PIN1 polarity above the wound (Sauer et al. 2006). Perturbations to auxin transport, such as treatment with auxin transport inhibitors (i.e., naphthylphthalamic acid), can significantly reduce auxin accumulation at the junctions and thus slow or inhibit graft connectivity (Wang et al. 2014). In grafted apple, a more-branching (MB) mutant scion reduced root growth, possibly due to scion-controlled reductions of *PIN1* and *ARF* expression in the root (Li et al. 2016). In tomato, auxin levels were found to have their highest expression in the scion; Prior to vascular formation, auxin levels were positively correlated with increased expression of *SlIAAs*, *SlARFs, PINs*, and *SlLAX2* (Cui et al. 2021; Duan et al. 2024).

While PAT plays a clear role in graft formation, endogenous auxin levels in the root are also important. In tobacco (*Nicotiana tabacum)*, auxin levels were modulated via tryptophan-2-monooxygenase auxin biosynthesis

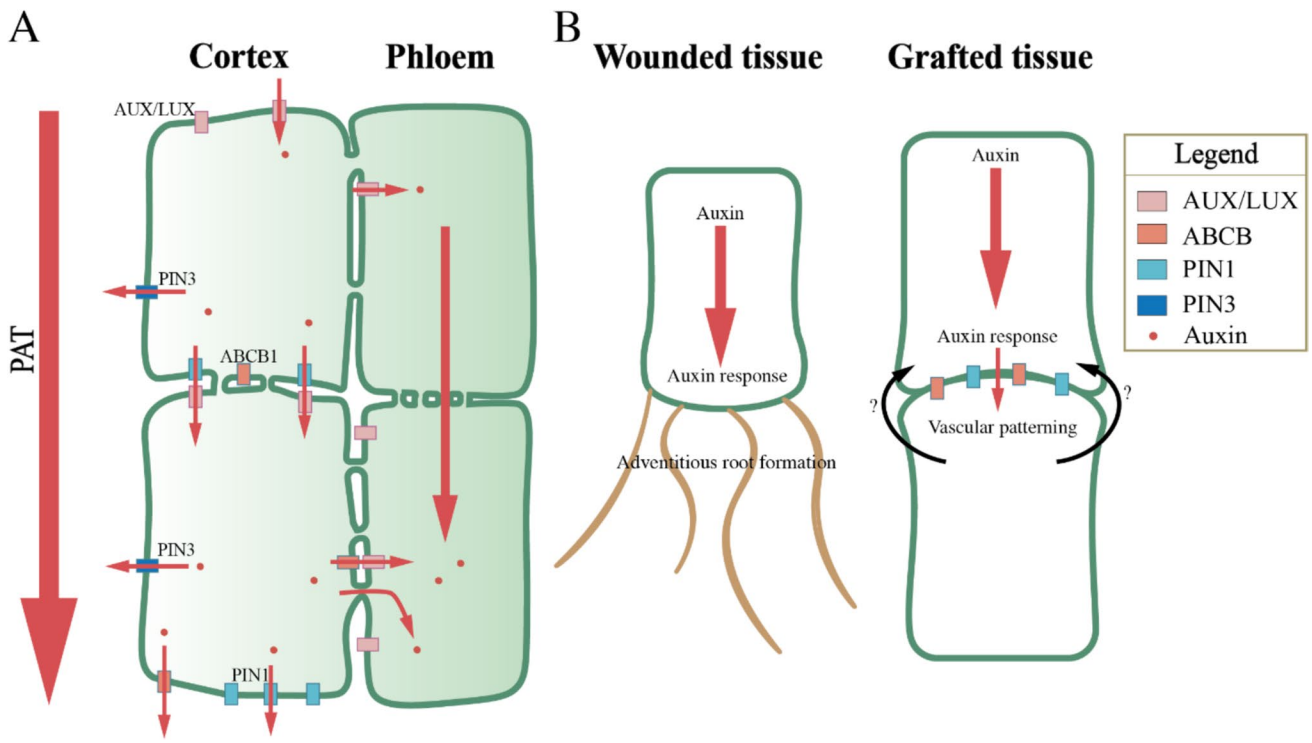

Fig. 4 Auxin is translocated cell-to-call to generate auxin gradients that promote adventitious roots or graft junctions. **A** Auxin generally moves basipetal from the growing tip to the roots. Bulk flow in the phloem and plasmodesmata facilitate symplastic cellular movement of auxin. Auxin can also be actively transported through transporters. Auxin is imported into cells by AUX/LUX importers and exported by ABCB1 and PIN exporters. PINs display polar localization, with PIN1 directing downward auxin flux and PIN3 critical for lateral auxin movement during phototropism. Together, all these mechanisms combine to achieve polar auxin transport. **B** During wounding without grafting, auxin accumulates at the cut site, where an auxin maximum induces adventitious root formation. In grafted tissue, newly expressed exporters pump auxin across the graft site. The scion and stock experience auxin responses that induce healing and differentiation of vascular tissue. In contrast to wounded tissue, an unknown mechanism from the stock may trigger vascular reconnection rather than root formation

or indoleacetic acid-lysine synthetase-mediated auxin conjugation (Li et al. 2017; Zhai et al. 2021). Higher levels of auxin in the root increased callus, cellular connectivity, and scion growth (Zhai et al. 2021). Despite basipetal auxin transport being the dominant direction, auxin-enriched rootstocks showed the upward movement of auxin into the scion, even prior to vascular connectivity. In watermelon (*Citrullus lanatus*), the removal of the cotyledons, an auxin source tissue, from the rootstock significantly reduced vascular connectivity and callus formation (Wang et al. 2024). Like Arabidopsis, ClPIN1 in watermelon cotyledons was key for auxin-induced graft healing (Matsuoka et al. 2016; Wang et al. 2024). The role of cotyledon-derived auxin during grafting supports previous work that found cotyledons were necessary for wound healing in tomato and cucumber (*Cucumis sativus*), although this work also identified the role of cotyledon-derived gibberellins (Asahina et al. 2002, 2007). Indeed, recent work, including mathematical modeling, has suggested that PAT alone is not capable of describing the auxin transport required for vascular reconnections following grafting and that other hormones are likely involved (Ongaro et al. 2008; Prusinkiewicz et al. 2009).

It has been known for quite some time that auxin transport is repressed at low temperatures and that higher temperatures correlate with increased PAT velocity (Morris 1979; Shibasaki et al. 2010). Reduced PAT at cold temperatures has been linked to inhibited intercellular trafficking and localization of PINs, but the exact mechanism underlying this remains elusive (Shibasaki et al. 2010). Outside of the location of PINs, much remains unknown about how light and temperature regulate PAT, and especially during grafting.

Auxin-induced wound response

PAT is also vital for the appropriate auxin response that triggers graft healing. Both grafted scions and separated apical parts trigger auxin responses, but only grafted rootstocks showed auxin responsiveness, highlighting how PAT from the scion to the stock is a key physiological step in grafting (Asahina et al. 2011; Melnyk et al. 2018). This is

curious, since, in the absence of a rootstock, PAT leads to adventitious root (AR) formation at the cut site (Fig. 4B). ARs are roots that form from any part of the plant other than the embryonic primary root. When explants are removed from their root system, auxin accumulates, triggering the formation of a new stem cell niche (SCN), from which roots form (Rovere et al., 2016; Fattorini et al., 2017). There is quite a lot of overlap between the formation of adventitious roots and graft healing, posing the question of: what differentiates wounding from grafting? Mere hours after grafting, auxin exporters were expressed in the cut scion surface (Melnyk et al. 2017). Similarly, in apple cuttings, all 8 MdPINs were differentially expressed during AR formation (Guan et al. 2020). Instead, it is likely that a signal from the stock or auxin diffusion across the junction is key to inducing vascular reconnection instead of AR formation (Fig. 4B). Perhaps a hormonal or other mobile signal induces a recognition event at the graft junction that facilitates grafting-required auxin signaling. While recent work has approached this question, the underlying mechanism remains unknown.

Research on wounding has shown that PAT in the Arabidopsis inflorescence stems facilitated healing, thus setting up this new tissue as a model to explore auxin-regulated wound healing (Mazur et al. 2016). Several important genes related to grafting were identified using this system. Following wounding, auxin immediately accumulates above the wound, stimulating healing through PAT from the apex (Asahina et al. 2011). The wound acts as a block to vascular auxin transport, so auxin must rely on PAT to pass cell-to-cell at the wound site (Yin et al. 2012). As auxin moves from the shoot to the root, various genes are regulated for healing. For example, high auxin above the inflorescence stem wound stimulates *NAC DOMAIN CONTAINING PROTEIN 71* and *96 (ANAC071* and *96*) expression, while lower auxin levels below the wound promote *RELATED TO AP2 6L (RAP2.6L)* expression (Asahina et al. 2011; Matsuoka et al. 2021). ANAC071 and RAP2.6L are necessary for non-vascular proliferation, while ANC071 and 96 are also crucial for wound-induced cambium formation (Matsuoka et al. 2021). Downstream of ANAC071 are *XTH19* and *20*, members of the xyloglucan endotransglucosylase/hydrolases family involved in cell enlargement (Pitaksaringkarn et al. 2014). Much like ANAC071, XTH19 and 20 are auxin-dependent regulators of pith connectivity. It is important to note that while similar to grafting, the inflorescence system does not always translate into the stem. For example, when *rap2.6L* mutants were used in Arabidopsis stem grafting, healing was unaffected, highlighting how tissue- and species- specific studies are needed to understand graft healing (Matsuoka et al. 2018).

The role of PAT and graft success

Auxin transport is also critical to graft healing. During graft healing, auxin diffuses into the auxin-depleted stock. As auxin is transported into the Arabidopsis stock, auxin-responsive genes such as *ABERRANT LATERAL ROOT FORMATION 4 (ALF4), AUXIN RESISTANT 1 (AXR1),* and *HIGH CAMBIAL ACTIVITY 2 (HCA2)* are initiated (Melnyk et al. 2015, 2018). When *alf4* or *arx1* mutants are used as rootstocks, auxin responses are significantly reduced below the junction, which impedes phloem connectivity (Melnyk et al. 2015). Similarly, suppression of HCA2 targets in the stock led to delayed phloem connectivity, emphasizing the critical role auxin plays in the spatial regulation of phloem reconnection (Melnyk et al. 2018). Further focus on *HCA2*, a member of the DOF family, identified other cambium and vasculature-related DOF TFs, including *TARGET OF MONOPTEROS 6 (TM06), DNA-BINDING ONE ZINC FINGER 2.1 (DOF2.1)*, and *DOF6*, which were all upregulated in grafting or separated apical parts in an auxin-dependent manner (Zhang et al. 2022). A quadruple mutant of these AtDOFs showed significant relevance to graft healing, with reduced vascular regeneration, wound healing, and callus formation. Furthermore, *CELLULASE 3 (CEL3)* was identified as a direct target of TMO6 (Zhang et al. 2022). *Atcel3* mutants showed delayed phloem and xylem connectivity (Zhang et al. 2022). *CEL3* (also known as *GLYCOSYL HYDROLASE 9B3*; *GH9B3*) *is a* β−1,4-glucanases, which degrades cellulose to aid in cellular growth and has recently been identified as an important gene during interfamily grafting (Notaguchi et al. 2020), suggesting that auxin regulation may play a key role in graft formation as well as compatibility.

