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REVIEW

Growth activity, rooting capacity, and tropism: three auxinic precepts fulfilled by melatonin

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Abstract Plant melatonin appears to be a multiregulatory molecule with multiple functions similar to those observed in animals. It induces growth in stems and stimulates root generation. It is also able to delay senescence by protecting photosynthetic systems and related processes. One of the most studied actions of melatonin is its effect on biotic and abiotic stresses in the plant, such as that produced by drought, extreme temperatures, chemical pollution, UV radiation, etc. Recent data have demonstrated its role as a modulator of gene expression in plants. In this review, we compare studies which show that melatonin behaves in a similar way to auxin, and present data that relate the physiological responses produced by melatonin with the action of auxin, such as promoting/inhibiting growth activity and rooting capacity. In addition, for the first time, the data presented demonstrate the possible involvement of melatonin in the tropic response of roots. The possible role of melatonin as a plant regulator and its relationship with auxin action and the signaling molecule nitric oxide is presented and discussed in a hypothetical model.

Keywords Auxin · Tropism · Melatonin · Phytomelatonin · Plant growth regulator · Plant stress · Rhizogenesis

Introduction

Melatonin (*N*-acetyl-5-methoxytryptamine) is a biogenic indoleamine that is structurally related to other important substances, such as tryptophan, tryptamine, and serotonin, and also with indolic compounds that are very important in plant physiology, such as the common auxin and indole-3-acetic acid (IAA). Many advances in understanding the role of melatonin have been made since its discovery in plants in 1995 (Dubbels et al. 1995; Hattori et al. 1995). The very amplitude of its biological actions in plants has led it to being called a multiregulatory molecule. Such actions include the ability to act as plant biostimulator against stress, both biotic and abiotic; the ability to regulate plant growth; the ability to regulate processes of plant vegetative development such as rooting, leaf senescence, photosynthetic efficiency, and biomass yield, as well as a role as a potential regulator in the processes of flowering, and the formation and ripening of fruits and seeds (Arnao and Hernández-Ruiz 2014, 2015; Nawaz et al. 2016; Reiter et al. 2015).

Undoubtedly, huge steps forward in melatonin research were the studies on the changes in gene expression modulated by melatonin. To date, many genes and transcription factors that are up- or down-regulated by melatonin have been identified, including the genes of some enzymes of the metabolism of plant hormones, of ion transporters, of primary metabolic pathway enzymes, and of transcription factors from abiotic stress situations, among many others (Shi et al. 2016; Weeda et al. 2014; Wei et al. 2016; Zhang et al. 2014a, b, 2015).

One of the most interesting and most controversial aspects in research on melatonin in plants is its postulated similar role to that of IAA, and the auxin activity of melatonin has been described in many cases (Arnao and

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Hernández-Ruiz 2006; Hernández-Ruiz et al. 2004). Besides the chemical similarities/differences that exist between melatonin and IAA (both are indolic compounds, but only IAA is an acid), both compounds have been compared in many specific physiological studies. The last 10–12 years have seen data that accumulate on the possible role of melatonin as auxin, perhaps, better expressed as the possible auxin activity of melatonin (although the way to describe this hormonal action differs according to individual authors). Although the term "auxin" may seem to be ambitious when applied to melatonin, there are many data that suggest that melatonin plays a role closely related with the classic actions of auxin.

Kögl and Haagen-Smit (1933) introduced the term "auxin", from "*auxano*", which means "growth" in Greek (Kögl et al. 1933; Kögl and Haagen-Smit 1933). Later "auxin" was applied to various natural and artificial substances that can promote cell elongation, in a similar way to IAA (Tukey et al. 1954). Any chemical with these qualities could be considered as an auxin. Thus, according to the precepts of the "Old Auxinology", the effects that auxin has in plants include the induction of cell growth through triggering cell wall loosening, the promotion of growth in stems, and the growth inhibition in roots (Lüthen 2015). This contrasted effect in stem and root can be explained by the different degrees of sensitivity of tissues to auxin levels or gradients. Previously, the idea that ''Wuchsstoff'' (growth-inducing substance) gradients were the physical basis of tropisms was first established by Cholodny in 1924 (Cholodny 1924), and is still referred to as the Cholodny–Went theory (Went 1928). Accordingly, differential auxin transport in tissues causes auxin gradients, leading to asymmetric growth, which drives phototropism and gravitropism in plants (Perrin et al. 2005).

