Microreviews in Cell and Molecular Biology 2012

# The effects of melatonin on autophagic cell death

Logan Edmonds
Physiology Junior
Department of Microbiology and Molecular Genetics, Oklahoma State University, Stillwater, OK 74078, USA

**Key Words:**
Anti-cancer, antioxidant, apoptosis, autophagy, cancer, circadian rhythm, cyclosporine A, free radical, melatonin, programmed cell death, sarcopenia, tumor

**Melatonin is known to have effects on the regulation of cellular autophagy. The role of melatonin in inducing and reducing the autophagic processes of cells could be of great importance in reduction of cellular stressors. Recently, melatonin has been found to induce autophagy in cancerous tumor cells. This mechanism of autophagic induction is potentially related to the prevention of tumor growth. Recent advances could reveal more information about the potential uses of melatonin as an anti-cancer agent.**

## Introduction

There are two main divisions of programmed cell death. The first, apoptotic cell death, is a key way in regulating the number of cells present during times of high stress. It is also a key mechanism in the clearance of cells. Apoptosis is typically associated with the de-polymerization and cleavage of cytoskeletal protein components [1]. Autophagic cell death is a method of breaking down and recycling proteins, ribosomes, and organelles. Autophagy acts to maintain homeostasis by disposing of unnecessary proteins and poorly functioning organelles [2]. Autophagy has been shown to have the ability to induce cell death in the event that apoptosis is not functioning and may generally act to suppress cancerous tumors [3]. However, autophagy is not only a type of programmed cell death, but also an important survival mechanism. The identity of autophagy as primarily a route of programmed cell death or as primarily a cell survival response has been greatly debated [1, 3]. Autophagy that acts as a survival mechanism can be observed during times of cellular stress, including metabolic, genotoxic, and hypoxic stressors [1].

Autophagy can be induced by a variety of factors. In cancerous cells, autophagocytosis acts as a form of tumor suppression. In this way, autophagy may have an important role in the progression of cancer, although this is still to be determined through future studies of the autophagic pathway in response to cancerous cells [3]. The immunosuppressive drug Cyclosporine A (CsA) is linked to high prevalence of autophagy of cells. This is most likely due to the general high toxicity of CsA, specifically the ability of CsA to cause neurotoxicity, hepatotoxicity, and nephrotoxicity. Considerable research supports the idea that these toxicities are a result of free radical production. This release of free radical species is a major driving force in autophagic cell death that occurs during times of high toxicity. The autophagy response caused by toxicities from CsA may act as a cell survival mechanism against the CsA toxicities [1]. Muscle diseases and sarcopenia have both been related to high levels of apoptosis and autophagy. Muscle diseases can affect skeletal muscle and significantly change tissue structure or cause weakening and wasting away of muscle. Apoptosis has been noted as a possible mechanism of muscular cell death as a result of muscle diseases. Sarcopenia is associated with the loss of skeletal muscle mass and muscle strength and is prevalent in elderly persons. In cases where skeletal and cardiac muscles are lost due to age associated factors, apoptosis and autophagy may be important causes of this muscle loss. In this way, sarcopenia may be a direct result of the apoptosis and autophagy of muscle cells [2].

Melatonin is a natural hormone produced in the pineal gland of mammals and is known to have multiple

 

anti-cancer characteristics that work antagonistically to tumors [1, 2]. In addition to anti-cancer properties, melatonin has also been shown as a possible agent in reduction of chemotherapeutic side effects [3]. Melatonin has also been observed as a broad-spectrum antioxidant and scavenger of cellular free radicals. Many studies have linked melatonin with a possible anti-apoptotic death rate in cells and a reduction of autophagy. These anti-apoptotic and autophagy inhibiting effects can be attributed to melatonin's ability to act as a strong antioxidant [1, 2]. An opposite effect has been attributed to melatonin in some cancer cell lines. In these cell lines, melatonin has been observed to induce autophagy. However, the exact role of melatonin in cancer cell progression is relatively unknown and the mechanism of autophagosome induction as a result of melatonin treatment in cancer cell lines will require further study. In this review, the role of melatonin on autophagy will be examined and the possible role of melatonin in cancer treatment will be further discussed.

