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MINIREVIEW

Blood Pressure Modulation and Cardiovascular Protection by Melatonin: Potential Mechanisms Behind

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Summary

The production of the pineal hormone melatonin is synchronized with day-night cycle *via* multisynaptic pathway including suprachiasmatic nucleus linking several physiological functions to diurnal cycle. The recent data indicate that impaired melatonin production is involved in several cardiovascular pathologies including hypertension and ischemic heart disease. However, the mechanisms of melatonin effect on cardiovascular system are still not completely understood. The activation of melatonin receptors on endothelial and vascular smooth muscle cells and antioxidant properties of melatonin could be responsible for the melatonin effects on vascular tone. However, the data from *in vitro* studies are controversial making the explanation of the melatonin effect on blood pressure *in vivo* difficult. *In vivo*, melatonin also attenuates sympathetic tone by direct activation of melatonin receptors, scavenging free radicals or increasing NO availability in the central nervous system. The central and peripheral antiadrenergic action of chronic melatonin treatment might eliminate the mechanisms counter-regulating decreased blood pressure, providing thus additional cardioprotective mechanism. The extraordinary antioxidant activity and antilipidemic effects of melatonin may enhance the modulation of blood pressure by melatonin and probably play the most important role in the amelioration of target organ damage by chronic melatonin treatment. Further investigation of these mechanisms should provide novel knowledge about pathophysiological mechanisms of cardiovascular diseases, additional explanation for their circadian and seasonal variability and potentially generate new impulses for the development of therapeutic arsenal.

Key words
Melatonin • Hypertension • Nitric oxide • Free radicals • Sympathetic nervous system

Introduction

Since the identification of melatonin by Lerner *et al.* (1958), it has been shown, that melatonin is involved in the regulation of many physiological systems, including cardiovascular system (Važan *et al.* 2003, 2004). Melatonin influences blood pressure (Arangino *et al.* 1999), myocardial contractility (Abete *et al.* 1997) and

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increases the antioxidant reserve (Girouard *et al.* 2004). Melatonin receptors were discovered in the heart (Pang *et al.* 2002) and arteries (Masana *et al.* 2002). Moreover, decreased melatonin levels were reported in various pathological conditions including hypertension with non-dipper pattern (Jonas *et al.* 2003), impairment of heart failure (Girotti *et al*. 2003), ischemic heart disease (Brugger *et al.* 1995), or in patients after acute myocardial infarction (Dominguez-Rodriguez *et al.* 2002). Melatonin is often available as dietary supplement without need of medical prescription, what increases the chance of regular melatonin intake by patients suffering from a cardiovascular disease. Therefore, melatonin is coming to the cutting edge of cardiovascular research and its effects on cardiovascular system in clinical situation are being discussed (Šimko and Paulis 2007). However, results obtained on models with different analytical level are partially controversial and the mechanisms behind melatonin influence on cardiovascular system are still not completely understood.

In this review we have summarized and confronted data from studies on isolated cells, animal experiments as well as small clinical trials. Our major aim was to suggest important mechanisms and pathways that might be responsible for the physiological and clinical importance of melatonin in the cardiovascular system. We paid special attention to the regulation of blood pressure, one of the most important determinants of cardiovascular risk. Moreover, we focused on melatonin impact on cardiovascular remodeling and myocardial reperfusion injury. The details about protection against damage to the brain (Reiter *et al*. 2005) or kidney (Tylicki *et al.* 2003) by melatonin represent, however, an extensive topic and are behind the scope of this review.