In addition, the concentration of auxin in tissues is a key factor in the regeneration of new organs. Root regeneration strongly depends on the IAA levels and also of its transport where the regeneration of roots occurs, whether by adventitious or lateral root promotion (Aloni 2004). In this sense, auxin (natural or synthetic) are used to induce rapid and prolific rooting of cuttings of a wide range of ornamentals, vines, trees, and shrubs (Rademacher 2015).

The three above mentioned responses (the promotion/inhibition of growth, rooting activity and tropisms) clearly define the action of a compound with possible auxinic activity. This is precisely the objective of this work: to compare studies in which melatonin behave in a way similar to auxin. More specifically, we look at three physiological responses that are clearly auxin-mediated: the promotion/inhibition of growth activity, rooting capacity, and tropisms in which melatonin may be involved. As regards tropism, we provide preliminary data, for the first time, concerning the possible involvement of melatonin in the tropic response (tropism) of lupin plants.

Growth activity of melatonin

Table 1 shows the studies in which exogenous melatonin has been seen to change the growth pattern in different species. In all the studies presented, melatonin induced growth in aerial parts and promoted or inhibited growth in roots, depending on the concentration assayed. In general, growth inhibition only occurs at high melatonin concentrations (100 μM), although in one recent study using *Zea mays,* melatonin was seen not to have any effect on growth-promoting/inhibiting activities in the conditions assayed. Nevertheless, in the presence of NaCl, melatonin partially restored NaCl-inhibited growth in maize coleoptiles and roots (Kim et al. 2016), which contrasted with the findings of a previous paper, in which leaves and roots of *Zea mays* were seen to respond as expected to melatonin treatment (Zhao et al. 2015) (see data in Table 1). In general, the growth-promoting effect of melatonin is high when a stress condition affects plant development, as can be seen in the case of *Helianthus* and *Zea mays* grown in saline conditions, and in *Arabidopsis* and *Cynodon* exposed to cold stress, among others (Table 1). Melatonin induced a 3–4-fold increase in growth compared to control plants in the aerial tissues of *Lupinus, Phalaris, Triticum, Hordeum, Arabidopsis,* and *Cucumis*, and a less pronounced increase in others (Table 1).

Rooting activity of melatonin

Table 2 shows some details of the studies in which rooting activity has been demonstrated to be promoted by melatonin. In the pioneering study in lupin by Arnao and Hernández-Ruiz (2007), where the strong induction of roots by melatonin was compared to the effect of IAA, it was seen that melatonin induced the root primordia from pericycle cells and clearly affected the new occurrence of both adventitious and lateral roots. Therefore, melatonin changed the pattern of distribution, the time-course, the number and length of adventitious roots, and the number of new lateral roots. Since this study, many other species have been used to confirm that melatonin promotes the generation of lateral and adventitious roots. For example, in *Arabidopsis thaliana*, melatonin was seen to increase the appearance of adventitious roots twofold and the appearance of lateral roots by up to threefold but to have no effect on root hair density (Pelagio-Flores et al. 2012). In addition, in three transgenic lines of *Arabidopsis thaliana* which over-produce melatonin, a high number of lateral roots (compared with wild type) were induced (Zuo et al. 2014). Similar data are shown in Table 2.

