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COMMENTARY

Melatonin in the management of perinatal hypoxic-ischemic encephalopathy: light at the end of the tunnel?

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Abstract: Perinatal hypoxic-ischemic encephalopathy (HIE) affects one to three per 1,000 live full-term births and can lead to severe and permanent neuropsychological sequelae, such as cerebral palsy, epilepsy, mental retardation, and visual motor or visual perceptive dysfunction. Melatonin has begun to be contemplated as a good choice in order to diminish the neurological sequelae from hypoxic-ischemic brain injury. Melatonin emerges as a very interesting medication, because of its capacity to cross all physiological barriers extending to subcellular compartments and its safety and effectiveness. The purpose of this commentary is to detail the evidence on the use of melatonin as a neuroprotection agent. The pharmacologic aspects of the drug as well as its potential neuroprotective characteristics in human and animal studies are described in this study. Melatonin seems to be safe and beneficial in protecting neonatal brains from perinatal HIE. Larger randomized controlled trials in humans are required, to implement a long-awaited feasible treatment in order to avoid the dreaded sequelae of HIE.

Keywords: melatonin, hypoxia, use, encephalopathy

Introduction

Perinatal asphyxia is defined as the interference of gas exchange or blood flow to and from the fetus in the perinatal age.[1] This interruption could be extended to partial asphyxia and sudden subtotal asphyxia because of a sentry experience or a combination of both.[2] Hypoxic-ischemic injuries to the brain might be the outcome if the perinatal asphyxia is of an adequate intensity or extended beyond the capability of the fetus to compensate.[3,4]

Perinatal hypoxic-ischemic encephalopathy (HIE) affects one to three per 1,000 live full-term births.[5,6] Up to one-fifth of the affected newborns will perish in the postnatal period, and a further 25% will develop severe and permanent neuropsychological sequelae, such as cerebral palsy, epilepsy, mental retardation, and visual motor or visual perceptive dysfunction.[7] Ensuing a hypoxic-ischemic (HI) abuse, an instantaneous phase of neuronal cell injury and enervation of energy stores occurs, followed by oxidative stress and apoptosis.[8] Many crucial neuronal groups are more susceptible to HI injury in newborns than in adults, especially those related to enhanced density and function of excitatory amino acid receptors.[9] The neonatal brain has higher water content, underdeveloped cortex, lower myelin, and a more conspicuous germinal matrix than the mature brain.[10] The morbidity and mortality of infants are strongly affected by their ability to preserve physiologic homeostasis.[11] Melatonin has begun to be contemplated as a good choice in order to diminish the neurological

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sequelae from HI brain injury. Melatonin emerges as a very interesting medication, because of its capacity to cross all physiological barriers extending to subcellular compartments and its safety and effectiveness.[12,13] It effortlessly crosses the blood–brain barrier and the placenta, stretching to subcellular compartments with minimal toxicity and high effectiveness,[12,14] rendering it a relatively safe treatment that could be administered to newborns.[15] The purpose of this commentary is to detail the evidence on the use of melatonin as a neuroprotection agent.

Search strategy and selection criteria

A search in PubMed, Google Scholar, and Embase databases was conducted using different combinations of the following terms: melatonin, perinatal, hypoxic, encephalopathy, treatment, prevention, and strategy. Moreover, the references of the identified articles were searched for further articles. Later, the abstracts and titles were inspected, and studies that were appropriate to the topic of interest were selected. Finally, the search was restricted to manuscripts that were published in Spanish and English from inception till July 2016 (Figure 1).