The role of auxin in the cambium was further elucidated. Blocking auxin signaling under procambium (*PHLOEM INTERCALATED WITH XYLEM (PXY)* and *HOMEOBOX GENE 8 (ATHB8)*) and cambial phloem-precursor (*SUPPRESSOR OF MAX2 1-LIKE 5 (SMXL5)*) specific promoters in Arabidopsis significantly reduced graft success by interfering with tissue attachment and phloem healing (Fig. 5) (Serivichyaswat et al. 2024). This is profound as it suggests that within the cambium, specific niches of cells expressing *PXY*, *ATHB8*, or *SMLX5* lead to callus, phloem, or xylem during graft formation (Fig. 5C). Similarly, Sl*WUSCHEL HOMEOBOX RELATED 4 (SlWOX4)*, an important gene in cambial-xylem maintenance, was found to be required for xylem reconnection in tomato (Thomas et al. 2022). Although self-grafted *wox4* mutants could form non-vascular connections, xylem formation was overproliferated and non-cohesive, leading to graft failure. In Arabidopsis, blocking auxin in cells expressing *WOX4* did not interfere with grafting, which posits that while auxin-dependent cambial stimulation requires WOX4, blocking auxin signaling in

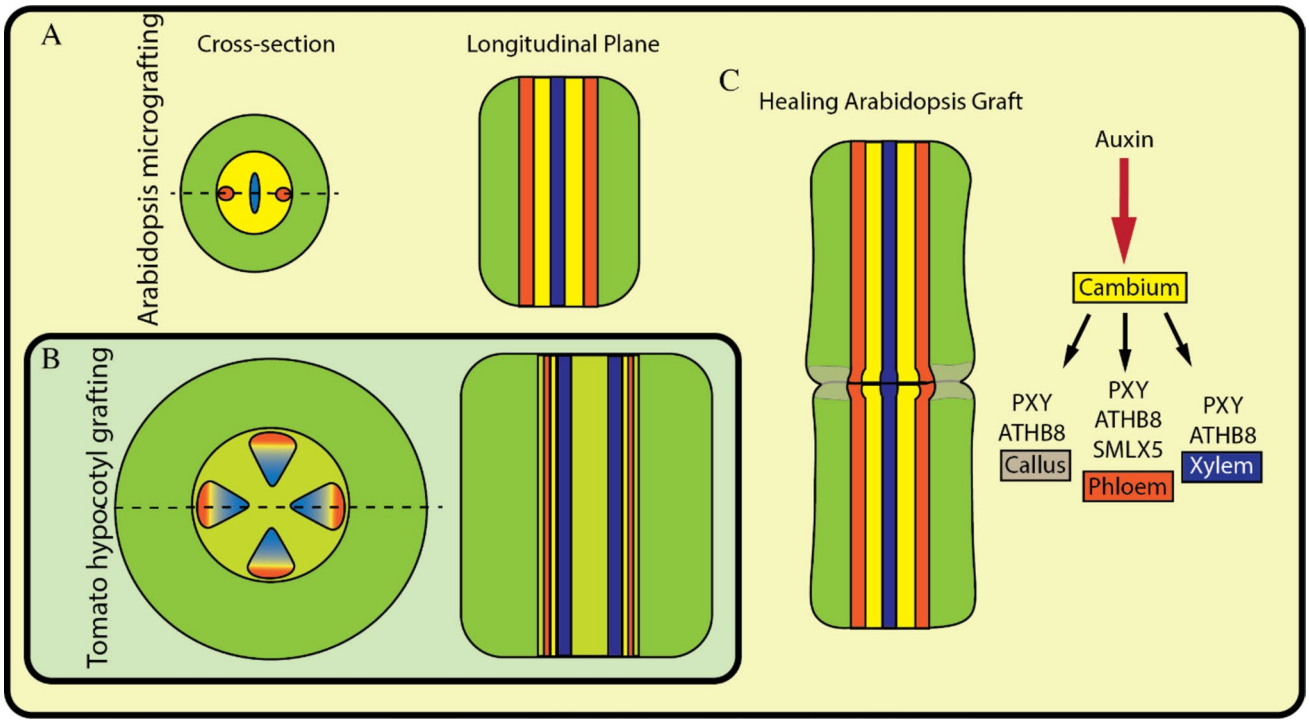

Fig. 5 Auxin signaling in the cambium is necessary for graft healing. In Arabidopsis micrografting, the hypocotyl stele contains two phloem poles, a central xylem region, and a more diffuse cambial region. **B** In tomato seedling grafts, the hypocotyl has developed vascular bundles, where the cambium is sandwiched between newly forming xylem and phloem cells. These different anatomies likely affect graft healing. **C** In Arabidopsis grafting, auxin signaling in distinct cell niches expressing a subset of cambial markers such as *PXY*, *ATHB8*, or *SMLX5* are required for callus, phloem, and xylem formation. For example, while auxin signaling in *PXY*-expressing cambial cells is required for callus, phloem, and xylem reconnection, auxin signaling in *SMLX5*-expressing cambial cells is only necessary for phloem healing

these cells fails to disrupt tissue adhesions completely (Suer et al. 2011; Serivichyaswat et al. 2024).

Another important distinction between tomato and Arabidopsis is the developmental stage during hypocotyl grafting. During Arabidopsis micrografting, the vascular tissue in the hypocotyl only has two phloem poles, and cambial expression covers a large portion of the stele (Fig. 5A) (Lehmann and Hardtke 2016; Serivichyaswat et al. 2024). This differs from young tomato seedlings, which have already formed vascular bundles, each with a layer of cambium cells (Fig. 5B) (Thomas et al. 2023). The positioning and alignment of cambium are thus different in these two types of grafting and are worth considering when comparing gene function between the two systems, such as with *WOX4*.

The importance of auxin to graft-induced meristematic tissue is further highlighted by *ETHYLENE RESPONSE FACTOR 114* and *115* (*ERF114* and *115*), which are also upregulated following grafting in Arabidopsis, spruce, pepper (*Capsicum annuum*), and tomato (Feng et al. 2024; Thomas et al. 2024; Zhang et al. 2022). *ERF115* was originally identified as an essential component of the regenerative properties of roots (Heyman et al. 2013). Following wounding, ERF115 sensitizes nearby cells to auxin by activating *ARF5/MONOPTEROS (MP)* to coordinate the formation of a new SCN (Canher et al. 2020). ERF115 and its homolog ERF114 were found to interact with *SCARECROW-LIKE21 (SCL21)* and *PHYTOCHROME A SIGNAL TRANSDUCTION1 (PAT1)* to promote SCN generation and callus production (Fig. 6) (Heyman et al. 2016). SCL21-PAT1, identified initially as FR light signaling components, can also regulate regeneration by activating *DOF3.4* and *IAA5* (Bisht et al. 2023; Torres-Galea et al. 2013). *erf114, erf115,* and *pat1* mutants showed reduced callus, phloem connectivity, and tissue attachment in Arabidopsis (Feng et al. 2024; Zhang et al. 2022). Additionally, ERF115-PAT1 may act upstream of *WOUND INDUCED DEDIFFERENTIATION 1 (WIND1)*, a critical TF upstream of cytokinin responses, to trigger callus formation, but if this is true, *WIND1* likely can also be triggered independently of this pathway as *ERF115* expression is not always present in newly forming callus (Heyman et al. 2016). While integral to callus formation, the role of cytokinin is less well understood during grafting compared to auxin. Following grafting and wounding, cytokinin-responsive genes, *RESPONSE REGULATOR 5*

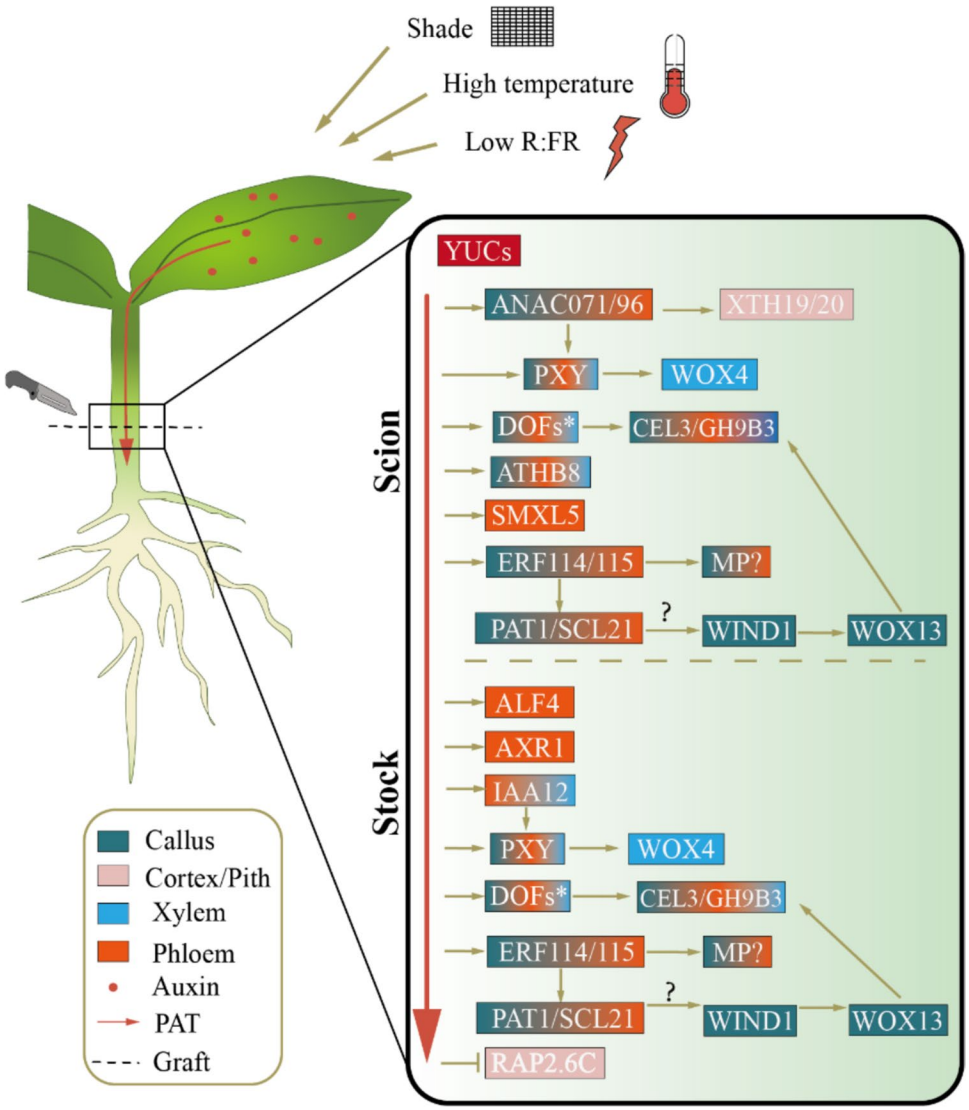

Fig. 6 Auxin regulates gene expression in the graft junction. Auxin from the scion is critical for graft healing. YUCs are required for scion-auxin signaling and graft healing. High auxin levels in the scion induce the expression of *ANAC071* and *96*, which are important for callus and phloem healing. ANC071/96 then induces *XTH19* and *20*. XTH19 and 20 are required for pith reconnection. ANAC71 and 96 also promote the expression of *PXY*, which regulates cambial promotion of callus, phloem, and xylem. PXY is upstream of *WOX4*, which is necessary for xylem formation and organization. Auxin signaling in the cambium is critical for phloem reconnection. DOFs* (*HCA2, TMO6, DOF2.1*, and *DOF6)* are induced following grafting and are necessary for callus, phloem, and xylem reconnection. TMO6 binds directly to *CEL3/GH9B3* during early graft adhesion to aid in callus, phloem, and xylem reconnection. *ERF114* and *115* are induced following wounding and mediate callus and phloem formation through *PAT1/SCL21, WIND1*, and *WOX13*. WOX13 also induces *GH9B3*. High auxin levels in the cambium induce genes such as *ATHB8* and *SMXL5*, where ATHB8 is required for callus, phloem, and xylem, while SMLX5 is only needed for phloem. In the stock, auxin responsive *ALF4 and AXR1* are required for phloem reconnection, while *IAA12* controls the phloem and xylem. In the stock, lower auxin levels promote *RAP2.6L*, which is required for non-vascular healing. The color of the gene highlights its tissue-specific role during graft healing. Gene involved in multiple tissue-types are depicted as a gradient

(ARR5) and *TWO COMPONENT SIGNALLING (TCSn),* are strongly activated in the scion and stock. Despite this, cytokinin mutants such as *wooden leg* (*wol)* and *LONELY GUY7* (*LOG7*) overexpression lines did not display any phenotypes associated with phloem reconnection (Melnyk et al. 2015).