Table 1 Effect of exogenous melatonin on growth in different organ and species

Specie	Organ	Melatonin treatment on growth	Maximum growth response (vs. without melatonin)	References
Lupinus albus	Hypocotyl (sections)	1.0–10 μM	↑ Up to 3.0-times	(Hernández-Ruiz et al. 2004)
	Hypocotyl (derooted)	0.1–10 μM	↑ Up to 3.5-times	
	Cotyledon	0.1–1.0 μM	↑ Up to 1.2-times	(Hernández-Ruiz and Arnao 2008b)
Phalaris canariensis	Coleoptile (sections)	0.1–10 μM	↑ Up to 3.6-times	(Hernández-Ruiz et al. 2005)
	Roots (intact)	0.1 nM–100 μM	↓ Up to 0.4-times	
Triticum aestivum	Coleoptile (sections)	10–100 μM	↑ Up to 3.5-times	
	Roots (intact)	10 nM–1 μM	↑ Up to 1.4-times	
		10–100 μM	↓ Up to 0.7-times	
Hordeum vulgare	Coleoptile (sections)	10 nM–1 μM	↑ Up to 3.3-times	
		10–100 μM	↓ Up to 0.8-times	
Avena sativa	Coleoptile (sections)	10 nM	↑ Up to 1.2-times	
		0.1 μM	↓ Up to 0.3-times	
	Roots (intact)	0.1 nM–100 μM	↓ Up to 0.7-times	
Brassica juncea	Roots (intact)	0.1 μM	↑ Up to 1.2-times	(Chen et al. 2009)
		100 μM	↓ Up to 0.6-times	
Brassica oleracea	Whole plant	1–10 μM	↑ Up to 1.2-times	(Posmyk et al. 2008)
	Whole plant in cooper	10 μM	↑ Up to 1.5-times	
Helianthus annuus	Hypocotyl	5–30 μM	↑ Up to 2.2-times	(Mukherjee et al. 2014)
	Hypocotyl with NaCl	15 μM	↑ Up to 1.3-times	
	Primary root	15–30 μM	↑ Up to 1.8-times	
	Primary root with NaCl	15 μM	↑ Up to 1.2-times	
Oryza sativa	Seminal root (wild type)	0.5–5 μM	↑ Up to 1.4-times	(Park and Back 2012)
	Seminal root (transgenic line)	–	↑ Up to 1.5-times	
Arabidopsis thaliana	Lateral roots	100–300 μM	↑ Up to 3.2-times	(Pelagio-Flores et al. 2012)
	Whole plant	10–100 μM	↑ Up to 1.4-times	(Bajwa et al. 2014)
	Whole plant at 4 °C	30 μM	↑ Up to 1.7-times	
	Primary roots	10–40 μM	↑ Up to 1.3-times	
	Primary roots at 4 °C	30 μM	↑ Up to 1.4-times	
	Primary roots	0.1 μM	↑ Up to 1.1-times	(Hernández et al. 2015)
	Primary roots	10–1000 μM	↓ Up to 0.4 times	(Wang et al. 2016)
Glycine max	Leaf area	50–100 μM	↑ Up to 1.3-times	(Wei et al. 2015)
Cynodon dactylon	Whole plant	4–100 μM	Not increases in growth	(Shi et al. 2015b)
	Whole plant in ClNa	20–100 μM	↑ Up to 1.4-times	
	Whole plant in drought	20–100 μM	↑ Up to 1.4-times	
	Whole plant at 4 °C	20–100 μM	↑ Up to 1.5-times	
Prunus avium x Prunus cerasus	Primary root	0.1–1 μM	↑ Up to 2.5-times	(Sarropoulou et al. 2012a, b)
Cucumis sativus	Shoot in osmotic stress	100–500 μM	↑ Up to 1.7-times	(Zhang et al. 2013)
	Root in osmotic stress	100–500 μM	↑ Up to 3.0-times	
Punica granatum	Adventitious roots	5 μM	↑ Up to 1.4-times	(Sarrou et al. 2014)
Solanum lycopersicum	Shoot with cadmium	25–500 μM	↑ Up to 2.4-times	(Hasan et al. 2015)
	Root with cadmium	25–500 μM	↑ Up to 3.3-times	

Table 1 continued

Specie	Organ	Melatonin treatment on growth	Maximum growth response (vs. without melatonin)	References
Malus hupehensis	Whole plants	0.1 µM	Not increases in growth	(Li et al. 2016)
	Whole plants in ClNa	0.1 µM	↑ Up to 1.2-times	
	Whole plants in K deficiency	0.1 µM	↑ Up to 1.3-times	
	Whole plants in all-nutrient deficiency	0.1 µM	↑ Up to 1.7-times	
Zea mays	Leaves	1–10 µM	↑ Up to 1.2-times	(Zhao et al. 2015)
		100–1000 µM	↓ Up to 0.5-times	
	Roots	10 µM	↑ Up to 1.2-times	
		100–1000 µM	↓ Up to 0.6-times	
	Coleoptiles	0.1–100 µM	Not increases in growth	(Kim et al. 2016)
	Coleoptiles with NaCl	10 µM	↑ Up to 1.3-times	
	Roots	1–100 µM	Not increases in growth	
	Roots with NaCl	10 µM	↑ Up to 1.1-times	