Background

The regulation of melatonin synthesis and melatonin concentrations

Melatonin, an endocrine product of the pineal gland, is formed predominantly during night-time. Light has an inhibitory effect on pineal melatonin secretion (Wurtman *et al.* 1964). Melatonin release is synchronized with day-light cycle by a multisynaptic pathway. Light stimulates retina to modulate the activity of suprachiasmatic nucleus (SCN) (Moore 1996), the site of the master biological clock (Dubocovich *et al.* 1998). GABA-ergic neurons are projected from SCN to paraventricular nucleus (PVN) (Moore 1996) intermittently inhibiting the constant excitatory output for sympathetic intermediolateral nucleus (Kalsbeek *et al.* 2000). The sympathetic stimulation from intermediolateral nucleus is after interpolation in superior cervical ganglion finally directed to the epiphysis to induce melatonin synthesis (Moore 1996). The activity of rate limiting enzymes in the epiphysis is regulated by norepinephrine binding to pineal β_1- and α_1-adrenoceptors (Klein *et al*. 1983, Reiter 1991, Ribelayga *et al.* 1997). Moreover, other neurohumoral systems, e.g. the local renin-angiotensin system, may modify melatonin secretion as well (Baltatu *et al.* 2002). The interference of melatonin synthesis and action with other neurohumoral systems plays an important role in the modulation of cardiovascular functions by melatonin. The concentrations of melatonin in the sera of healthy subjects reach 10^{-10} to 10^{-9} mol/l during the night and an order lower value during the day (Kennaway and Voultsios 1998). It should be noted, that physiological blood levels of melatonin do not reach peak concentrations achieved by pharmacological application (10^{-7} to 10^{-5} mol/l after 60-150 min) (Waldhauser *et al.* 1984, DeMuro *et al.* 2000).

Specific mechanisms of melatonin effects

Many authors suppose that melatonin acts mainly *via* its membrane receptors (Costa *et al.* 1995). However, melatonin's lipophilic nature allows it to act also intracellularly i.e. on its nuclear receptor (Mor *et al.* 1999).

Since the identification of melatonin binding site (Vaneček *et al.* 1987), two G-protein-coupled melatonin membrane receptor subtypes have been identified in mammals: MT_1 (formerly Mel 1a or mt_1) and MT_2 (Mel 1b) (Dubocovich *et al.* 1998). The MT_1 receptor was shown to associate with various second messengers: G_i-coupled decrease in cyclic AMP levels (Capsoni *et al.* 1994, Reppert *et al.* 1996, Witt-Enderby and Duboccovich 1996), G_q-coupled phospholipase-C activation resulting in increased cytosolic Ca^{2+} (Brydon *et al.* 1999) or G-coupled activation of the Kir 3 K-channels (Nelson *et al.* 1996). MT_2 receptor was demonstrated to couple with G_q-protein mediated phosphatidylinositol-4,5-bisphosphate hydrolysis (Dubocovich 1995). The third receptor type MT_3, which has a lower affinity, is probably not coupled with G protein (Mor *et al.* 1999).

The highest density of melatonin receptors was shown to be in central nervous system, particularly in the adenohypophysis (Williams and Morgan 1988, Malpaux

et al. 1995, 2001), SCN (Vaneček *et al.* 1987, Vaneček and Janský 1989), PVN (Duncan *et al.* 1989) and area postrema (Williams *et al.* 1995). In the cardiovascular system melatonin receptors were first revealed in 1990 in rat caudal artery (Viswanathan *et al.* 1990). MT_1-receptor was identified in chicken (Pang *et al.* 2002) and human (Ekmekcioglu *et al.* 2001) coronary arteries as well as in chicken (Pang *et al.* 1993) and rat (Abete *et al.* 1997) heart, whereas MT_2-receptor in the human heart, coronary arteries and the aorta (Ekmekcioglu *et al.* 2003). The precise localization of these receptors is not completely revealed. It was hypothesized that while MT_1-receptor is localized primary on vascular smooth muscle cells, the MT_2-receptor appears on endothelial as well as vascular smooth muscle cells (Masana *et al.* 2002).

Non-specific mechanisms of melatonin effects

Despite the high density of melatonin receptors in the central nervous system, and on the periphery as well, other mechanisms of melatonin action independent on specific receptors have been reported.