Auxin and graft compatibility

The ability of two plant varieties or species to heal completely following grafting is known as graft compatibility (Moore 1984). Graft incompatibility often manifests as poorly connected vascular tissue between the scion and stock (Jefree and Yeoman 1983; Moore and Walker

1981). The exact determination of graft compatibility has been posited for hundreds of years. More closely related species are more likely to be compatible (Thomas et al. 2023), but there are instances, such as Petunia (*Petunia hybrida*) and tobacco, which show broad compatibility even when grafted to plants from different families (Kurotani et al. 2022; Notaguchi et al. 2020). This hypercompatibility may be due to early adhesion facilitated by *GH9B3* (Notaguchi et al. 2020), but it remains unclear if this gene can be utilized in other species to expand compatibility. Graft compatibility is a serious issue for horticultural production since grafting is an expensive and time-consuming practice. Graft failure due to compatibility poses a significant issue, especially in woody crops, which have slower growth. The role of auxin on graft compatibility has not been clearly explored. For example, cambium-specific auxin signaling has been shown to be crucial for grafting in Arabidopsis (Serivichyaswat et al. 2024), and the cambium-xylem maintenance gene, Sl*WOX4*, is also critical for graft healing in tomato (Thomas et al. 2022). In litchi (*Litchi chinensis)*, compatible grafts accumulated higher levels of auxin, largely due to upregulated biosynthesis genes such as *TAA1* and *YUCs* early during healing, whereas incompatible grafts showed poor healing and unaffected auxin regulation (Chen et al. 2017). Similarly, in pomelo (*Citrus grandis)*, incompatible grafts had reduced auxin signaling components such as *AUX/IAAs* and *SAURs* (He et al. 2018). It is clear that auxin levels increase during grafting, and it appears that incompatible grafts fail to accumulate the appropriate levels of auxin for healing. What remains unknown is if failure to regulate auxin is the cause of incompatibility or a symptom.

Another interesting type of graft incompatibility was identified between two legumes, pea and lupin (*Lupinus angustofolius),* where only pea is capable of arbuscular mycorrhizal (AM) development (Foo et al. 2016). Previous studies showed a directionally specific incompatibility; When AM-forming species were grafted to lupin rootstocks, the resulting plants were incompatible (Gianinazzi-Pearson and Gianinazzi 1992). When lupin was grafted onto pea rootstocks, the plants could survive, but strigolactone exudates and AM were suppressed in the root (Foo et al. 2016). Auxin has a positive effect on AM formation, so PAT was accessed in the grafts. Despite the formation of apparent xylem, radiolabeled ^{3}H-IAA from the scion was blocked at the graft junction, suggesting that the phloem remained poorly connected (Foo et al. 2016; Wulf et al. 2019). The relationship between auxin and compatibility appears to be intimately linked to vascular connectivity, but more research is required to clarify this interaction.

Graft mobile auxin signals

In addition to hormones, plants can also transport other signals over long distances, such as mRNA, proteins, metabolites, electrical signals, and small RNAs (sRNAs) (Choi et al. 2016; Durbak et al. 2012; Kehr and Kragler 2018; Kong et al. 2019; Müller and Harrison 2019; Parent et al. 2012). Grafted plants are excellent model systems to study these mobile signals (Thomas and Frank 2019). Recently, graft-mobile signaling has earned significant interest due to the finding that CRISPR-associated protein 9 (Cas9) and guide RNAs can be deployed into the scion from transgenic rootstocks in Arabidopsis (Yang et al. 2023). Numerous types of genetic information have been reported to pass across the graft junction from nucleic acids, proteins, extrachromosomal circular DNAs, and even whole organelles (Haroldsen et al. 2012; Hertle et al. 2021; Yang et al. 2015; Zhang et al. 2024). For mRNA, methylated and high-abundance transcripts are more likely to be deemed mobile (Calderwood et al. 2016; Yang et al. 2019). However, there are challenges associated with many of these experiments, especially those that rely solely on transcript detection (Heeney and Frank 2023). A recent meta-analysis of published mobile transcript datasets found that limitations in the computation techniques used to identify mobile transcripts generate inherently noisy data with largely false positives.

Nonetheless, it is apparent that mobile signals exist. One such signal, Cyp1, is a graft-mobile protein in tomato. *cyp1* mutants are auxin insensitive, displaying reduced secondary growth, perturbed vascular development, and a lack of lateral roots (Zobel 1973). When *cyp1* was used as a scion, it altered wild-type tomato (WT) roots, but when WT scions were grafted to *cyp1* rootstocks, wild-type-like roots were restored, proving the presence of a SlCyp1-derived signal (Spiegelman et al. 2015). Further experiments were able to show that CYP1 protein accumulation and its subsequent transport in the phloem are induced by light to regulate shoot and root growth by promoting transcription of NAC genes involved in auxin response. Thus, this graft-mobile protein modulates auxin response to coordinate resource allocation.

Grafting can also induce increased yield and growth, a process known as graft-induced vigor (Cerruti et al. 2021). One mechanism to describe this phenomenon is reduced small interfering RNA (siRNA)-directed DNA methylation (RdDM) (Wendte and Pikaard 2017). sRNA are mobile non-coding RNAs that direct cysteine methylation (Molnar et al. 2010). Mutations that interrupt RdDM show increased transgenerational vigor in tomato due to hypomethylation of the auxin-gene pathway, highlighting auxin signaling as central to graft vigor (Kundariya et al. 2020).

The effect of light on grafting

The effect of light quality on horticultural crops

Light quality also affects the growth of plants. Historically, light quality has been challenging to study due to confounding variabilities such as temperature and radiation (Fletcher and Zalik 1964). Indeed, light intensity is critical in these experiments. Low-intensity red light is more inhibitory to hypocotyl elongation than blue, while high-intensity blue light is more inhibitory (Fortanier 1954; Meijer 1959). Light quality exerts regulation on auxin accumulation, which increases hypocotyl height. In pea, blue light-treated seedlings accumulated the most auxin and had the highest hypocotyl height, whereas red light showed the lowest auxin and plant height (Fletcher and Zalik 1964). With the advent of LEDs came a new era of light-based research. Despite significant work in this field, most data appears contradictory. For example, in tomato, red light has been shown to promote hypocotyl elongation, cotyledon expansion, plant height, and leaf area (Izzo et al. 2020). Meanwhile, tobacco had the highest levels of shoot-derived auxin under blue light, while red light increased auxin transport to the roots (Meng et al. 2015). Despite many studies looking at the effect of individual light qualities on plant growth, white or red/blue mixed light is often associated with the best plant growth (Paradiso and Proietti 2022). These diverse findings mirror sentiments made 60 years ago that the main driving determinant of light quality and growth may be species-specific (Fortanier 1954). Additionally, discrepancies between experimental setups, such as light intensity or temperature, could also introduce conflating factors.

Despite the lack of clarity on the role of light quality and its effect on hormonal regulation and growth, horticulturalists have been eager to assay for graft-related benefits associated with distinct light qualities. While most grafted crops fare best after a period of low light treatment, grafts that were left to heal in extended periods of darkness consistently performed the worst across species (Lee et al. 2016; Moosavi-Nezhad et al. 2021; Vu et al. 2014). Still, the relationship between light quality and graft formation show species- and even experimental-based inconsistencies. In Solanaceae and Cucurbitaceae, white, blue, red, and mixtures of light were all capable of graft healing, but the timing of recovery and overall plant growth were affected by the quality. Red light is sometimes associated with slower, less effective graft recovery due to delayed junction attachment (Yousef et al. 2021b) and increased transpiration (Lee et al. 2016). In contrast, other assays found red light to be the optimal treatment during grafting (Bantis et al. 2020; Vu et al. 2014). Blue light was rarely the most beneficial choice for solanaceous graft healing (Moosavi-Nezhad et al. 2021). Much like with plant growth, red/blue and white light were commonly found to be ideal for grafting (Bantis et al. 2020; Jang et al. 2013; Lee et al. 2016; Moosavi-Nezhad et al. 2021; Yousef et al. 2021b,a). Indeed, mixed red and blue light has been associated with higher auxin biosynthesis in some cases (Yousef et al. 2021b). Bantis et al. (2021) found that blue light during the first three days of healing, followed by red light, increased vascular connectivity without the growth penalty associated with prolonged blue light. Interestingly, they found that this condition was associated with reduced auxin and cytokinin levels in the scion.

While a significant body of research has assayed the effect of light quality on grafting, significant experimental discrepancies exist between species and conditions. In addition to the likely possibility that different species respond preferentially to various light conditions, these experiments may have utilized varying light intensity, duration, and other abiotic conditions such as humidity and temperature. In summary, the intersection of light quality and graft success still needs to be explored.

Blue light sensing

Grafting and auxin are intrinsically linked, and auxin is regulated by both light and temperature. While some work has shown links between high temperature and auxin-regulated graft healing, the role of light on grafting remains a black box. PhyB is capable of regulating graft success as a thermosensor (Meng et al. 2025). While not demonstrated, PhyB likely can also regulate grafting through R:FR sensing. Similarly, PAT1 implicates PhyA as potentially involved in graft healing (Feng et al. 2024). In contrast, no molecular sensors related to blue-light have been associated with grafting, despite numerous crop-based research profiling the effect of blue light on growth and graft recovery. Additionally, blue light has been shown to regulate auxin signaling, so efforts to detangle light quality and graft success should remain prioritized in plant biology.