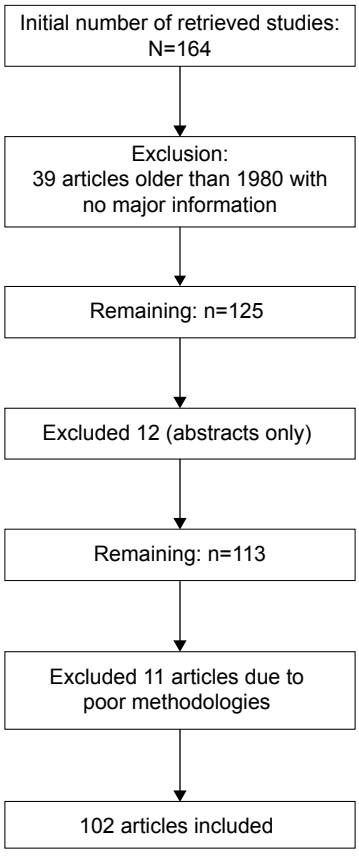

Figure 1 Flow diagram of selection of references.

Melatonin

Melatonin (*N*-acetyl-5-methoxytryptamine) is an endogenously produced indolamine originating from the pineal gland.[16] Melatonin, a naturally appearing neuroendocrine molecule is released as a result of environmental light–dark cycles, effortlessly crosses biological membranes and performs through receptor-dependent and receptor-independent mechanisms to adjust gene expression and cell signaling.[17,18] It has a short half-life.[19,20] The augmentation in serum levels and excretion after oral ingestion of melatonin is rapid (60–150 min).[21] Melatonin is usually metabolized in the liver and excreted by the kidney.[22]

It comprises several other biological functions in various tissues and organs,[23] which involve harmonizing energy metabolism,[24] protecting against oxidative stress,[25] delaying aging,[26] and embellishing immune function.[27]

Autogenous melatonin is indispensable to normal neurodevelopment and preserves the developing brain from injury.[28–30] Maternal melatonin levels are increased during pregnancy and melatonin effortlessly crosses the blood–brain barrier and placenta.[31,32] Neonates born at term produce, to some extent, low pineal melatonin and therefore have deficiencies in diurnal variation for the first week of life.[33] Moreover, another raise in serum melatonin levels occurs in very sick children as well as in patients with cerebrovascular accidents entailing an aspect of melatonin in an autogenous protective reaction.[34,35]

Melatonin accomplishes its strong neuroprotective result through anti-inflammatory, anti-apoptotic, and antioxidant processes through nuclear and cell membrane receptors[28,29,36] and by boosting glial and neuronal development.[37,38] Evolving young brain is easily exposed to free radical damage,[39,40] and the strong free radical-scavenging characteristics of melatonin and its metabolites contribute to a crucial neuroprotective process.[25,41,42]

Under abnormal circumstances, one of the most crucial regulators of apoptotic cell death is mitochondrial deterioration, as the severance of its membrane integrity can limit cell survival by dispensation of apoptotic proteins, impaired calcium homeostasis, and generation of reactive oxygen species.[43]

Studies have shown that melatonin decreased or blocked caspase (a family of protease enzymes playing essential roles in programmed cell death-1) and caspase-3 activation,[44–46] interfered with release of cytochrome *c*,[45,46] decreased *Bad* (B-cell lymphoma 2-associated death promoter),[47,48] and *Bax* (B-cell lymphoma 2-associated X protein)[49] pro-apoptotic proteins, and diminished the

number of terminal deoxynucleotidyl transferase dUTP nick end labeling (TUNEL)-positive cells.[50–53] Melatonin can also produce antiexcitatory effects on neurons through the modulation of gamma-aminobutyric acid and glutamate receptors,[54,55] leading to the decrease in cytosolic calcium concentrations.[56,57]

The approach moderated by melatonin and its main metabolites circumscribes the downregulation of some inflammation-associated molecules such as prostaglandin,[58] cyclooxygenase,[59,60] 5-lipoxygenase,[61] tumor necrosis factor-α, cytokines, interleukin-6, and interleukin-8,[62–65] and also the hindrance of neuronal nitric oxide (nNO).[66,67] Melatonin hence diminishes vascular endothelial growth factor concentration, NO production, and, therefore, vascular permeability that results increased after hypoxic exposure.[68]