The antioxidant properties of melatonin, especially in pharmacological doses seem to be the most pronounced (Ianas *et al.* 1991). Melatonin was able to reduce lipid peroxidation more effectively than vitamin C or E (Gitto *et al.* 2001). The potent antioxidant ability can be explained by the potential to scavenge hydroxyl (Bromme *et al.* 2000), superoxide (Sewerynek *et al.* 1996), peroxyl (Pieri *et al.* 1994) but also NO (Noda *et al.* 1999) free radical. The reported NO scavenging could have adverse effects on cardiovascular system. Okatani *et al.* (2001) attributed the vasoconstriction of the human umbilical artery observed after melatonin administration to the potential of melatonin to scavenge NO. However, vasoconstriction was seen only at high melatonin concentrations (Okatani *et al.* 2001) and *in vitro* experiments have shown that melatonin scavenges NO only in the presence of oxygen and it possibly interacts with peroxynitrite rather than NO alone (Blanchard *et al.* 2000). The antioxidant activity of melatonin is enhanced by the induction of antioxidant enzymes (Antolin *et al.* 1996, Barlow-Walden *et al.* 1995), the stability of the oxidized form of melatonin (Kojšová *et al.* 2006) and high free radical scavenging activity of melatonin metabolites as well (Tan *et al.* 2007). The antioxidant properties of melatonin *in vivo* are demonstrated by the ability of melatonin to reduce ischemia-reperfusion injury in various organs, including the heart (Tan *et al.* 1998), kidney (Sahna *et al.* 2003), brain (Cho *et al.* 1997) and liver (Sewerynek *et al.* 1996).

Non-specific mechanisms of melatonin action can also reside in direct interaction with calmodulin (Turjanski *et al.* 2004), inhibition of Ca^{2+} channels (Satake *et al.* 1986, Shibata *et al.* 1989) or calcium pump stimulation observed in cardiomyocytes (Chen *et al.* 1993). In the central nervous system the ability of melatonin to bind and activate GABA-receptors (Wang *et al.* 2003) along with its ability to reduce oxidative load and to enhance NO signaling may also participate in the central regulation of sympathetic tone.

Melatonin and blood pressure

Since hypertension is one of the most prevalent and pronounced risk factors for various cardiovascular alterations, the effect of melatonin on blood pressure deserves special attention among its influence on cardiovascular system.

Experimental hypertension

In experimental conditions the surgical removal of epiphysis, pinealectomy, which was associated with decreased melatonin production, caused vasoconstriction (Cunnane *et al.* 1980), unchanged cardiac output (Harlow 1987) and temporary hypertension in adult rats (Zanoboni and Zanoboni-Mucciaci 1967, Zanoboni *et al.* 1978). Administration of melatonin reversed pinealectomy-induced hypertension (Holmes and Sudgen 1976). The exposure of experimental animals to continuous light (24 hours/day), which prevented the nocturnal rise of melatonin serum levels (Brown *et al.* 1991), also resulted in suppression of circadian heart rate and blood pressure variability (Briaud *et al.* 2004). Therefore under the experimental conditions it could be even spoken about "melatonin-deficient" hypertension.

In spontaneously hypertensive rats (SHR) melatonin production was shown to decline with aging more rapidly than in normotensive rats (Kawashima *et al.* 1987). The pharmacological treatment with melatonin for five days in adult SHR resulted in graduate decrease in blood pressure, heart rate and plasma renin activity (Kawashima *et al*. 1987). Although in some studies melatonin administration to SHR reverted established hypertension only partially (Pecháňová *et al.* 2007), its antihypertensive effect was more pronounced than the effect of the antioxidant N-acetylcysteine (Kojšová *et al.* 2006) and was comparable with the effect of spironolactone (Paulis *et al.* 2006).

In another experiment in melatonin-treated SHR, the reduction of blood pressure and heart rate was associated with increased endothelium-dependent vasodilatation and increased sensitivity to NO-synthase inhibitor (Girouard *et al.* 2001) suggesting improved NO signaling. This hypothesis is also supported by the observation of increased cytosolic Ca^{2+} levels in endothelial cells (Pogan *et al.* 2002), which may result in increased NO production *via* enhancement of NO-synthase activity, increased cyclic GMP levels and decreased intracellular Ca^{2+} in smooth muscle cells with subsequent vasodilatation (Anwar *et al.* 2001). Interestingly, the antioxidant N-acetylcysteine reduced blood pressure, heart rate and catecholamine concentration and β-adrenoceptor expression (both impaired in SHR) in a similar level as melatonin (Girouard *et al.* 2003). Moreover, the decreased blood pressure and improved baroreflex in SHR correlated with improved antioxidant capacity after long-term melatonin administration (Girouard *et al*. 2004) suggesting an association of antioxidant melatonin properties with its ability to decrease sympathetic tone.