Blue light is involved in many processes and is perceived by blue light sensors, CRYs, and PHOTs. CRYs are photoactivatable nuclear flavoproteins that regulate circadian rhythm, seed germination, leaf senescence, and SAS, among other roles (Bognár et al. 1999; Meng et al. 2013; Pedmale et al. 2014, 2015). When CRYs are treated with blue light, the receptor is oligomerized and phosphorylated to become active. Activated CRYs can directly bind to PIF4 and 5 (Pedmale et al. 2015). CRYs have also been shown to modulate plant thermogenesis. In full light or high blue light, active CRYs repress *PIF4* and *5*, but during low blue light, CRY1 and 2 physically interact with PIF4 and 5 to promote the expression of genes related to hypocotyl growth (Pedmale

et al., 2015). This is also true during elevated temperatures, where elevated PIF4 leads to hypocotyl elongation, but when plants are grown under high temperatures and only blue light, this phenomenon is counteracted. While *CRY1* expression is not regulated by elevated temperature, during exposure high temperatures and only blue light, *PIF4* expression is reduced, which attenuates the expression of YUC genes involved in auxin biosynthesis (Ma et al. 2016). PHOTs are membrane-bound protein kinases that, when phosphorylated under blue light, regulate both stomatal activity and chloroplast movement (Briggs and Christie 2002). PHOTs are also crucial regulators of phototropism. When activated by blue light, PHOT1 leads to NPH3 dephosphorylation and relocalization of PIN3 away from the illuminated side via Calrithin-mediated endocytosis (Fig. 3) (Ding et al. 2011; Folta and Spalding 2001; Keuskamp et al. 2010; Zhang et al. 2017).

While not an exhaustive profile of blue light regulation, these findings highlight how blue light is involved in various processes such as SAS and auxin transport. While no research to date has identified a blue-light sensor or TF as required for grafting, it seems likely that there are unexplored components involved. Future work on the effect of light quality on grafting should focus on how blue light, CRYs, and PHOTs regulate graft healing and success. Understanding the role of light quality has immediate implications on crop production, as environmental conditions can be easily adjusted in greenhouses.

The role of light and sugars on grafted plants

The main function of light is as an energy source, fueling photosynthesis and growth. Because as it is not easy to separate the role of light between signaling and energy generation, the exact role of photosynthesis and metabolism on grafting also remains an open question.

Light and darkness during graft healing

For most horticulturally grafted crops, plants recover for several days in dark or low-light environments following grafting to reduce transpiration. So clearly, the formation of early graft adhesion does not require high light. This observation is logical as many plant species generate callus best in dark environments (Efferth 2019), and indeed, Arabidopsis forms more callus when cultured in the dark (Ikeuchi et al. 2017). During wound-induced callus formation, *WIND1* is quickly induced and, via cytokinin-mediated signaling, triggers callus formation (Ikeuchi et al. 2013; Iwase et al. 2011a,b; Mason et al. 2005). While overexpression of *WIND1* can ectopically induce callus, only by inhibiting all downstream WIND1 targets could wound-induced callus be reduced, suggesting there is redundancy in the callus-forming mechanism (Iwase et al. 2011a). *WIND1* induction activates auxin-related genes such as *YUC4* and *PIN1* (Iwase et al. 2011b, 2021). Furthermore, *WIND1* induction partially regulates *WOX13*, a crucial gene for callus formation, which is induced by PAT (Ikeuchi et al. 2022; Tanaka et al. 2023). WOX13 regulates numerous genes involved in cell wall modification, including *CEL3/GH9B3*.

Similarly to wounding, *WIND1* is strongly upregulated in the scion and stock following grafting; *WIND1* is also induced in cut but not grafted scions but not in the rootstock, suggesting apical auxin may act as the signal to induce *WIND1* expression (Ikeuchi et al. 2022; Melnyk et al. 2015). This finding contrasts with previous work, which has shown that *WIND1* can be upregulated in wounded stocks that were grown on sucrose, suggesting that sugars, in addition to auxin, may be a requirement for *WIND1* expression in Arabidopsis (Iwase et al. 2011a). While a short period in the dark can be beneficial for callus formation, plants fail to heal following grafting if left for prolonged periods in the dark. These outcomes show that early graft processes, such as tissue attachment and callus formation, do not require high levels of light but that for later steps, such as vascular connection, light-induced signaling, and sugars are necessary. In callus explants, light-regulated shoot regeneration by inducing differentiation of photosynthesis-rich chlorenchyma cells showed higher expression of auxin biosynthesis genes (Song et al. 2023). This work led to the hypothesis that endogenous photosynthesis, in addition to light signaling, may be capable of inducing auxin biosynthesis. While it is not clear how similar graft healing and shoot regeneration may be, the intriguing possibility remains that photosynthesis and endogenous sugar content could also regulate graft success.

Sugars as potential signaling molecules for graft success

Outside of the role of light-induced regulation, sugars are also thought to be signals during graft healing. During Arabidopsis grafting, the scion accumulates starch and activates a sugar-response genetic program, whereas the stock shows a starvation response (Fig. 7A) (Melnyk et al. 2018). This sugar-regulated gene asymmetry is relieved once phloem transport resumes (Melnyk et al. 2018). The importance of differential sucrose content between the scion and stock for Arabidopsis remains unclear as some data suggest low sucrose in the stock improves graft success (Marsch-Martínez et al. 2013), while other work suggests otherwise (Melnyk et al. 2018). This area is further complicated by the fact that Arabidopsis heals in lit conditions, meaning the scion has a constant source of sugars accumulating above the graft site.

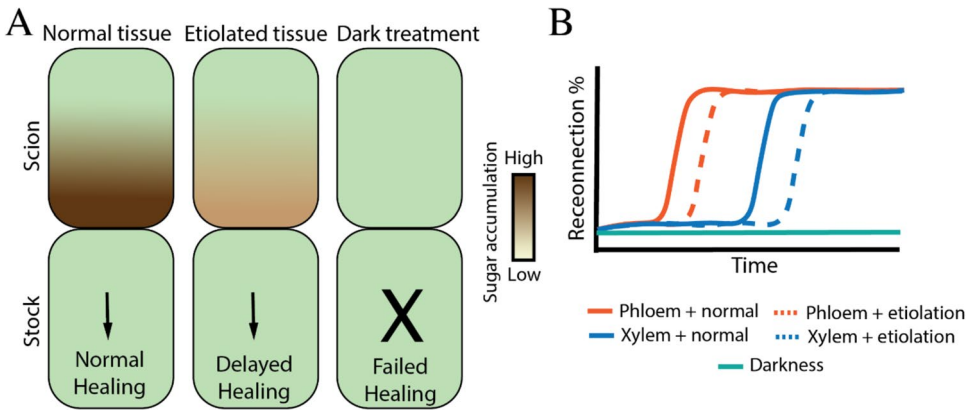

Fig. 7 Sugars are necessary in the scion for graft healing. **A** In typical grafted plants, the photosynthesizing scion will accumulate sugar and starch above the graft junction. When etiolated plants are used for grafting, reduced sugars are available. Grafted plants left for extended periods in the dark always fail to recover, likely due to a complete lack of sugars, amongst other issues. **B** When normal tissue is grafted, the phloem heals first, followed by the xylem. Etiolated scions show developmental delays in healing that cannot be complemented by exogenous glucose application. Dark-treated grafts fail to heal (Modified from (Miao et al. 2021))

Perhaps more relevant studies for horticultural crops include plants that require a period of darkness following grafting. For example, in healing cucumber-pumpkin (*Cucurbita moschata)* grafts, which require a 3-day low light recovery, sucrose, raffinose, stachyose, and starch were higher in the scion than stock prior to phloem connectivity (Miao et al. 2021). When etiolated seedlings containing lower sugar levels were used to graft, grafting success, biomass, and vascular healing speed were reduced (Fig. 7) (Miao et al. 2021). When exogenous glucose was applied to the scion, biomass and graft success could be rescued. However, exogenous glucose could not overcome the developmental delays induced by scions containing low sugar prior to grafting (Fig. 7B). Furthermore, tomato, which requires a low-light period, showed asymmetric genes expressed related to starvation under the graft junction prior to vascular healing (Cui et al. 2021). More work is required to fully understand how asymmetric sugar signals may regulate other genes during grafting or if scion sugar accumulation is simply a byproduct of the physiology of grafting. Sugar metabolism can also be altered by grafting, such as a reduction in roots (Li et al. 2016) or an increase in the fruit (Sun et al. 2023), but the exact mechanisms that induce sugar alterations following grafting have yet to be determined. It is of note that grafted plants left in continuous darkness consistently fail to heal, likely due to a complete lack of photosynthates.

Despite considerable progress focused on the role of phloem connectivity during grafting, the role of sugars and asymmetric sugar-related gene expression remains to be determined. Sugars are necessary for complete graft healing, but the exact ratio of scion to stock sugar and the role that exogenous sugars play in graft success remains to be seen. This area is a clear focus for future research in the field of graft biology, especially in crops that require a dark period for successful healing, as this has a significant effect on the available photosynthates in the scion.

Future perspectives

The future of graft biology is strong, with much still unknown about the regulation and mechanism of graft healing. Light and temperature regulation, sugar metabolism, compatibility, and mobile molecules all integrate at auxin signaling. Currently, studies have profiled the effects of light and temperature on graft healing in horticultural environments, but much remains unknown about the molecular mechanisms that underlie these responses. Tomato provides a unique model system for studying graft biology due to its use as a commercially grafted crop and its emergence as a model system.

Recent work has highlighted the integral role of auxin during grafting. While decades of research have been spent elucidating the role of light and temperature on auxin biosynthesis, there remains a significant disconnect between these fields and graft biology. Future work unraveling how light, temperature, and hormonal regulation underpin grafting is needed to translate our understanding of abiotic stress responses into the context of plant wounding and recovery. Additionally, most of the work focused on grafting and the molecular signaling of temperature regulation has been conducted in Arabidopsis. This complicates our ability to translate this knowledge into horticulturally relevant grafted crops. The next steps in graft biology will likely revolve around identifying conserved mechanisms between Arabidopsis and other crops and validating well-studied signaling pathways from Arabidopsis in species like tomato and