Other important functions of melatonin at the molecular level include maintenance of the mitochondrial integrity and the upregulation of antioxidant enzymes.[25,42] Several animal studies have demonstrated that melatonin decreases oxidative injury to cerebral lipids[69–74] and improves cerebral energy breakdown[75] and apoptosis.[74–78] Melatonin promotes protection after HI[75–80] and in lipopolysaccharide-sensitized HI.[81]

Several actions are arbitrated through the G-protein-coupled melatonin receptors in cellular membranes.[82] Other molecular actions of melatonin include its cooperation with molecules such as calmodulin in the cytosol and orphan nuclear receptors.[83] Nonreceptor-mediated actions of melatonin are usually associated with its capability to detoxify reactive oxygen species.[84]

Melatonin as a neuroprotector in animal models

The neuroprotective outcomes of melatonin in the fetal brain have been evaluated in several animal models. Melatonin administration to both preterm and near-term fetal sheep has been demonstrated to decrease oxidative stress[69] and debilitate cell injury and death in the fetal brain, in association with a decreased inflammatory reaction.[79]

Following acute neonatal hemorrhagic brain injury in neonatal rats, melatonin was administered to assess germinal matrix hemorrhage. Lekic et al[85] concluded that melatonin is an efficient antioxidant that can guard the infant's brain from the posthemorrhagic ramifications of mental retardation and cerebral palsy. Husson et al[86] showed that melatonin, through its adenylate cyclase inhibition, showed neuroprotection of the murine periventricular white matter against neonatal excitotoxicity challenge. Mice that received melatonin through the intraperitoneal route expressed 82% reduction in size of ibotenate-induced white matter cysts. The study also concluded that melatonin can bolster secondary lesion repair of white matter lesions. Robertson et al[75] studied the neuroprotective outcome of adding melatonin to therapeutic hypothermia after transient HI in a swine model of perinatal asphyxia using magnetic resonance spectroscopy biomarkers backed by immunohistochemistry. Amid global HI insult, 17 newborn piglets were randomized to receive either therapeutic hypothermia (33.5°C from 2 to 26 h after resuscitation, n=8) or therapeutic hypothermia plus intravenous melatonin (n=9). The dose of melatonin used was 5 mg/kg/h over 6 h initiated at 10 min after resuscitation and repeated for 24 h. The study showed that plasma levels of melatonin were 10,000 times higher in the hypothermia plus melatonin compared to the hypothermia alone group. In addition, melatonin-increased hypothermia greatly decreased the HI-induced area, as projected by the proton magnetic resonance spectroscopy lactate/*N*-acetyl aspartate and lactate/total creatine ratios in the deep gray matter.

Pazar et al[87] investigated the neuroprotective effects of melatonin in an experimental hemolysis-induced hyperbilirubinemia in 72 newborn Sprague–Dawley rats. Phenylhydrazine hydrochloride (PHZ; 75 mg/kg) was injected at 0 and 24 h. The rats were administered either saline or melatonin (10 mg/kg) half an hour prior to the first and second PHZ injections and 24 h after the second PHZ injections. The control group (n=24) were injected with saline, but not PHZ. The study concluded that augmented TUNEL cells in the hippocampus of saline-treated PHZ group were decreased by melatonin treatment, indicating the neuroprotective and antiapoptotic effects of melatonin on the oxidative neuronal damage of the newborn rats with hemolysis and hyperbilirubinemia.

Melatonin as a neuroprotector in human clinical studies

The neuroprotective outcomes of melatonin in the fetal brain have been assessed in several human studies. In a randomized controlled pilot study, Aly et al[16] tested the clinical outcomes of melatonin and neurophysiological aftermath of neonates with HIE. The study included 45 newborns, 30 with HIE and 15 healthy controls. The dose of melatonin used was 10 mg/kg daily for a total of five doses. Serum nitric oxide (NO) and melatonin as well as plasma superoxide dismutase (SOD) were measured for the 45 newborns and at 5 days for the HIE groups (N=30). The study concluded that compared with healthy neonates, the HIE groups had increased melatonin, SOD, and NO. Moreover, at 5 days, the melatonin/hypothermia group

had a higher level of melatonin ($P<0.001$) and decrease in NO ($P<0.001$), and SOD ($P=0.004$). The melatonin/hypothermia group had fewer seizures on follow-up electroencephalogram and fewer white matter abnormalities on MRI. Furthermore, at 6 months, the melatonin/hypothermia group had better mortality rate without developmental or neurological abnormalities ($P<0.001$).