Acute administration of melatonin lowered blood pressure and reduced norepinephrine blood levels in SHR (K-Laflamme *et al.* 1998). *In vitro,* melatonin attenuated constriction of aortic ring in SHR by inhibiting phospholipase C cascade independently on MT-receptor or α_1-adrenoceptor blockade (K-Laflamme *et al.* 1998). Since similar inhibitory effect was also achieved with an antioxidant enzyme superoxide dismutase, it might be assumed that the beneficial effect of melatonin was mediated by its ability to prevent excess oxidative load, which was reported to contribute to enhanced vasoconstriction in spontaneous hypertension (Wu *et al.* 1998).

Melatonin in normotensive and hypertensive humans

The blood pressure lowering effect of melatonin was reported in healthy women receiving contraception (Cagnacci *et al.* 1997), postmenopausal women on hormonal substitution therapy (Cagnacci *et al.* 2001) and healthy men (Arangino *et al.* 1999). The reduction of blood pressure after acute melatonin administration was associated with reduced norepinephrine levels and pulse index (Cagnacci *et al.* 1998). The unchanged heart rate and decreased pulse index in these experiments indicate that the blood pressure decrease after melatonin is *in vivo* mediated by attenuation of the peripheral resistance achieved potentially by increased NO formation (Cagnacci *et al.* 2001).

Impaired circadian rhythm of autonomic tone was observed in several studies on hypertension (Guzzetti *et al*. 1991, Nakano *et al.* 2001). This phenomenon was associated with disturbed neurotransmission in supra-chiasmatic nucleus as proven on humans *post-mortem* (Goncharuk *et al.* 2001), which is the regulatory center for melatonin secretion (Klein and Weller 1972) and autonomic tone (Scheer *et al.* 1999, 2001). Decreased nocturnal melatonin concentrations were observed in patients with non-dipping blood profile (Zeman *et al.* 2005). Although in a double-blind randomized cross-over study acute melatonin administration failed to influence blood pressure, chronic 3-week melatonin administration reduced blood pressure and amplified the night time blood pressure decrease (Scheer *et al.* 2004). The authors explained the inability of melatonin to decrease blood pressure after acute administration by the possible involvement of SCN. While in previous studies melatonin was reported to decrease blood pressure after administration during day-time when the SCN activity is high, in this study melatonin was given before sleep onset, when the SCN activity is low and further inhibition is hardly possible. On the other hand, the long-term regular intake before sleep supported normal rhythm in melatonin concentrations and was therefore effective in restoring circadian variability in blood pressure (Scheer *et al.* 2004).

The mechanisms of the effect of melatonin on blood pressure

From the mechanistic point of view, blood pressure is the function of cardiac output and peripheral resistance. Studies aimed to investigate the direct influence of melatonin on vascular reactivity were performed in various laboratories. However, the data obtained from these experiments are partly contradictory. Melatonin was reported to have no effect on basal arterial tone (Monroe and Watts 1998), to cause vasoconstriction (Doolen *et al.* 1998) or to cause vasodilatation (Weekly 1991, 1993).

Also the correlation of these findings with known intracellular pathways mediating the effects of melatonin remains controversial. The constrictive effect of melatonin can be explained by receptor mediated decrease in cyclic AMP levels (Capsoni *et al.* 1994, Witt-Enderby and Dubocovich 1996) and phosphatidyl-inositol-4,5-bisphosphate hydrolysis which was reported