Table 2 Effect of exogenous melatonin on rooting in different species

Species	Melatonin treatment	Maximum rooting response (vs. without melatonin)	References
Lupinus albus	10 µM	↑ Up to 6.0-times	(Arnao and Hernández-Ruiz 2007)
Adventitious roots	0.01 µM	↑ up to 3.0-times	
Lateral roots			
Arabidopsis thaliana	450 µM	↑ Up to 2.0-times	(Pelagio-Flores et al. 2012)
Adventitious roots	300–450 µM	↑ up to 3.0-times	(Koyama et al. 2013)
Lateral roots	150–450 µM	No effect	
Root hairs			
Arabidopsis thaliana	Transgenic line	↑ Up to 1.5-times	(Zuo et al. 2014)
Lateral roots	(up to 4-times more melatonin than WT)		
Diverse sweet cherry	0.05–1 µM	↑ Up to 3.8-times	(Sarropoulou et al. 2012a, b)
rootstocks (*Prunus sp.*)			
Shoot tip explants			
Cucumis sativus	100–500 µM	↑ Up to 2.2-times	(Zhang et al. 2013, 2014b)
Lateral roots			
Punica granatum	5 µM	↑ Up to 1.7-times	(Sarrou et al. 2014)
Adventitious roots			
Solanum lycopersicum	25–50 µM	↑ Up to 2.0-times	(Wen et al. 2016)
Adventitious roots			

Despite evidence presented by many authors concerning the possible auxinic action of melatonin, there are some facts that distinguish the mode of action of melatonin from the well-known action of the auxin IAA. Root primordial induction by melatonin is self-dependent of the IAA-signaling pathway. IAA, but not melatonin, is able to activate the auxin-inducible gene expression marker DR5:GUS in *Arabidopsis* (Koyama et al. 2013; Pelagio-Flores et al. 2012). These data suggest that melatonin can act in a parallel way to IAA, in both lateral and adventitious root induction (Arnao and Hernández-Ruiz 2014). This latter suggestion would agree with the observations made concerning many genes up- and down-regulated by melatonin and related with rhizogenesis (up to 320 genes) in *Cucumber sativus*. Some of these transcription factors and also some ethylene-related transcription factors are able to negatively regulate the genes related to rooting and, therefore, to suppress the formation of new roots. Many of the factors named above were regulated by melatonin, allowing the formation of new roots (Zhang et al. 2014b). Curiously, in the first molecular studies of melatonin, the expression pattern of auxin-related genes exhibited minimal changes in melatonin-treated *Arabidopsis* plants. Only one IAA-amino synthase was up-regulated, with no apparent changes in the expression of auxin biosynthetic genes (Weeda et al. 2014). Nevertheless, a recently quantitative real-time PCR study in *Arabidopsis* showed that some transcript levels of YUCCA (*YUC1*, *YUC2*, *YUC5*, *YUC6*, and *TAR2*) proteins, which play important roles in IAA biosynthesis, significantly decreased after treatment with 600 µM melatonin, while other transcripts (*YUC3*, *YUC4*, *YUC7,* and *YUC8*) increased. As a result, the endogenous IAA content in melatonin-treated roots was slightly lower than that of the control (Wang et al. 2016).