Fulia et al[88] investigated the implications of free radicals in the pathogenesis of neonatal asphyxia and its complications. The study that included 20 asphyxiated newborns measured malondialdehyde (a product of lipid peroxidation) and the nitrite/nitrate levels before and after the administration of melatonin within the first 6 h of life. One group of newborns received a total of 80 mg of melatonin orally (eight doses of 10 mg each separated by 2 h intervals). A single blood sample was gathered prior to melatonin administration, and two further blood samples (at 12 and 24 h) were gathered after the oral administration of melatonin. The study showed that serum malondialdehyde and nitrite + nitrate concentrations in newborns with asphyxia prior to treatment were markedly higher than those in infants without asphyxia. In the asphyxiated group that received melatonin, there was a marked decrease in levels of malondialdehyde and nitrite/nitrate levels at both 12 and 24 h. Thirty percent of the asphyxiated children who did not receive melatonin died within 72 h after birth, whereas all the asphyxiated newborns who received melatonin lived. The study concluded that the melatonin may be useful and advantageous in the management of newborn infants with asphyxia.

Carloni et al[89] investigated whether administering melatonin before or after HI in immature rats provides significant protection and long-lasting benefit on ischemic outcomes. The authors concluded that brain injury was significantly debilitated in the melatonin-treated ischemic group. Signorini et al[90] studied the production of oxidative damage mediators and the likely outcomes of melatonin administration in a model of HIE in newborn rats (7 days old). The study showed that HI induces an augmentation in desferrioxamine-chelatable free iron in the cerebral cortex, which can lead to cerebral oxidative stress, whereas the cerebral injury by the oxidative stress may be avoided by melatonin administration.

Maternal–fetal transfer of melatonin in pregnant women near term has been documented by measuring the concentration of melatonin in the fetal circulation after its administration. The oral administration of 3 mg of melatonin resulted in augmentation in the serum levels with maximum values being observed 2 h after drug administration; the blood levels of melatonin in the umbilical vein corresponded well to those in the maternal vein.[91] These results solidify the concept that melatonin is easily transferred from maternal to fetal circulation and can be an option for its therapeutic use as an antioxidant in patients with preeclampsia.

Safety profiles and possible side effects of melatonin

Human investigations have demonstrated melatonin toxicity to be very low with negligible side effects.[16,88,92] Melatonin can be used in children with severe learning disorders to ameliorate sleep patterns[93,94] and learning disabilities,[93] in neonates with respiratory distress syndrome,[95–97] and in children with attention deficit/hyperactivity disorder and chronic sleep onset insomnia.[98]

Experiments on animals have shown that even high doses of melatonin (200 mg/kg/day) from gestational days 6–19 did not have any teratogenic or any negative side effect on rat newborns.[99] Another study conducted by Sadowsky et al[100] showed that high melatonin dose did not affect the myometrial electromyography activity in the pregnant sheep at 138–142 days gestation (term =147 days gestation).

However, there is a concern with regard to the inhibition effect of melatonin on the synthesis of prostaglandins, and hence on the endocrine and circulatory capacity in the fetus.[101] In addition, there was a report of decreased threshold of seizure activities in four children with neurologic impairments treated with melatonin.[102]

Conclusion

Melatonin seems to be safe and beneficial in protecting neonatal brains from perinatal HIE. However, the number of infants included in the studies were small, and therefore, larger randomized controlled trials in humans are needed to implement a long-waited feasible treatment to avoid the dreaded sequelae of HIE.

Disclosure

The authors report no conflicts of interest in this work.

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