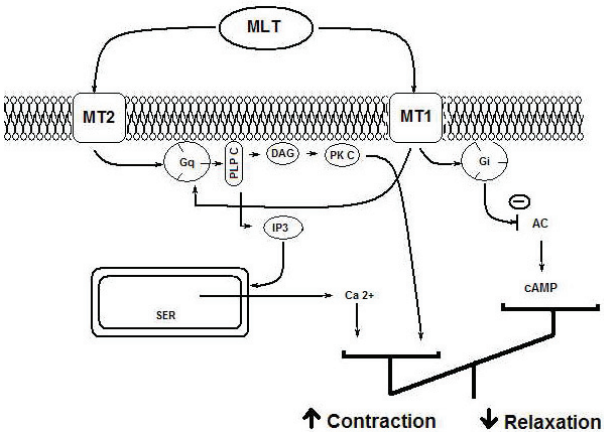

Fig. 1. Potential pathways directly mediating melatonin-induced vasoconstriction *in vitro*. Melatonin resceptors associated with G_i and G_q proteins decrease cyclic AMP levels (Capsoni *et al.* 1994, Witt-Enderby and Dubocovich 1996) and stimulate phosphatidylinositol-4,5-bisphosphate hydrolysis (Dubocovich 1995). Activation of melatonin receptors on vascular smooth muscle cells should therefore diminish vasodilatation or enhance vasoconstriction. MLT, melatonin, MT1, MT2, melatonin receptor MT_1, MT_2, respectively, Gq, Gi, G-proteins, PLP C, phospholipase C, DAG, diacylglycerol, PK C, protein kinase C, IP3, inositol-1,4,5-trisphosphate, SER, smooth endoplasmatic reticulum, cAMP, cyclic adenosine monophosphate.

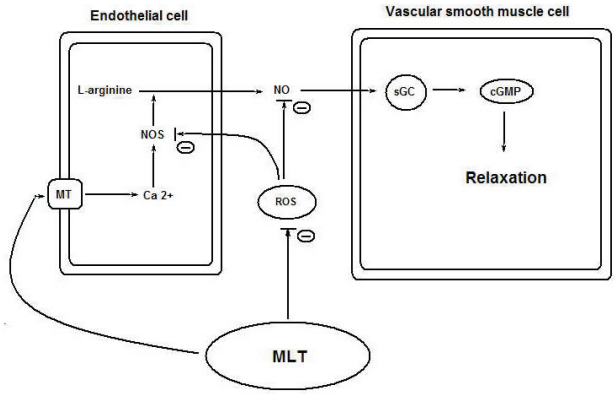

Fig. 2. Possible pathways mediating melatonin-induced vasodilatation. The activation of MT_2 receptors on endothelial cells could increase cytosolic Ca^{2+} in endothelial cells (Pogan *et al.* 2002). Activated endothelial cells are stimulated to NO production. Antioxidant action of melatonin may further enhance the formation and availability of NO, which stimulates guanylate cyclase in smooth muscle cells leading to vasodilatation. MT, melatonin receptors, NOS, NO-synthase, ROS, reactive oxygen species, sCG, solubile guanylate cyclase, cGMP, cyclic guanosine monophosphate.

by several *in vitro* studies (Dubocovich 1995) (Fig. 1).

Despite the fact that the vasodilatation after melatonin is congruent with decreased blood pressure after melatonin administration, it is difficult to explain it on the basis of melatonin receptor stimulation. The activation of melatonin receptors in the majority of experimental models was associated with cyclic AMP decrease and phosphatidylinositol-4,5-bisphosphate hydrolysis, which lead to inhibition of vasodilatation or to vasoconstriction. Nevertheless, the activation of MT_2 receptors on endothelial cells (Masana *et al.* 2002) could increase cytosolic Ca^{2+} in endothelial cells, which was observed by Pogan *et al.* (2002). Activated endothelial cells are then stimulated to increase the production of NO, which is additionally protected by antioxidant properties of melatonin. This hypothesis is supported by the findings of Anwar *et al.* (2001) who observed decreased oxidative load and increased NO in blood serum associated with decreased cytosolic Ca^{2+} and increased cyclic GMP in vascular smooth muscle cells (Fig. 2).

The consistent results of melatonin influence on blood pressure from *in vivo* experiments and clinical trials are in contrast with inconsistent data from *in vitro* experiments and suggest the involvement of central regulatory mechanisms in the mediation of melatonin effects on blood pressure *in vivo*. Improved baroreflex responses (Girouard *et al.* 2004), decreased sympathetic output (K-Laflamme *et al.* 1998) and association of decreased heart rate or cardiac output with blood pressure fall after melatonin administration (Kawashima *et al.* 1987, Arangino *et al.* 1999, Scheer *et al.* 2003) support the idea of central action of melatonin.