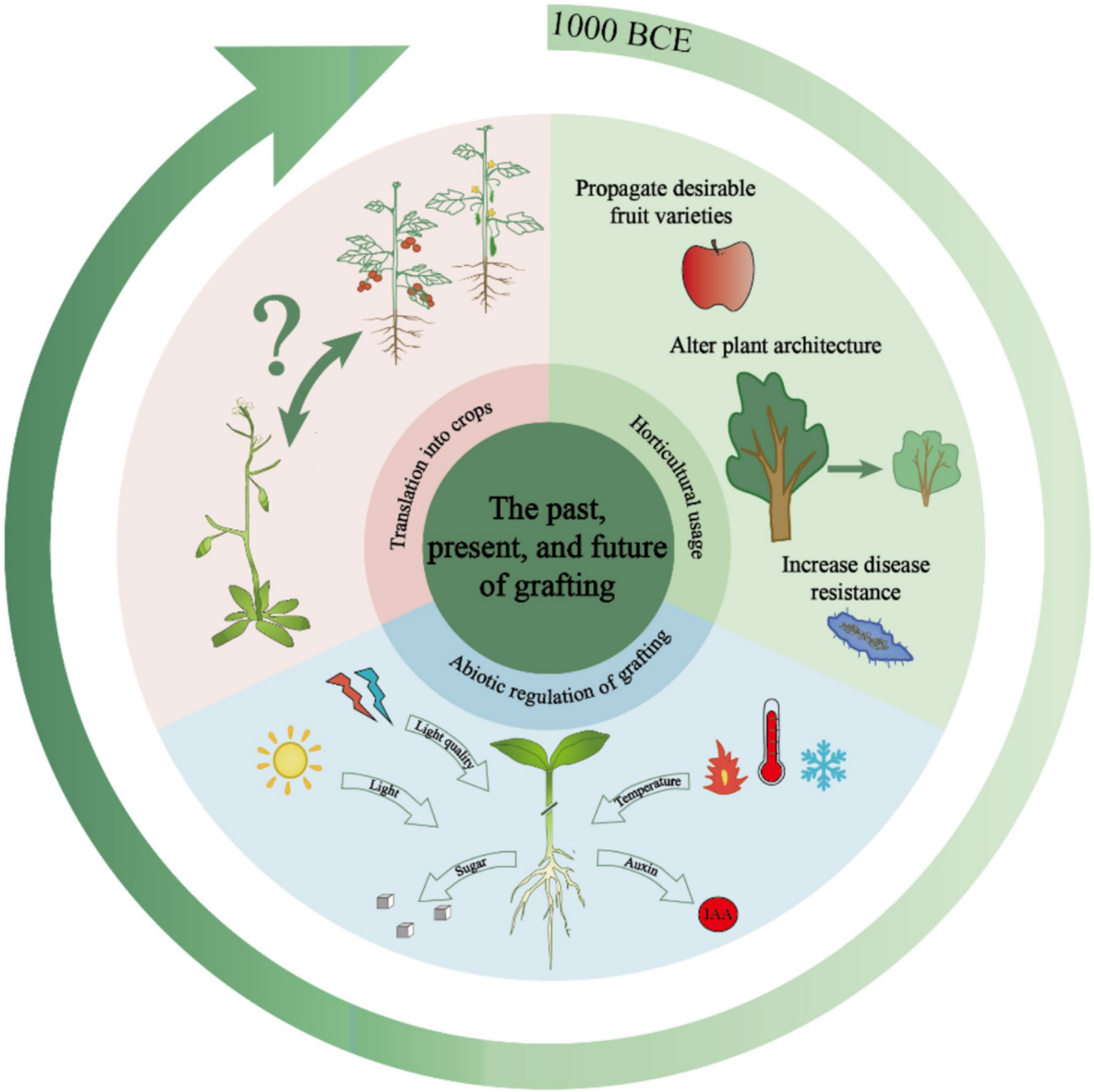

Fig. 8 The past, present, and future of graft science. Grafting was originally used to propagate woody food crops. Over the years, humans have adopted the technique to confer improved traits, graft herbaceous plants, and study mobile molecules (green wedge). Currently, abiotic factors such as light and temperature have highlighted the central role of auxin in graft success (blue wedge). Future efforts to translate knowledge regarding light, temperature, sugar signaling, and grafting from the model system Arabidopsis into horticultural crops are the next important step in graft biology (red wedge). Much like its humble beginnings, the future of graft biology will return to food production (green arrow)

cucumber. Arabidopsis is a powerful model system, but the challenging next step of translating this work into relevant crops is now needed to affect modern agriculture (Fig. 8). Based on profound progress in graft biology over the last few decades, the breadth of graft innovation appears limitless. In a world with an increasing population, destabilized weather patterns, and challenges maintaining stable food supplies, any horticultural techniques that benefit crop production deserve ample attention.

Acknowledgements We would like to thank Huijia Kang for their suggestions and insights.

Author contributions RDH led the literature review, and all authors (RDH, RL, YZ, and HRT) equally contributed to the writing of this manuscript.

Funding This work is funded by the National Natural Science Foundation of China (32330094) and (U21 A20233). HRT is funded by the ZJU Hundred Talents Start-up Fund.

Declarations

Conflict of interest There are no competing interests.

Consent to publish All authors have consented to publication.

Open Access This article is licensed under a Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International License, which permits any non-commercial use, sharing, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if you modified the licensed material. You do not have permission under this licence to share adapted material derived from this article or parts of it. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit http://creativecommons.org/licenses/by-nc-nd/4.0/.

References

Alba R, Kelmenson PM, Cordonnier-Pratt MM, Pratt LH (2000) The phytochrome gene family in tomato and the rapid differential evolution of this family in angiosperms. Mol Biol Evol 17:362–373

Al-Sady B, Ni W, Kircher S, Schäfer E, Quail PH (2006) Photoactivated phytochrome induces rapid PIF3 phosphorylation prior to proteasome-mediated degradation. Mol Cell 23:439–446

Asahina M, Iwai H, Kikuchi A, Yamaguchi S, Kamiya Y, Kamada H, Satoh S (2002) Gibberellin produced in the cotyledon is required for cell division during tissue reunion in the cortex of cut cucumber and tomato hypocotyls. Plant Physiol 129:201–210

Asahina M, Yamauchi Y, Hanada A, Kamiya Y, Kamada H, Satoh S, Yamaguchi S (2007) Effects of the removal of cotyledons on endogenous gibberellin levels in hypocotyls of young cucumber and tomato seedlings. Plant Biotechnol 24:99–106

Asahina M, Azuma K, Pitaksaringkarn W, Yamazaki T, Mitsuda N, Ohme-Takagi M, Yamaguchi S, Kamiya Y, Okada K, Nishimura T et al (2011) Spatially selective hormonal control of RAP2.6L and ANAC071 transcription factors involved in tissue reunion in Arabidopsis. Proc Natl Acad Sci USA 108:16128–16132

Assunção M, Santos C, Brazão J, Eiras-Dias JE, Fevereiro P (2019) Understanding the molecular mechanisms underlying graft success in grapevine. BMC Plant Biol 19:1–17

Bantis F, Koukounaras A, Siomos AS, Fotelli MN, Kintzonidis D (2020) Bichromatic red and blue LEDs during healing enhance the vegetative growth and quality of grafted watermelon seedlings. Sci Hortic 261:109000

Bantis F, Panteris E, Dangitsis C, Carrera E, Koukounaras A (2021) Blue light promotes vascular reconnection, while red light boosts the physiological response and quality of grafted watermelon seedlings. Sci Rep 2021(11):1–14

Bartusch K, Trenner J, Melnyk CW, Quint M (2020) Cut and paste: temperature-enhanced cotyledon micrografting for Arabidopsis thaliana seedlings. Plant Methods 16:1–11

Bellstaedt J, Trenner J, Lippmann R, Poeschl Y, Zhang X, Friml J, Quint M, Delkera C (2019) A mobile auxin signal connects temperature sensing in cotyledons with growth responses in hypocotyls. Plant Physiol 180:757–766

Bhalerao RP, Eklöf J, Ljung K, Marchant A, Bennett M, Sandberg G (2002) Shoot-derived auxin is essential for early lateral root emergence in Arabidopsis seedlings. Plant J 29:325–332

Bisht A, Eekhout T, Canher B, Lu R, Vercauteren I, De Jaeger G, Heyman J, De Veylder L (2023) PAT1-type GRAS-domain proteins control regeneration by activating DOF3.4 to drive cell proliferation in Arabidopsis roots. Plant Cell 35:1513–1531

Bognár LK, Hall A, Ádám É, Thain SC, Nagy F, Millar AJ (1999) The circadian clock controls the expression pattern of the circadian input photoreceptor, phytochrome B. Proc Natl Acad Sci USA 96:14652–14657

Bradley S, Garner RJ (2017) The Grafter's Handbook: Revised & Updated Edition. London: Hachette

Briggs WR, Christie JM (2002) Phototropins 1 and 2: versatile plant blue-light receptors. Trends Plant Sci 7:204–210

Calderwood A, Kopriva S, Morris RJ (2016) Transcript abundance explains mrna mobility data in Arabidopsis thaliana. Plant Cell 28:610–615

Cambridge AP, Morris DA (1996) Transfer of exogenous auxin from the phloem to the polar auxin transport pathway in pea (Pisum sativum L.). Planta 199:583–588

Canher B, Heyman J, Savina M, Devendran A, Eekhout T, Vercauteren I, Prinsen E, Matosevich R, Xu J, Mironova V et al (2020) Rocks in the auxin stream: wound-induced auxin accumulation and ERF115 expression synergistically drive stem cell regeneration. Proc Natl Acad Sci USA 117:16667–16677

Cerruti E, Gisbert C, Drost HG, Valentino D, Portis E, Barchi L, Prohens J, Lanteri S, Comino C, Catoni M (2021) Grafting vigour is associated with DNA de-methylation in eggplant. Hortic Res 2021(8):1–10

Chae K, Isaacs CG, Reeves PH, Maloney GS, Muday GK, Nagpal P, Reed JW (2012) Arabidopsis SMALL AUXIN UP RNA63 promotes hypocotyl and stamen filament elongation. Plant J 71:684–697

Chen S, Lory N, Stauber J, Hoecker U (2015) Photoreceptor specificity in the light-induced and COP1-mediated rapid degradation of the repressor of photomorphogenesis SPA2 in Arabidopsis. PLoS Genet 11:e1005516

Chen Z, Zhao J, Hu F, Qin Y, Wang X, Hu G (2017) Transcriptome changes between compatible and incompatible graft combination of Litchi chinensis by digital gene expression profile. Sci Rep 7:1–12

Chen L, Huang XX, Zhao SM, Xiao DW, Xiao LT, Tong JH, Wang WS, Li YJ, Ding Z, Hou BK (2020) IPyA glucosylation mediates light and temperature signaling to regulate auxin-dependent hypocotyl elongation in Arabidopsis. Proc Natl Acad Sci USA 117:6910–6917

Choi WG, Hilleary R, Swanson SJ, Kim SH, Gilroy S (2016) Rapid, long-distance electrical and calcium signaling in plants. Annu Rev Plant Biol 67:287–307

Clack T, Mathews S, Sharrock RA (1994) The phytochrome apoprotein family in Arabidopsis is encoded by five genes: the sequences and expression of PHYD and PHYE. Plant Mol Biol 25:413–427

Cui Q, Xie L, Dong C, Gao L, Shang Q (2021) Stage-specific events in tomato graft formation and the regulatory effects of auxin and cytokinin. Plant Sci 304:110803