Effect of melatonin on root tropism

As regard tropisms, no evidence has previously been provided concerning the possible participation of melatonin in phototropic responses but, the data provided in the present work in the case of root tropism represent the first evidence, to our knowledge, of such a relation. Figure 1 shows the different behaviour of lupin primary roots grown in contact with agar blocks containing IAA or melatonin. In all cases, except the control (agar block without indoles), a significant curvature in the root from the point where the agar block was placed (marked in blue) can be seen. In contrast, no curvature effect was observed in the control seedling, where the root grew straight, parallel to gravitational force. The curvature effect was evident at all three concentrations of IAA tested, and also with melatonin at 10 and 100 µM. Figure 1, inset, shows the growth trajectories of roots containing agar blocks with IAA and melatonin at 10 and 100 µM, respectively. As can be seen, both trajectories are similar in shape and time, and both affect the normal tropic response of the root. The response of the lupin seedlings depicted in Fig. 1 (though limited and preliminary) clearly demonstrates the involvement of melatonin in another response frequently attributed to compounds with auxinic activity: the tropic response. The disruption (through IAA- or melatonin-enriched agar blocks) of the natural level of auxin in lupin roots gave rise to an artificial internal gradient that provoked a high lateral growth of roots, with the consequent loss of verticality. Thus, melatonin induces negative tropism (roots grow away from side to which was applied), similar to the effect of calcium application on maize root apices (Ishikawa and Evans 1992). Once the auxinic gradient was normalized on both sides of the root, vertical growth was re-established guided by the force of gravity (Fig. 1, inset). These qualitative data show the effect that an exogenous imbalance of IAA or melatonin has on the tropic response of lupin root. At present, we are working to obtain more data (qualitative and quantitative) concerning the action of melatonin in this respect.

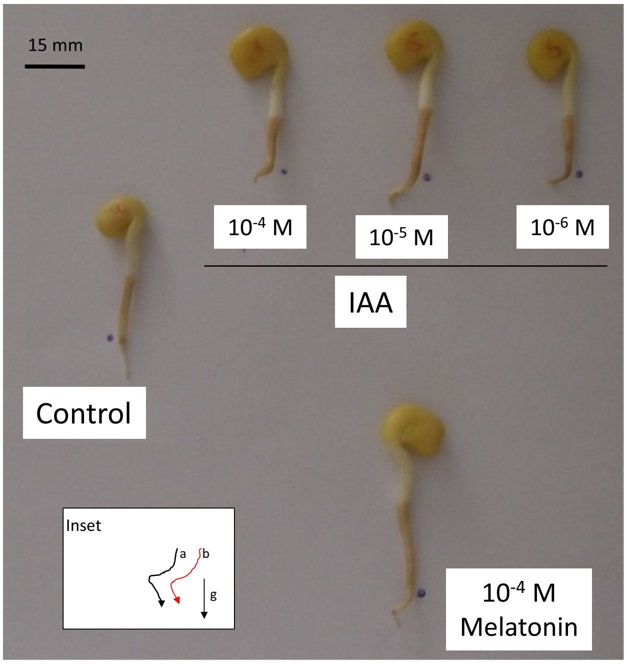

Fig. 1 Representative images of lupin primary root responses 24 h after of the localized treatment (marked with a *blue point*) with agar blocks containing IAA or melatonin at the indicated concentrations. Only an agar block with water was applied in the control treatment. The *inset* shows the trajectories of primary roots treated with (*a*) 100 µM melatonin and (*b*) 10 µM IAA

IAA is clearly involved in tropic responses (gravitropism, phototropism, and others) through the changes that it provokes in the action of the PIN-FORMED (PIN) family of auxin efflux carriers and the auxin-influx carrier protein-1 (AUX1), which belong to a small gene family comprising four highly conserved genes, *AUX1* and *LIKE-AUX1 (LAX)* (amino acid/auxin permease superfamily), which can modulate polar auxin transport in roots (Swarup and Péret 2012). Of the four *AUX/LAX* genes known, melatonin treatment in *Arabidopsis* severely down-regulated *AUX1*, which is involved in the regulation of lateral root development, root gravitropism, root hair development, and leaf phyllotaxy (Swarup et al. 2005). These data show that melatonin may influence the action of auxin through changes in auxin carriers by changing the local IAA gradients (Weeda et al. 2014). Curiously, in lupin roots and some monocot roots, a melatonin gradient similar to the IAA gradient has been described, suggesting that both gradients co-participate in plant growth (Hernández-Ruiz et al. 2004; Hernández-Ruiz and Arnao 2008a). Recently, Wang and co-workers demonstrated that melatonin down-regulated auxin biosynthesis and also the auxin response in

Arabidopsis. Therefore, TIBA (an auxin polar transporter inhibitor) does not co-participate in the reduction of root meristem size by melatonin, indicating that polar auxin transport could be necessary in the regulation of root meristem size by melatonin (Wang et al. 2016).