**Recent Progress**

*Findings that support melatonin as a suppressor of autophagy*

Two recent studies that examine the effects of melatonin on CsA-induced autophagy in rat pituitary GH3 cells and NO-induced autophagy in mice myoblast cells found that melatonin decreased cell death caused by autophagy [1, 2]. In the study of CsA-induced autophagy it was determined that CsA toxicity causes Endoplasmic Reticulum (ER) stress which further leads to cellular autophagy. The characteristic toxicity of CsA is most likely due to the increased free radical production from CsA. Melatonin's antioxidant powers were found to protect cells from CsA toxicity and were determined as a factor attenuating autophagy. Melatonin was also found to increase the antioxidant enzyme levels and stabilize intracellular calcium levels in the pituitary cells. Both of these factors are likely to be in direct opposition to the known toxicity of CsA. The study determined many novel effects of melatonin; it was found that melatonin prevents cardiotoxicity from CsA and has a shielding effect against renal malfunction during CsA produced nephrotoxicity [1]. In the study of mice myoblast cells, the same general effect of melatonin as an autophagy inhibitor was observed. Melatonin was found to protect against both apoptotic and autophagic cell death in myoblast cells. Melatonin lowered NO-induced apoptosis and serum deprivation induced autophagy [2].

In both studies, melatonin was identified as changing the expression levels of proteins concerned with apoptotic and autophagic signaling. Melatonin decreased expression of pro-apoptotic proteins while increasing anti-apoptotic protein expression. Reduced expressions of all autophagy-associated proteins (LC3-II, Beclin-1) was observed in cells treated with melatonin, which is highly suggestive of melatonin as a cell survival mechanism that protects against autophagy [1, 2]. In both studies, the Bcl-2 protein family was found as an important indicator of autophagosome levels. Bcl-xL acts as an antiapoptotic protein while Bcl-2 acts as an anti-autophagic protein. Bcl-2 concentration was found to increase with increases in melatonin levels [2].

*Findings that support melatonin as an inducer of autophagy*

In a study involving mouse hepatoma tumor cells, it was found that melatonin can inhibit tumor growth similarly to its inhibition of CsA induced toxicity. In contrast to the other two studies examined in this review, this study identified melatonin as triggering autophagy in H22 cancer cell bearing mice. The inhibition of tumor growth was likely a direct result of melatonin derived autophagy of tumor cells. In this way, melatonin is likely to activate a protective mechanism through autophagy that prevents further tumor growth. Circadian rhythm control that is associated with melatonin levels was also given as a possible explanation for the inhibition of tumor growth that occurs after melatonin treatment in H22-bearing mice. Unlike the previous two studies examined, expression of the autophagy-associated proteins LC3-II and Beclin-1 was increased after treatment of melatonin on H22-bearing mice [3].

**Discussion**

A question arising from these studies is whether or not melatonin induces autophagy. The results of the studies noted in this review give conflicting evidence. The studies involving pituitary and myoblast cells indicate that melatonin reduces autophagy [1, 2], while melatonin induces autophagy in H22 tumor cells. These results indicate that melatonin may have different mechanisms dependent upon the type of cells it is acting upon. In the H22 cells, melatonin was found to specifically activate a protective autophagic response that could prevent these cancerous cells from dying. Inhibition of this protective autophagic response resulted in enhancement of the anti-tumor growth properties of melatonin. It was also found that the combination of therapeutic drugs with autophagy inhibitors could potentially synergistically inhibit the growth of tumors [3]. These results seem contradictory and require further study of melatonin in cancer progression. Future research could reveal important uses of melatonin as an anti-cancer agent.

Another issue arising from these studies is the role of autophagy as both a type of programmed cell death and as a cellular survival mechanism. Results from these studies would indicate that autophagy is of particular use as a cellular survival mechanism, as in its role as a tumor suppressor. Autophagy appears to act as a type of

 

programmed cell death specifically when other methods of programmed cell death fail [1, 2, 3]. There is generally a poor knowledge of apoptotic and autophagic protein activation mechanisms and the role that each plays relative to the other. It is possible that a concerted relationship exists between apoptotic and autophagic processes [3]. An important area of future research could be related to the relationship between apoptotic and autophagic cell mechanisms.

**References**

[1] Yoo Y, Jeung E. Melatonin suppresses cyclosporine A-induced autophagy in rat pituitary GH3 cells. Journal of Pineal Research 2010; 48: 204-211

[2] Kim C, Kim K, Yoo Y. Melatonin protects against apoptotic and autophagic cell death in C2C12 murine myoblast cells. Journal of Pineal Research 2011; 50: 241-249.

[3] Liu C, Jia Z, Zhang X, Hou J, Wang L, Hao S, Ruan X, Yu Z, Zheng Y. Involvement of melatonin in autophagy-mediated mouse hepatoma H22 cell survival. International Immunopharmacology 2012; 12: 394-401.

 