Although, the mechanisms participating in central effect of melatonin are yet not completely known, several pathways can be suggested:

First, neurons in PVN directly or after interpolation in rostral ventrolateral medulla (RVLM) project to medullar intermediolateral nucleus, which innervates sympathetic ganglia generating thus sympathetic tone and regulating arterial pressure (Coote *et al.* 1998, Pyner and Coote 2000, Cano *et al.* 2001, 2004, Stocker *et al.* 2006). The constant excitatory output of PVN is intermittently inhibited by GABA-ergic innervation from SCN (Kalsbeek *et al.* 2000), which enables circadian oscillations in sympathetic output synchronized with day-light (Buijs *et al.* 1999, Scheer *et al.* 2003). The same sites (SCN, PVN, intermedioalteral nucleus) are also responsible for the excitation of sympathetic neurons in the superior cervical ganglion that regulates pineal melatonin synthesis (Moore 1996). It could be hypothesized that the modulation of SCN activity by melatonin (Reppert *et al.* 1988, Dubocovich *et al.* 1998) alters sympathetic tone and thus represents a protective mechanism against excessive sympathetic excitation. Moreover, neurons in area postrema were suggested to be epigenetically modified by melatonin (Irmak and Sizlan 2006). These neurons are believed to

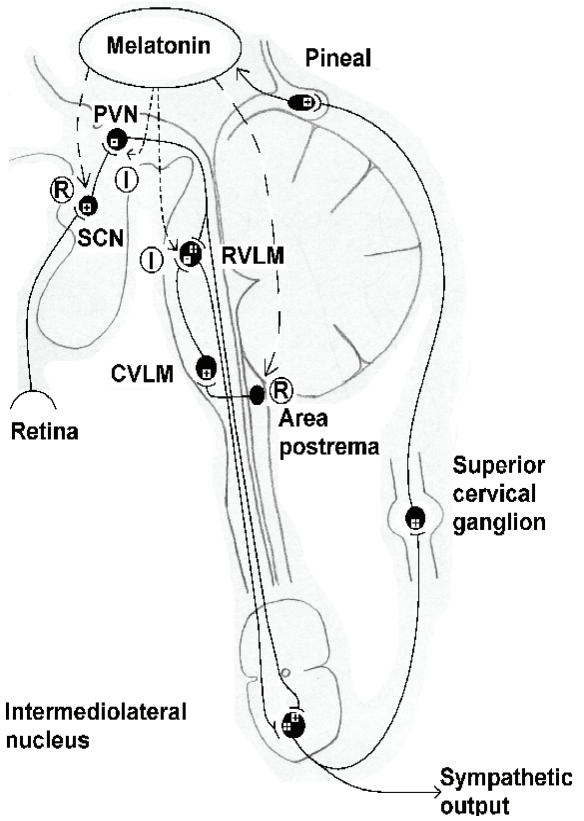

Fig. 3. Possible modulation of central sympathetic tone mediating melatonin effect on blood pressure. First, melatonin may bind to its receptors especially in SCN and area postrema, which show an especially high receptor density (Vaneček *et al.* 1987, Williams *et al.* 1995). Second, melatonin may enhance inhibitory GABA (Wang *et al.* 2003) and NO (Rossi *et al.* 2004) signalization in the PVN and RVLM (Patel *et al.* 2001). SCN, suprachiasmatic nucleus, PVN, paraventricular nucleus, RVLM, rostral ventrolateral medulla, CVLM, caudal ventrolateral medulla, + excitatory synapse (glutamate, acetylcholine or norepinephrine), – inhibitory synapse (GABA, γ-aminobutyric acid), R, potential modulation by receptor binding, I, potential modulation by enhancement of inhibitory GABA and NO signaling.

set reference point for blood pressure regulation (Irmak and Sizlan 2006) and inhibit the activity of RVLM through caudal ventrolateral medulla (CVLM) (Patel *et al.* 2001).