Delker C, Sonntag L, James GV, Janitza P, Ibañez C, Ziermann H, Peterson T, Denk K, Mull S, Ziegler J et al (2014) The DET1-COP1-HY5 pathway constitutes a multipurpose signaling module

regulating plant photomorphogenesis and thermomorphogenesis. Cell Rep 9:1983–1989
Della Rovere F, Fattorini L, D'Angeli S, Veloccia A, Falasca G, Altamura MM (2013) Auxin and cytokinin control formation of the quiescent centre in the adventitious root apex of Arabidopsis. Ann Bot 112:1395–1407
Dharmasiri N, Dharmasiri S, Estelle M (2005) The F-box protein TIR1 is an auxin receptor. Nat 435:441–445
Ding Z, Galván-Ampudia CS, Demarsy E, Łangowski Ł, Kleine-Vehn J, Fan Y, Morita MT, Tasaka M, Fankhauser C, Offringa R et al (2011) Light-mediated polarization of the PIN3 auxin transporter for the phototropic response in Arabidopsis. Nature Cell Biol 13:447–452
Donadio LC, Lederman IE, Roberto SR, Stucchi ES (2019) Dwarfing-canopy and rootstock cultivars for fruit trees. Revista Brasileira de Fruticultura 41:e-997
Duan Y, Zhang F, Meng X, Shang Q (2024) Spatio-temporal dynamics of phytohormones in the tomato graft healing process. Hortic Plant J 10:1362–1370
Durbak A, Yao H, McSteen P (2012) Hormone signaling in plant development. Curr Opin Plant Biol 15:92–96
Efferth T (2019) Biotechnology applications of plant callus cultures. Engineering 5:50–59
Fattorini L, Veloccia A, Della Rovere F, D'angeli S, Falasca G, Altamura MM (2017) Indole-3-Butyric acid promotes adventitious rooting in Arabidopsis thaliana thin cell layers by conversion into indole-3-acetic acid and stimulation of anthranilate synthase activity. BMC Plant Biol 17:1–14
Feng M, Zhang A, Nguyen V, Bisht A, Almqvist C, De Veylder L, Carlsbecker A, Melnyk CW (2024) A conserved graft formation process in Norway spruce and Arabidopsis identifies the PAT gene family as central regulators of wound healing. Nature Plants 10:53–65
Fiorucci AS, Galvão VC, Ince YÇ, Boccaccini A, Goyal A, Allenbach Petrolati L, Trevisan M, Fankhauser C (2020) PHYTOCHROME INTERACTING FACTOR 7 is important for early responses to elevated temperature in Arabidopsis seedlings. New Phytol 226:50–58
Fletcher RA, Zalik S (1964) Effect of light quality on growth and free indoleacetic acid content in phaseolus vulgaris. Plant Physiol 39:328–331
Folta KM, Spalding EP (2001) Unexpected roles for cryptochrome 2 and phototropin revealed by high-resolution analysis of blue light-mediated hypocotyl growth inhibition. Plant J 26:471–478
Foo E, Heynen EMH, Reid JB (2016) Common and divergent shoot–root signaling in legume symbioses. New Phytol 210:643–656
Fortanier E (1954) Some observations on the influence of spectral regions of light on stem elongation, flower bud elongation, flower bud opening and leaf movement in. N.P.
Franklin KA, Lee SH, Patel D, Kumar SV, Spartz AK, Gu C, Ye S, Yu P, Breen G, Cohen JD et al (2011) Phytochrome-Interacting Factor 4 (PIF4) regulates auxin biosynthesis at high temperature. Proc Natl Acad Sci USA 108:20231–20235
Friml J, Wiśniewska J, Benková E, Mendgen K, Palme K (2002) Lateral relocation of auxin efflux regulator PIN3 mediates tropism in Arabidopsis. Nature 415:806–809
Friml J, Vieten A, Sauer M, Weijers D, Schwarz H, Hamann T, Offringa R, Jürgens G (2003) Efflux-dependent auxin gradients establish the apical–basal axis of Arabidopsis. Nature 426:147–153
Gälweiler L, Guan C, Müller A, Wisman E, Mendgen K, Yephremov A, Palme K (1998) Regulation of polar auxin transport by AtPIN1 in Arabidopsis vascular tissue. Science 282:2226–2230
Gangappa SN, Kumar SV (2017) DET1 and HY5 control PIF4-mediated thermosensory elongation growth through distinct mechanisms. Cell Rep 18:344–351
Gao C, Liu X, De Storme N, Jensen KH, Xu Q, Yang J, Liu X, Chen S, Martens HJ, Schulz A et al (2020) Directionality of plasmodesmata-mediated transport in Arabidopsis leaves supports auxin channeling. Curr Biol 30:1970-1977.e4
Geisler M, Blakeslee JJ, Bouchard R, Lee OR, Vincenzetti V, Bandyopadhyay A, Titapiwatanakun B, Peer WA, Bailly A, Richards EL et al (2005) Cellular efflux of auxin catalyzed by the Arabidopsis MDR/PGP transporter AtPGP1. Plant J 44:179–194
Gianinazzi-Pearson V, Gianinazzi S (1992) Influence of intergeneric grafts between host and non-host legumes on formation of vesicular–arbuscular mycorrhiza. New Phytol 120:505–508
Guan L, Li Y, Huang K, Cheng ZMM (2020) Auxin regulation and MDPIN expression during adventitious root initiation in apple cuttings. Hortic Res, 7:143
Haroldsen VM, Szczerba MW, Aktas H, Lopez-Baltazar J, Odias MJ, Chi-Ham CL, Labavitch JM, Bennett AB, Powell ALT (2012) Mobility of transgenic nucleic acids and proteins within grafted rootstocks for agricultural improvement. Front Plant Sci 3:21458
He W, Wang Y, Chen Q, Sun B, Tang HR, Pan DM, Wang XR (2018) Dissection of the mechanism for compatible and incompatible graft combinations of Citrus grandis (L.) Osbeck ('Hongmian Miyou'). Int J Mole Sci 19:505
Heeney M, Frank MH (2023) The mRNA mobileome: challenges and opportunities for deciphering signals from the noise. Plant Cell 35:1817–1833
Hertle AP, Haberl B, Bock R (2021) Horizontal genome transfer by cell-to-cell travel of whole organelles. Sci Adv. https://doi.org/10.1126/sciadv.abd8215
Heyman J, Cools T, Vandenbussche F, Heyndrickx KS, Van Leene J, Vercauteren I, Vanderauwera S, Vandepoele K, De Jaeger G, Van Der Straeten D et al (2013) ERF115 controls root quiescent center cell division and stem cell replenishment. Science 342:860–863
Heyman J, Cools T, Canher B, Shavialenka S, Traas J, Vercauteren I, Van Den Daele H, Persiau G, De Jaeger G, Sugimoto K et al (2016) The heterodimeric transcription factor complex ERF115–PAT1 grants regeneration competence. Nature Plants 2:1–7
Hornitschek P, Kohnen MV, Lorrain S, Rougemont J, Ljung K, López-Vidriero I, Franco-Zorrilla JM, Solano R, Trevisan M, Pradervand S et al (2012) Phytochrome interacting factors 4 and 5 control seedling growth in changing light conditions by directly controlling auxin signaling. Plant J 71:699–711
Ikeuchi M, Sugimoto K, Iwase A (2013) Plant callus: mechanisms of induction and repression. Plant Cell 25:3159–3173
Ikeuchi M, Iwase A, Rymen B, Lambolez A, Kojima M, Takebayashi Y, Heyman J, Watanabe S, Seo M, de Veylder L et al (2017) Wounding triggers callus formation via dynamic hormonal and transcriptional changes. Plant Physiol 175:1158–1174
Ikeuchi M, Iwase A, Ito T, Tanaka H, Favero DS, Kawamura A, Sakamoto S, Wakazaki M, Tameshige T, Fujii H et al (2022) Wound-inducible WUSCHEL-RELATED HOMEOBOX 13 is required for callus growth and organ reconnection. Plant Physiol 188:425–441
Iwase A, Mitsuda N, Koyama T, Hiratsu K, Kojima M, Arai T, Inoue Y, Seki M, Sakakibara H, Sugimoto K et al (2011a) The AP2/ERF transcription factor WIND1 controls cell dedifferentiation in Arabidopsis. Current Bioltogy 21:508–514
Iwase A, Ohme-Takagi M, Sugimoto K (2011b) WIND1: A key molecular switch for plant cell dedifferentiation. Plant Signal Behav 6:1943–1945
Iwase A, Kondo Y, Laohavisit A, Takebayashi A, Ikeuchi M, Matsuoka K, Asahina M, Mitsuda N, Shirasu K, Fukuda H et al (2021) WIND transcription factors orchestrate wound-induced callus formation, vascular reconnection and defense response in Arabidopsis. New Phytol 232:734–752

