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THE EFFECT OF EXOGENOUS MELATONIN ON THE CONCENTRATION OF CALCIUM IONS IN MICE BLOOD SERUM *(SHORT COMMUNICATION)*

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The aim of the study was to measure calcium ions Ca^{2+} concentration in the serum of mice, who were intraperitoneally treated with melatonin at a dose of 5 mg/kg, 10 mg/kg and 15 mg/kg b.w.
Twenty four hours after melatonin injection, blood serum Ca^{2+} concentration was significantly decreased.

Key words: exogenous melatonin, blood serum, calcium ions Ca^{2+} mice

INTRODUCTION

The concentration of calcium ions in intracellular fluid and serum is mainly regulated by parathyroid hormone, vitamin D and calcitonin (DELUCA, 1985, WASSERMAN and FULLMER, 1981, SCHACHTER and KOWARSKI, 1985).

Calcium ions are the most universal regulator of cellular processes. They are second order transducers and regulate the levels of cyclic nucleotides, cause muscle and muscle tissue contraction, releases neurotransmitters, are involved in mitotic processes in intercellular communication and proteolysis reactions (BLAUSTEIN, 1985, CARAFOLI, 1982, KUŹNICKI and DRABIKOWSKI, 1980, GRABAREK and KUŹNICKI, 1980, VAN ELDIK et al., 1982).

Melatonin (N-acetyl-5-methoxytryptamine) arouses great interest among researchers because it has many different activities. This hormone exerts its biological effects by activating specific receptors. This process takes place through: binding of the hormone by membrane receptors ML_1 and ML_2; interaction with nuclear receptors RZalpha and RZbeta – activation or transcription of certain genes; direct interaction with cytoplasmic proteins calmodulin (CaM) – influence on biochemical calcium-dependent processes (ZAWILSKA and NOWAK, 1997).

Furthermore, all ML_1 subtypes have also been implicated in the Ca^{2+} dependent release of dopamine in the mammalian retina, as well as various light-dependent processes such as phagocytosis

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of retinal photopigment discs (Brzeziński, 1997, Reppert et al., 1996). ML_1 receptor activation results in the inhibition of adenylyl cyclase, and consequently decreas in 3', 5'-cAMP levels, via its coupling to a G_1-protein (Brzeziński, 1997, Reppert et al., 1996, Dubocovich, 1995). Moreover, only Mel_{1a} receptor has been shown to induce a parallel signal-transduction of phospholipase activation (Godson et al., 1997). In addition, the Mel_{1c} receptor has been shown to exert a modulatory influence on intracellular concentrations of 3', 5' – cGMP levels (Jokers et al., 1997), the only melatonin receptor substitute known to affect this particular cyclic nucleotide.

Calcium ions play active role in melatonin biosynthesis and sodium and potassium ions, by involvement in the membranes polarization, influence melatonin penetrating possibility into cells, where this active molecule can exert its physiological functions (Boguszewska and Pasternak, 2004).

Hence, it seems interesting to measure exogenous melatonin influence on the concentration of calcium in mice blood serum.

MATERIALS AND METHODS

The experiment was conducted on 24 male mice of Swiss strain, with an average weight of 25g, kept in constant conditions (LD 12:12) and fed standard food with free access to water.

Animals were divided into one control group and three experimental animal groups (n=6). Control mice received intraperitoneal injection of saline in a volume of 0.3 ml, while the experimental animals were injected with melatonin. Animals of the first experimental group received a single dose of melatonin at 5 mg / kg b.w., the second 10 mg / kg b.w. and the third 15 mg / kg b.w. Twenty four hours after injection, blood samples were collected and concentrations of calcium ions (Ca^{2+}) in serum were measured by colorimetric method (Biochemtest, Poland).

RESULTS

The serum level of calcium in blood of control mice was 2.22 ±0.006 µmol/l (Fig. 1). Injection of 5 mg/kg b.w. melatonin significantly decreased the level of calcium ($p<0.05$). Higher doses (10 and 15 µg/kg b.w.) of exogenous melatonin decreased the calcium level to 1.89±0.02 and 1.71±0.03 µmol/l, respectively ($p<0.01$)

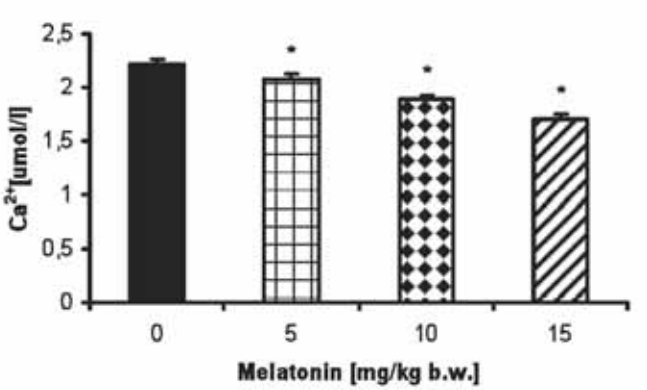

Fig. 1. The effect of melatonin on serum calcium level (X±SE, *p<0.05-0.01)

DISCUSSION

The obtained results clearly showed that pharmacological doses of melatonin decreased serum calcium concentration in mice.