Second, GABA-ergic signalization is involved in the inhibition of PVN by SCN (Kalsbeek *et al.* 2000) and in the inhibition of RVLM by CVLM (Patel *et al.* 2001). Melatonin was reported to enhance GABA-ergic signalization (Wang *et al.* 2003), which may contribute to inhibition of these nuclei and subsequent decrease in sympathetic tone.

Third, NO formation was shown to potentiate GABA-ergic inhibitory effects in PVN (Rossi *et al.* 2004) and RVLM (Patel *et al.* 2001). The potential of melatonin to increase NO availability may additionally augment inhibition in these areas (Fig. 3).

The effect of melatonin on cardiovascular remodeling

Beside the effects of melatonin on blood pressure, an important question remains whether melatonin treatment may beneficially modify the deterioration of organ structure and function associated with hypertension, coronary artery disease or dyslipidemia.

The effect of melatonin on vascular structure

Although extensive attention was devoted to the modulation of vascular function by melatonin, the effect of melatonin on vascular morphology was less intensively investigated. Nevertheless, improved NO production and decreased oxidative load after melatonin administration (Anwar *et al.* 2001) may lead to prevention of endothelial structural alterations. Most promising results were obtained in models with altered metabolic conditions, which produce pronounced endothelial damage. In rats on high-fat diet melatonin administration attenuated atheromatous changes in arteries along with the normalization of blood pressure, body weight, blood glucose, improvement of antioxidant capacity and lipid profile (Hussein *et al.* 2007). In hypertriglyceridemic rats, melatonin prevented intimal infiltration by foam cells induced by cholesterol in association with modified plasmatic fatty acid composition (Pita *et al.* 2002). The prevention of endothelial damage may help to explain more effective blood pressure reduction in type 1 diabetic patients than in controls (Cavallo *et al.* 2004). However, under the conditions of less compromised endothelium melatonin can also improve endothelial function. The prevention of early atheromatous changes and endothelial damage with direct impact on arterial function may help several organs especially the brain to cope with ischemia-reperfusion injury or other pathological conditions.

Deficit of melatonin induced by pinealectomy decreased cross-sectional area, attenuated compliance and increased stiffness of rat cerebral arterioles, whereas low-dose melatonin treatment prevented the development of these alterations (Regrigny *et al*. 2001). Beside this fact, there is lack of other evidence for modulation of medial structure by melatonin. Yet, there are several attributes of melatonin, which could potentially lead to improvement of vascular remodeling. Numerous melatonin-induced changes such as attenuation of hemodynamic overload, decrease in sympathetic output (Girouard *et al.* 2003),

reduction of oxidative load (Tan *et al*. 2007), increase in bioavailability of NO, which has antiproliferative and antiproteosynthetic effects (Šimko and Šimko 2000) as well as reduction of serum cholesterol levels and lipid profile normalization (Wakatsuki *et al.* 2001, Sandyk and Awerbuch 1994) encourage future investigations of the modulation of vascular structure by melatonin.

The effect of melatonin on left ventricular hypertrophy

Melatonin prevented cardiac hypertrophy in hyperthyroid rats along with reduced oxidative load and altered expression of metabolically important genes (Ghosh *et al.* 2007). This study indicates that the effect of melatonin on hemodynamic overload, NO availability, free radicals and lipid profile may modify myocardial remodeling as well. On the other hand, despite the reduction of blood pressure melatonin failed to ameliorate left ventricular hypertrophy in SHR (Šimko *et al.* 2006), but some promising results were obtained in melatonin-deficient models. Pinealectomy increased heart weight and fibrosis (Mizrak *et al.* 2004, Sahna *et al.* 2002) and continuous light increased collagen types I/III ratio (Paulis *et al.* 2007). Since the increase in heart weight was proportional to the increase in body weight (Sahna *et al.* 2002, Paulis *et al.* 2007) and the gain in heart weight did not correlate with blood pressure rise (Sahna *et al.* 2002), the hemodynamic load was probably not the decisive factor for the development of these alterations. Pinealectomy was also associated with several metabolic alterations including enhanced isoproterenol-induced lipolysis in rats (Borges-Silva *et al.* 2005), hypercholesterolemia and transient hypertriglyceridemia in type 2 diabetic rats (Nishida *et al.* 2003) and hypercholesterolemia and hyperlipidemia in rabbits (Damian 1976). Insufficient effect of melatonin on food efficiency and growth factors and insulin levels (Wolden-Hanson *et al.* 2000) in melatonin-deficient conditions may participate in the development of cardiac remodeling. It can be expected that blood pressure reducing and metabolic impact of melatonin may be especially beneficial against cardiovascular damage associated with metabolic syndrome.