Izzo LG, Hay Mele B, Vitale L, Vitale E, Arena C (2020) The role of monochromatic red and blue light in tomato early photomorphogenesis and photosynthetic traits. Environ Exp Bot 179:104195
Jang Y, Mun B, Seo T, Lee J, Oh S, Chun C (2013) Effects of light quality and intensity on the carbon dioxide exchange rate, growth, and morphogenesis of grafted pepper transplants during healing and acclimatization. Hortic Sci Technol 31:14–23
Jefree CE, Yeoman MM (1983) Development of intercellular connections between opposing cells in a graft union. New Phytol 93:491–509
Jia Y, Kong X, Hu K, Cao M, Liu J, Ma C, Guo S, Yuan X, Zhao S, Robert HS et al (2020) PIFs coordinate shade avoidance by inhibiting auxin repressor ARF18 and metabolic regulator QQS. New Phytol 228:609–621
Jung JH, Domijan M, Klose C, Biswas S, Ezer D, Gao M, Khattak AK, Box MS, Charoensawan V, Cortijo S et al (2016) Phytochromes function as thermosensors in Arabidopsis. Science 354:886–889
Kehr J, Kragler F (2018) Long distance RNA movement. New Phytol 218:29–40
Keuskamp DH, Pollmann S, Voesenek LACJ, Peeters AJM, Pierik R (2010) Auxin transport through PIN-FORMED 3 (PIN3) controls shade avoidance and fitness during competition. Proc Natl Acad Sci USA 107:22740–22744
King SR, Davis AR, Liu W, Levi A (2008) Grafting for disease resistance. HortScience 43:1673–1676
Knauss S, Rohrmeier T, Lehle L (2003) The auxin-induced maize gene ZmSAUR2 encodes a short-lived nuclear protein expressed in elongating tissues. J Biol Chem 278:23936–23943
Koini MA, Alvey L, Allen T, Tilley CA, Harberd NP, Whitelam GC, Franklin KA (2009) High temperature-mediated adaptations in plant architecture require the BHLH transcription factor PIF4. Curr Biol 19:408–413
Kong CH, Xuan TD, Khanh TD, Tran HD, Trung NT (2019) Allelochemicals and signaling chemicals in plants. Molecules 24:2737
Kundariya H, Yang X, Morton K, Sanchez R, Axtell MJ, Hutton SF, Fromm M, Mackenzie SA (2020) MSH1-induced heritable enhanced growth vigor through grafting is associated with the RdDM pathway in plants. Nat Commun 11:1–14
Kurotani KI, Huang C, Okayasu K, Suzuki T, Ichihashi Y, Shirasu K, Higashiyama T, Niwa M, Notaguchi M (2022) Interfamily grafting capacity of petunia. Hortic Res 9:uhab056
Lee JM, Kubota C, Tsao SJ, Bie Z, Echevarria PH, Morra L, Oda M (2010) Current status of vegetable grafting: diffusion, grafting techniques, automation. Sci Hortic 127:93–105
Lee KM, Lim CS, Muneer S, Jeong BR (2016) Functional vascular connections and light quality effects on tomato grafted unions. Sci Hortic 201:306–317
Lee S, Wang W, Huq E (2021) Spatial regulation of thermomorphogenesis by HY5 and PIF4 in Arabidopsis. Nat Commun 12:1–12
Legris M, Klose C, Burgie ES, Rojas CC, Neme M, Hiltbrunner A, Wigge PA, Schäfer E, Vierstra RD, Casal JJ (2016) Phytochrome B integrates light and temperature signals in Arabidopsis. Science 354:897–900
Lehmann F, Hardtke CS (2016) Secondary growth of the Arabidopsis hypocotyl—vascular development in dimensions. Curr Opin Plant Biol 29:9–15
Leivar P, Monte E (2014) PIFs: systems integrators in plant development. Plant Cell 26:56–78
Li L, Ljung K, Breton G, Schmitz RJ, Pruneda-Paz J, Cowing-Zitron C, Cole BJ, Ivans LJ, Pedmale UV, Jung HS et al (2012) Linking photoreceptor excitation to changes in plant architecture. Genes Dev 26:785–790
Li G, Ma J, Tan M, Mao J, An N, Sha G, Zhang D, Zhao C, Han M (2016) Transcriptome analysis reveals the effects of sugar metabolism and auxin and cytokinin signaling pathways on root growth and development of grafted apple. BMC Genomics 17:1–17
Li W, Fang C, Krishnan S, Chen J, Yu H, Murphy AS, Merewitz E, Katin-Grazzini L, McAvoy RJ, Deng Z et al (2017) Elevated auxin and reduced cytokinin contents in rootstocks improve their performance and grafting success. Plant Biotechnol J 15:1556–1565
Ljung K, Bhalerao RP, Sandberg G (2001) Sites and homeostatic control of auxin biosynthesis in Arabidopsis during vegetative growth. Plant J 28:465–474
Lorrain S, Allen T, Duek PD, Whitelam GC, Fankhauser C (2008) Phytochrome-mediated inhibition of shade avoidance involves degradation of growth-promoting bHLH transcription factors. Plant J 53:312–323
Lu XD, Zhou CM, Xu PB, Luo Q, Lian HL, Yang HQ (2015) Red-light-dependent interaction of phyB with SPA1 promotes COP1-SPA1 dissociation and photomorphogenic development in Arabidopsis. Mol Plant 8:467–478
Ma D, Li X, Guo Y, Chu J, Fang S, Yan C, Noel JP, Liu H (2016) Cryptochrome 1 interacts with PIF4 to regulate high temperature-mediated hypocotyl elongation in response to blue light. Proc Natl Acad Sci USA 113:224–229
Marsch-Martínez N, Franken J, Gonzalez-Aguilera KL, de Folter S, Angenent G, Alvarez-Buylla ER (2013) An efficient flat-surface collar-free grafting method for Arabidopsis thaliana seedlings. Plant Methods 9:1–9
Mashiguchi K, Tanaka K, Sakai T, Sugawara S, Kawaide H, Natsume M, Hanada A, Yaeno T, Shirasu K, Yao H et al (2011) The main auxin biosynthesis pathway in Arabidopsis. Proc Natl Acad Sci USA 108:18512–18517
Mason MG, Mathews DE, Argyros DA, Maxwell BB, Kieber JJ, Alonso JM, Ecker JR, Schaller GE (2005) Multiple Type-B response regulators mediate cytokinin signal transduction in Arabidopsis. Plant Cell 17:3007–3018
Matsuoka K, Sugawara E, Aoki R, Takuma K, Terao-Morita M, Satoh S, Asahina M (2016) Differential cellular control by cotyledon-derived phytohormones involved in graft reunion of Arabidopsis hypocotyls. Plant Cell Physiol 57:2620–2631
Matsuoka K, Yanagi R, Yumoto E, Yokota T, Yamane H, Satoh S, Asahina M (2018) RAP2. 6L and jasmonic acid–responsive genes are expressed upon Arabidopsis hypocotyl grafting but are not needed for cell proliferation related to healing. Plant Mol Biol 96:531–542
Matsuoka K, Sato R, Matsukura Y, Kawajiri Y, Iino H, Nozawa N, Shibata K, Kondo Y, Satoh S, Asahina M (2021) Wound-inducible ANAC071 and ANAC096 transcription factors promote cambial cell formation in incised Arabidopsis flowering stems. Commun Biol. https://doi.org/10.1038/s42003-021-01895-8
Mattsson J, Sung ZR, Berleth T (1999) Responses of plant vascular systems to auxin transport inhibition. Development 126:2979–2991
Mazur E, Benková E, Friml J (2016) Vascular cambium regeneration and vessel formation in wounded inflorescence stems of Arabidopsis. Sci Rep 6:1–15
Mei J, Tang X, Gu Y, Lu H, Yang Y, Shen Q, Yang L, Li B, Zuo J, Singh VP et al (2024) Role of TIR1/AFB family genes during grafting in Carya cathayensis. Front Plant Sci 15:1494579
Meijer G (1959) The spectral dependence of flowering and elongation 1). Acta Botanica Neerlandica 8:189–246
Melnyk CW (2017) Grafting with Arabidopsis thaliana. Methods Mol Biol 1497:9–18
Melnyk CW, Schuster C, Leyser O, Meyerowitz EM (2015) A developmental framework for graft formation and vascular reconnection in Arabidopsis thaliana. Curr Biol 25:1306–1318

Melnyk CW, Meyerowitz EM, Gabel A, Robinson S, Hardcastle TJ, Grosse I, Miyashima S (2018) Transcriptome dynamics at Arabidopsis graft junctions reveal an intertissue recognition mechanism that activates vascular regeneration. Proc Natl Acad Sci 115:E2447–E2456

Meng Y, Li H, Wang Q, Liu B, Lin C (2013) Blue light–dependent interaction between Cryptochrome2 and CIB1 regulates transcription and leaf senescence in soybean[W]. Plant Cell 25:4405–4420

Meng L, Song W, Liu S, Dong J, Zhang Y, Wang C, Xu Y, Wang S (2015) Light quality regulates lateral root development in tobacco seedlings by shifting auxin distributions. J Plant Growth Regul 34:574–583

Meng X, Zhang F, Yang W, Shang Q (2025) A PHYB-PIF4-auxin module promotes tomato graft formation in response to elevated ambient temperature. Hortic Plant J 11(1):227–238

Miao L, Li Q, Shu Sun T, Chai S, Wang C, Bai L, Sun M, Li Y, Qin X, Zhang Z et al (2021) Sugars promote graft union development in the heterograft of cucumber onto pumpkin. Hortic Res. https://doi.org/10.1038/s41438-021-00580-5

Mo Z, Zhang Y, Hu L, Zhai M, Xuan J (2023) Genome-wide identification and expression analysis of auxin response factor (ARF) gene family in pecan indicates its possible roles during graft union formation. Sci Hortic 322:112401

Molnar A, Melnyk CW, Bassett A, Hardcastle TJ, Dunn R, Baulcombe DC (2010) Small silencing RNAs in plants are mobile and direct epigenetic modification in recipient cells. Science 328:872–875

Moore R (1984) A model for graft compatibility-incompatibility in higher plants. Am J Bot 71:752

Moore R, Walker DB (1981) Studies of vegetative compatibility-incompatibility in higher plants. I. A structural study of a compatible autograft in Sedum telephoides (Crassulaceae). Am J Bot 68:820

Moosavi-Nezhad M, Salehi R, Aliniaeifard S, Tsaniklidis G, Woltering EJ, Fanourakis D, Żuk-Gołaszewska K, Kalaji HM (2021) Blue light improves photosynthetic performance during healing and acclimatization of grafted watermelon seedlings. Int J Mol Sci 22(15):8043

Morris DA (1979) The effect of temperature on the velocity of exogenous auxin transport in intact chilling-sensitive and chilling-resistant plants. Planta 146:603–605

Mudge K, Janick J, Scofield S, Goldschmidt EE (2009) A history of grafting. Hortic Rev 35:437–493

Müller LM, Harrison MJ (2019) Phytohormones, miRNAs, and peptide signals integrate plant phosphorus status with arbuscular mycorrhizal symbiosis. Curr Opin Plant Biol 50:132–139

Muneer S, Ko CH, Wei H, Chen Y, Jeong BR (2016) Physiological and proteomic investigations to study the response of tomato graft unions under temperature stress. PLoS One 11:e0157439

Navarro L, Dunoyer P, Jay F, Arnold B, Dharmasiri N, Estelle M, Voinnet O, Jones JDG (2006) A plant miRNA contributes to antibacterial resistance by repressing auxin signaling. Science 312:436–439

Notaguchi M, Kurotani KI, Sato Y, Tabata R, Kawakatsu Y, Okayasu K, Sawai Y, Okada R, Asahina M, Ichihashi Y et al (2020) Cell-cell adhesion in plant grafting is facilitated by b-1,4-glucanases. Science 369:698–702

Nozue K, Harmer SL, Maloof JN (2011) Genomic analysis of circadian clock-, light-, and growth-correlated genes reveals PHYTOCHROME-INTERACTING FACTOR5 as a modulator of auxin signaling in Arabidopsis. Plant Physiol 156:357–372

Ongaro V, Bainbridge K, Williamson L, Leyser O (2008) Interactions between axillary branches of Arabidopsis. Mol Plant 1:388–400

Paradiso R, Proietti S (2022) Light-quality manipulation to control plant growth and photomorphogenesis in greenhouse horticulture: the state of the art and the opportunities of modern led systems. J Plant Growth Regul 41:742–780

Parent JS, de Alba AEM, Vaucheret H (2012) The origin and effect of small RNA signaling in plants. Front Plant Sci 3:30926

Park E, Park J, Kim J, Nagatani A, Lagarias JC, Choi G (2012) Phytochrome B inhibits binding of phytochrome-interacting factors to their target promoters. Plant J 72:537–546

Pedmale UV, Carol Huang S, Zander M, Cole BJ, Hetzel J, Ljung K, Reis BPA, Sridevi P, Nito K, Nery JR et al (2014) A role for barley CRYPTOCHROME1 in light regulation of grain dormancy and germination. The Plant Cell 26:1094

Pedmale UV, Huang SSC, Zander M, Cole BJ, Hetzel J, Ljung K, Reis PAB, Sridevi P, Nito K, Nery JR et al (2015) Cryptochromes interact directly with PIFs to control plant growth in limiting blue light. Cell 164:233

Pitaksaringkarn W, Matsuoka K, Asahina M, Miura K, Sage-Ono K, Ono M, Yokoyama R, Nishitani K, Ishii T, Iwai H et al (2014) XTH20 and XTH19 regulated by ANAC071 under auxin flow are involved in cell proliferation in incised Arabidopsis inflorescence stems. Plant J 80:604–614

Podolec R, Ulm R (2018) Photoreceptor-mediated regulation of the COP1/SPA E3 ubiquitin ligase. Curr Opin Plant Biol 45:18–25

Procko C, Crenshaw CM, Ljung K, Noel JP, Chory J (2014) Cotyledon-generated auxin is required for shade-induced hypocotyl growth in Brassica rapa. Plant Physiol 165:1285–1301

Prusinkiewicz P, Crawford S, Smith RS, Ljung K, Bennett T, Ongaro V, Leyser O (2009) Control of bud activation by an auxin transport switch. Proc Natl Acad Sci USA 106:17431–17436

Pucciariello O, Legris M, Rojas CC, Iglesias MJ, Hernando CE, Dezar C, Vazquez M, Yanovsky MJ, Finlayson SA, Prat S et al (2018) Rewiring of auxin signaling under persistent shade. Proc Natl Acad Sci USA 115:5612–5617

Qiu L, Jiang B, Fang J, Shen Y, Fang Z, Saravana Kumar RM, Yi K, Shen C, Yan D, Zheng B (2016) Analysis of transcriptome in hickory (Carya cathayensis), and uncover the dynamics in the hormonal signaling pathway during graft process. BMC Genomics 17:1–13

Radwanski ER, Last RL (1995) Tryptophan biosynthesis and metabolism: biochemical and molecular genetics. Plant Cell 7:921

Rovere DF, Fattorini L, Ronzan M, Falasca G, Altamura MM (2016) The quiescent center and the stem cell niche in the adventitious roots of Arabidopsis thaliana. Plant signal behav 11(5):e1176660

Roosjen M, Paque S, Weijers D (2018) Auxin Response Factors: output control in auxin biology. J Exp Bot 69:179–188