The above suggests the possibility that melatonin: (i) interacts directly with auxin carriers (as described above) or (ii) alters local IAA gradients. In this last case, only limited data on the inter-relationship between melatonin and IAA levels exist. Table 3 shows the effect that melatonin treatment has on endogenous IAA levels. Only in the case of *Brassica juncea* has an increase in endogenous IAA been described. In other cases using transgenic plants that over-produce melatonin, a substantial decrease in IAA levels was reported. In the cases described in Table 3, the phenotypic effects clearly resemble auxinic responses, including a reduced apical dominance, a high degree of branching, and fewer lateral leaflets. This suggests that melatonin does not replace IAA in the apical dominance function, but that both take part in the same physiological action. Although IAA and melatonin share tryptophan as a precursor in their respective biosynthetic pathways, to date, no conclusive data have been presented to confirm metabolic inter-conversion between melatonin and IAA in plants (Arnao and Hernández-Ruiz 2015). Recently, in *Arabidopsis,* melatonin was seen to negatively regulate IAA biosynthesis while the exogenous application of low IAA concentrations increases, slightly, the endogenous melatonin content in roots, although the data should perhaps be treated as preliminary (Wang et al. 2016).

Proposed model and future perspectives

One of the factors that may explain some of the actions of melatonin in plants is the signaling molecule nitric oxide (NO), which mediates in a great variety of physiological processes during the development of plants, being a modulator of responses to stressors both abiotic and biotic (Di et al. 2015; Hussain et al. 2016).

In tomato seedlings, adventitious root formation induced by melatonin was mediated by NO as a downstream signal. Melatonin increases the endogenous NO level, up-regulating nitrate reductase (which reduces nitrite to NO using NADPH). In addition, melatonin treatment provokes changes in acropetal auxin transport through the up-regulation of several auxin efflux genes (*PIN1, PIN3, and PIN7*) and auxin signaling transduction genes (*IAA19* and *IAA24*). NO increased the level of melatonin in tomato roots, indicating a possible NO feedback mechanism, while it is also interesting that melatonin can act as a scavenger of NO (Wen et al. 2016). By contrast, these same auxin PIN proteins (PIN1/3/7) were down-regulated in *Arabidopsis,* influencing auxin transport (Wang et al. 2016). In *Arabidopsis thaliana*, auxin induces NO production in roots mediated by nitrate reductase and the induction of S-nitrosothiols from proteins, regulating the activation of

Table 3 Effect of melatonin treatment or over-production on endogenous IAA level and phenotypic response

Species	Melatonin treatment or endogenous level	IAA level (vs. control)	Phenotypic effects	References
Brassica juncea/roots	0.01–0.5 µM	↑ up to 2-times	↑ Root growth	(Chen et al. 2009)
Tomato over-expressing sheep SNAT/ HIOMT	↑ Up to 6-times	↓ Up to 7-times	↓ Apical dominance Branching phenotype ↑ tolerance to drought	(Wang et al. 2014)
Arabidopsis thaliana over-expressing apple HIOMT	↑ Up to 4-times	↓ Up to 1.4-times	↑ Lateral roots ↑ biomass ↑ tolerance to drought	(Zuo et al. 2014)
Tomato over-expressing rice indoleamine 2,3-dioxygenase	↓ Up to 4-times	nd	↓ Lateral leaflets Leaflets more flattened and less serrated	(Okazaki et al. 2010)
Tomato derooted plants	15 µM	↑ Up to 2-times in hypocotyls	↑ Adventitious root formation	(Wen et al. 2016)
Arabidopsis thaliana/roots	600 µM	↓ Up to 0.75 times	↓ Root meristem size	(Wang et al. 2016)

SNAT Serotonin *N*-acetyl transferase, *HIOMT* hydroxyindol-*O*-methyl transferase, *nd* not determined cell division and subsequent adventitious or lateral root formation (Correa-Aragunde et al. 2015b).