Melatonin and cardiac reperfusion injury

There is evidence for cardioprotective effect of melatonin against ischemia-reperfusion injury. Melatonin reduced the infarct size/risk area (Chen *et al.* 2003, Sahna *et al.* 2005) and the incidence of reperfusion arrhythmias (Lagneux *et al.* 2000, Lee *et al.* 2002, Sahna *et al.* 2002).

Since ischemia is associated with formation of oxygen free radicals from the residual molecular oxygen (Jennings *et al.* 2001), the cardioprotective effect is probably associated with melatonin ability to scavenge free radicals (Allegra *et al.* 2003, Sahna *et al.* 2005) and to induce the expression of antioxidant enzymes (Reiter 2000). Melatonin was even more efficient in reducing the severity of reperfusion arrhythmias than the antioxidant vitamin C (Tan *et al.* 1998). There are following possible explanations for the especially high effectiveness of melatonin in preventing reperfusion injury of various organs:

First, beside antioxidant action other mechanisms take part in the protective effect of melatonin. Melatonin was reported to decrease cytosolic calcium in cardiomyocytes (Chen *et al.* 1993) what could modify the electrical stability of the myocardium and contribute to protective action of melatonin against ischemia-induced arrhythmias (Lagneaux *et al.* 2000). These changes in cytosolic calcium may result from stimulation of melatonin receptors, which are present in cardiomyocytes (Pang *et al.* 2002) or from other non-specific melatonin effects like direct interaction with calmodulin (Turjanski *et al.* 2004), inhibition of Ca^{2+} channels (Satake *et al.* 1986, Shibata *et al.* 1989) or calcium pump stimulation (Chen *et al.* 1993).

Second, the antioxidant action of melatonin is extraordinary high. Melatonin takes a special place among other antioxidants. Melatonin does not undergo redox cycling (Tan *et al.* 2000), enhances the activity of antioxidant enzymes (Antolin *et al.* 1996, Barlow-Walden *et al.* 1995), and its primary, secondary and tertiary metabolites also posses high free radical scavenging activity (Tan *et al.* 2007). Most importantly, melatonin may penetrate lipid cell membrane (Mor *et al.* 1999) and act as intracellular antioxidant unleashing its scavenging properties.

Conclusions

Melatonin was shown to be involved in several cardiovascular pathologies and its therapeutic use is being considered (Šimko and Paulis 2007). However, the underlying mechanisms of melatonin protection within the cardiovascular system are not properly understood.

Although melatonin decreases peripheral resistance, results of experiments on isolated arteries are partly controversial and vasodilatation cannot be completely explained only by activation of second

messenger cascades associated with melatonin receptors. Thus, more sophisticated approach should be taken into consideration:

On the periphery, receptor-mediated vasoconstriction on vascular smooth cells might be counterbalanced by receptor-mediated NO release from endothelial cells, which is further enhanced by antioxidant properties of melatonin.

In the central nervous system, enhancement of GABA-ergic signaling, scavenging free radicals and augmentation of NO availability may substantially participate in the reduction of sympathetic output resulting in blood pressure decrease observed *in vivo*.

Additionally to the reduction of blood pressure, melatonin may be expected to prevent target cardiovascular damage as well. Melatonin protects heart against ischemia-reperfusion injury by its extraordinary antioxidant activity and its antilipidemic effects may attenuate undesirable vascular alterations.

Further investigation of these mechanisms may extend the knowledge on pathogenetic mechanisms of cardiovascular diseases, provide additional explanation for their circadian and seasonal variability and potentially generate new impulses for the development of novel therapeutic approaches.

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