Sauer M, Balla J, Luschnig C, Wiśniewska J, Reinöhl V, Friml J, Benková E (2006) Canalization of auxin flow by Aux/IAA-ARF-dependent feedback regulation of PIN polarity. Genes Dev 20:2902–2911

Serivichyaswat PT, Bartusch K, Leso M, Musseau C, Iwase A, Chen Y, Sugimoto K, Quint M, Melnyk CW (2022) High temperature perception in leaves promotes vascular regeneration and graft formation in distant tissues. Development 149(5):dev200079

Serivichyaswat PT, Kareem A, Feng M, Melnyk CW (2024) Auxin signaling in the cambium promotes tissue adhesion and vascular formation during Arabidopsis graft healing. Plant Physiol 196:754–762

Shafiq I, Hussain S, Ma R, Iqbal N, Ma A, Raza A, Fang FY, Mumtaz M, Shoaib M, Ansar M et al (2021) Crop photosynthetic response to light quality and light intensity. J Integr Agric 20:4–23

Sheerin DJ, Menon C, Zur Oven-Krockhaus S, Enderle B, Zhu L, Johnen P, Schleifenbaum F, Stierhof Y-D, Huq E, Hiltbrunner A (2015) Light-Activated Phytochrome A and B interact with members of the SPA Family to promote photomorphogenesis in Arabidopsis by reorganizing the COP1/SPA complex. Plant Cell 1:189–201

Shibasaki K, Uemura M, Tsurumi S, Rahman A (2010) Auxin response in Arabidopsis under cold stress: underlying molecular mechanisms. Plant Cell 21:3823–3838

Song X, Guo P, Xia K, Wang M, Liu Y, Chen L, Zhang J, Xu M, Liu N, Yue Z et al (2023) Spatial transcriptomics reveals light-induced chlorenchyma cells involved in promoting shoot regeneration in tomato callus. Proc Natl Acad Sci USA 120:e2310163120

Spartz AK, Lee SH, Wenger JP, Gonzalez N, Itoh H, Inzé D, Peer WA, Murphy AS, Overvoorde PJ, Gray WM (2012) The SAUR19 subfamily of SMALL AUXIN UP RNA genes promote cell expansion. Plant J 70:978–990

Spartz AK, Ren H, Park MY, Grandt KN, Lee SH, Murphy AS, Sussman MR, Overvoorde PJ, Gray WM (2014) SAUR inhibition of PP2C-D phosphatases activates plasma membrane H+-ATPases to promote cell expansion in Arabidopsis. Plant Cell 26:2129–2142

Spiegelman Z, Ham BK, Zhang Z, Toal TW, Brady SM, Zheng Y, Fei Z, Lucas WJ, Wolf S (2015) A tomato phloem-mobile protein regulates the shoot-to-root ratio by mediating the auxin response in distant organs. Plant J 83:853–863

Suer S, Agusti J, Sanchez P, Schwarz M, Greb T (2011) WOX4 imparts auxin responsiveness to cambium cells in Arabidopsis. Plant Cell 23(9):3247–3259

Sun J, Qi L, Li Y, Chu J, Li C (2012) PIF4–Mediated activation of YUCCA8 expression integrates temperature into the auxin pathway in regulating arabidopsis hypocotyl growth. PLoS Genet 8:e1002594

Sun N, Ma Y, Wang X, Ying Q, Huang Y, Zhang L, Zhu Z, Wang Y, He Y (2023) Grafting onto pumpkin alters the evolution of fruit sugar profile and increases fruit weight through invertase and sugar transporters in watermelon. Sci Hortic 314:111936

Tanaka H, Hashimoto N, Kawai S, Yumoto E, Shibata K, Tameshige T, Yamamoto Y, Sugimoto K, Asahina M, Ikeuchi M (2023) Auxin-Induced WUSCHEL-RELATED HOMEOBOX13 mediates asymmetric activity of callus formation upon cutting. Plant Cell Physiol 64:305–316

Tao Y, Ferrer JL, Ljung K, Pojer F, Hong F, Long JA, Li L, Moreno JE, Bowman ME, Ivans LJ et al (2008) Rapid synthesis of auxin via a new tryptophan-dependent pathway is required for shade avoidance in plants. Cell 133:164–176

Thomas HR, Frank MH (2019) Connecting the pieces: uncovering the molecular basis for long-distance communication through plant grafting. New Phytol 223:582–589

Thomas H, Van den Broeck L, Spurney R, Sozzani R, Frank M (2022) Gene regulatory networks for compatible versus incompatible grafts identify a role for SlWOX4 during junction formation. Plant Cell 34:535–556

Thomas HR, Gevorgyan A, Frank MH (2023) Anatomical and biophysical basis for graft incompatibility within the solanaceae. J Exp Bot 74:4461–4470

Thomas HR, Gevorgyan A, Hermanson A, Yanders S, Erndwein L, Norman-Ariztía M, Sparks EE, Frank MH (2024) Graft incompatibility between pepper and tomato elicits an immune response and triggers localized cell death. Hortic Res. https://doi.org/10.1093/hr/uhae255

Torres-Galea P, Hirtreiter B, Bolle C (2013) Two GRAS proteins, SCARECROW-LIKE21 and PHYTOCHROME A SIGNAL TRANSDUCTION1, function cooperatively in phytochrome A signal transduction. Plant Physiol 161:291–304

Vu NT, Kim YS, Kang HM, Kim IS (2014) Influence of short-term irradiation during pre- and post-grafting period on the graft-take ratio and quality of tomato seedlings. Hortic Environ Biotechnol 55:27–35

Wang J, Jin Z, Yin H, Yan B, Ren ZZ, Xu J, Mu CJ, Zhang Y, Wang MQ, Liu H (2014) Auxin redistribution and shifts in PIN gene expression during Arabidopsis grafting. Russ J Plant Physiol 61:688–696

Wang Y, Li W, Xu X, Qiu C, Wu T, Wei Q, Ma F, Han Z (2019) Progress of apple rootstock breeding and its use. Hortic Plant J 5:183–191

Wang X, Yu R, Wang J, Lin Z, Han X, Deng Z, Fan L, He H, Deng XW, Chen H (2020) The asymmetric expression of SAUR genes mediated by ARF7/19 promotes the gravitropism and phototropism of plant hypocotyls. Cell Rep 31(3):107529

Wang X, Xiong M, Xu J, Zhang T, Kadeer A, Bie Z, Notaguchi M, Huang Y (2024) PIN1a- mediated auxin release from rootstock cotyledon contributes to healing in watermelon as revealed by seeds soaking-VIGS and cotyledon grafting. Hortic Res. https://doi.org/10.1093/hr/uhae329

Wendte JM, Pikaard CS (2017) The RNAs of RNA-directed DNA methylation. Biochimica et biophysica acta. Gene Regul Mech 1860:140–148

Wulf KE, Reid JB, Foo E (2019) Auxin transport and stem vascular reconnection—has our thinking become canalized? Ann Bot 123:429–439

Xie L, Dong C, Shang Q (2019) Gene co-expression network analysis reveals pathways associated with graft healing by asymmetric profiling in tomato. BMC Plant Biol 19:1–12

Yamada M, Greenham K, Prigge MJ, Jensen PJ, Estelle M (2009) The TRANSPORT INHIBITOR RESPONSE2 gene is required for auxin synthesis and diverse aspects of plant development. Plant Physiol 151:168–179

Yang Y, Hammes UZ, Taylor CG, Schachtman DP, Nielsen E (2006) High-Affinity auxin transport by the AUX1 influx carrier protein. Curr Biol 16:1123–1127

Yang Y, Mao L, Jittayasothorn Y, Kang Y, Jiao C, Fei Z, Zhong GY (2015) Messenger RNA exchange between scions and rootstocks in grafted grapevines. BMC Plant Biol 15:1–14

Yang L, Perrera V, Saplaoura E, Apelt F, Bahin M, Kramdi A, Olas J, Mueller-Roeber B, Sokolowska E, Zhang W et al (2019) M5C Methylation guides systemic transport of messenger RNA over graft junctions in plants. Curr Biol 29:2465-2476.e5

Yang L, Machin F, Wang S, Saplaoura E, Kragler F (2023) Heritable transgene-free genome editing in plants by grafting of wild-type shoots to transgenic donor rootstocks. Nat Biotechnol 41:958–967

Yin H, Yan B, Sun J, Jia P, Zhang Z, Yan X, Chai J, Ren Z, Zheng G, Liu H (2012) Graft-union development: a delicate process that involves cell–cell communication between scion and stock for local auxin accumulation. J Exp Bot 63:4219–4232

Yousef AF, Ali MM, Rizwan HM, Ahmed MAA, Ali WM, Kalaji HM, Elsheery N, Wróbel J, Xu Y, Chen F (2021a) Effects of light spectrum on morpho-physiological traits of grafted tomato seedlings. PLoS One 16:e0250210

Yousef AF, Ali MM, Rizwan HM, Gad AG, Liang D, Binqi L, Kalaji HM, Wróbel J, Xu Y, Chen F (2021) Light quality and quantity affect graft union formation of tomato plants. Scientific Rep 11:1–11

Yuan H, Zhao L, Qiu L, Xu D, Tong Y, Guo W, Yang X, Shen C, Yan D, Zheng B (2017) Transcriptome and hormonal analysis of grafting process by investigating the homeostasis of a series of metabolic pathways in Torreya grandis cv. Merrillii INdustrial Crops Products 108:814–823

Žádníkova P, Petrášek J, Marhavý P, Raz V, Vandenbussche F, Ding Z, Schwarzerová K, Morita MT, Tasaka M, Hejatko J et al (2010) Role of PIN-mediated auxin efflux in apical hook development of Arabidopsis thaliana. Development 137:607–617

Zeist AR, de Resende JTV, Zanin DS, da Silva ALBR, Perrud AC, Bueno GA, Arantes JHV, de Lima DP (2020) Effect of acclimation environments, grafting methods and rootstock RVTC-66

on the seedling development and production of tomato. Sci Hortic 271:109496
Zhai L, Wang X, Tang D, Qi Q, Yer H, Jiang X, Han Z, McAvoy R, Li W, Li Y (2021) Molecular and physiological characterization of the effects of auxin-enriched rootstock on grafting. Hortic Res 8:1–13
Zhang Y, Yu Q, Jiang N, Yan X, Wang C, Wang Q, Liu J, Zhu M, Bednarek SY, Xu J et al (2017) Clathrin regulates blue light-triggered lateral auxin distribution and hypocotyl phototropism in Arabidopsis. Plant Cell Environ 40:165–176
Zhang A, Matsuoka K, Kareem A, Robert M, Roszak P, Blob B, Bisht A, De Veylder L, Voiniciuc C, Asahina M et al (2022) Cell-wall damage activates DOF transcription factors to promote wound healing and tissue regeneration in Arabidopsis thaliana. Curr Biol 32:1883-1894.e7
Zhang A, Wang T, Yuan L, Shen Y, Liu K, Liu B, Xu K, Elsadek MA, Wang Y, Wu L et al (2024) Horizontal transfer of plasmid-like extrachromosomal circular DNAs across graft junctions in Solanaceae. Mol Hortic. https://doi.org/10.1186/s43897-024-00124-0
Zobel RW (1973) Some physiological characteristics of the ethylene-requiring tomato mutant diageotropica. Plant Physiol 52:385–389

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.