NO has extensively been reported to regulate auxin responses in roots, adventitious roots, lateral roots, root hair formation, and root gravitropism (Correa-Aragunde et al. 2004; Hu et al. 2005; Lombardo et al. 2006; Pagnussat et al. 2002). Interestingly, the application of NO donors can mimic the effect of auxin, which suggests an important role for NO in auxin-induced processes (Chen and Kao 2012). In addition, NO mediates the auxin response leading to adventitious root formation, demonstrating that NO acts downstream of auxin (Correa-Aragunde et al. 2015a; Pagnussat et al. 2004). Much of these effects can be attributed to the action of NO as a signaling molecule. All these mentioned relationships are represented in a hypothetical model in Fig. 2. This model reflects the curious parallelism between IAA and melatonin data on the three effects studied, in which NO seems to be a common factor. As can be seen, melatonin and auxin use NO as a common signaling molecule in rooting, growth, and tropism. This double control of NO levels probably permits the fine-tuning of responses, without forgetting that NO is also involved in the signaling cascades of other plant hormones, such as abscisic acid. In addition, although not the subject of this review, the protective role that melatonin has been seen to present in diverse situations of stress (biotic and abiotic) may also be mediated by NO. Thus, melatonin-treated plants increase the accumulation of diverse sugars and also glycerol, which increase the endogenous NO level, resulting in an enhancing of innate immunity against bacterial pathogens, via salicylic acid and an NO-dependent pathway in *Arabidopsis thaliana*. In these cases, some models have been presented in which melatonin acts upstream NO and ROS signals, being salicylic acid and jasmonic acid involved (Shi et al. 2012, 2015a; Qian et al. 2015).

Much information on the physiological actions in which melatonin is involved has been published, but very few on its mode of action. This is possibly due to the fact that the presence of a melatonin receptor in plants has not been confirmed, and therefore, any effects of this methoxindole have been attributed to its antioxidant activity, a non-specific action. Nonetheless, the factors currently considered the most determinant in the action of melatonin in plants are conditioned by the following questions: Do melatonin receptors exist in plants? Where are they located at cellular level, (membrane/nucleus)? In which tissue(s) is melatonin synthesized? How and through what system (xylem/phloem) is melatonin transported? Which are the melatonin metabolic pathways, including those involving biosynthesis, degradation, and conjugation? In which tissues can melatonin act? What are the elements of the signal transduction chain? Can melatonin share items with other plant hormones? Only when we know the answers to these questions will we be able to understand the multiple interrelations that may exist between melatonin and many plant hormones (auxin, gibberellins, abscisic acid, ethylene, jasmonic acid, and salicylic acid). As regard the auxinic actions of melatonin, current data suggest that melatonin is able to regulate the processes of plant growth and the *de novo* generation of roots, and modulate the tropic responses of these roots.

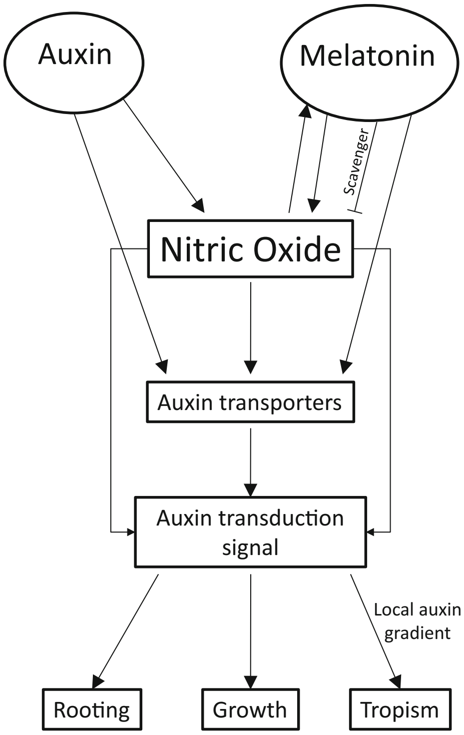

Fig. 2 Hypothetical model representing the activation of the three physiological responses studied: rooting, growth, and tropism, mediated by auxin and melatonin. Melatonin and auxin induce nitric oxide biosynthesis and changes in the auxin transporters of cells. Nitric oxide induces melatonin biosynthesis and melatonin can also act as nitric oxide scavenger. Localized gradients of auxin or melatonin in primary root cause tropic response (tropism)

Author contribution statement MBA conceived and designed work. JHR designed and performed the experiments. MBA and JHR analyzed the data. MBA wrote the manuscript. MBA and JHR critically revised the final version of manuscript. MBA and JHR read and approved the final article.

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