Previous study showed that melatonin is able to penetrate into the cells (Boguszewska and Pasternak, 2004). Kumari et al. (2011) showed melatonin induced rise in intra cellular calcium in human platelets. Futhermore, higher levels of melatonin potentiated the effect of thrombin in raising platelet calcium. It has been also shown that exogenous melatonin increases calcium in pancreatic cell lines probably due to facilitation of release of digestive enzymes downstream of physiological secretagogues (Bach, et al 2005). It also known that melatonin interacts with intracellular proteins – calmodulin, calreticulin or tubulin and antagonizes the binding of calcium to calmodulin (Pandi et al. 2008).

The results suggest that lower level of calcium in serum was due to its penetration into the different cells. It seems probable that melatonin at the higher doses may regulate physiological processes by affecting calcium extracellular levels.

REFERENCES

Bach, A.G., S. Wolgast, E. Mühlbauer. and E. Peschke. 2005. Melatonin stimulates inositol 1,4,5-triphosphate and calcium release from INS1 insulinoma cells. *J. Pineal Res.* 39, 316-323.

Blaustein, M. P. 1985. Intracellular calcium as a secon messenger: What's so special about calcium? W: Calcium in Biological Systems, Rubin R. et al. (red.), *Plenum Press*, New York, London, 23-33.

Boguszewska, A. and K. Pasternak. 2004. Melatonin and bioelements. *Pol. Merkuriusz Lek.* Nov, 17(101), 528 – 529.

Brzeziński, A. 1997. Melatonin in humans. *N Engl. J Med.* 336, 186-195.

Carafoli, E. 1982. The regulation of intercellular calcium. Regulation of phosphate and mineral metabolism, Massry i wsp. (red.), *Plenum Publishing Corporation*, 461-472.

DeLuca, M. F. 1985. The vitamin D – calcium axis – 1983. Calcium in Biological Systems, Rubin i wsp. (red.), *Plenum Press*, New York, London, 491-511.

Dubocovich, M. L. 1995. Melatonin receptors: Are there multiple substitutes? *Trends Pharmacol. Sci.*15, 50-56.

Godson, C. and S.M. Reppert. 1997. The Mel1a melatonin receptor coupled to paralel Signac transduction pathways. *Endocrinology* 138, 397-404.

Grabarek, Z. and J. Kuźnicki. 1980. Homologiczne białka wiążące wapń. *Post. Biochem.* 26, 209-224.

Jockers, R., L. Petit, I. Lacroix, P. de Coppet, P. Barrett, P.J. Morgan, B. Guardiola, P. Delagrange, S. Marullo and A.C. Strosberg. 1997. Novel isoformsof Mel1c melatonin receptorsmodulating intracellular cyclic guanosine 3' 5' – monophosphate levels. *Mol. Endocrinol.*, 11. 1070-1081.

Kumari, S. and D. Dush. 2011. Melatonin elevates intracellular free calcium in human platelets by inositol 1,4,5-triphosphate independent mechanism. *Febs Lett.*; 585(14):2345-51.

Kuźnicki, J. and W. Drabikowski. 1980. Kalmodulina-aktywator procesów regulowanych przez jony wapnia. *Post. Biochem.*, 26, 265-290.

Pandi Perumal, S.R., I. Trakht, V. Srinivasan, D.W. Spence, G.J. Maestroni, N. Zisapel and D.P. Cardinali. 2008. Physiological effects of melatonin: Role of melatonin receptors and signal transduction pathways. *Prog. Neurobiol.* 85, 335-353.

Reppert, S. M., D.R. Weaver and C. Godson. 1996. Melatonin receptor step into the light: Cloning and classification of substytutes. *Trends Pharmacol. Sci.*, 17, 100-102.

Schachter, D. and S. Kowarski. 1985. Vitamin D and the intestinal calcium-binding protein. W: Calcium in Biological Systems. Rubin i wsp. (red.), *Plenum Press*, New York, London, 513-518.

Sorenson, J. R. 2002. Cu, Fe, Mn and Zn chelates offer a medicinal chemistry approach to overcoming radiation injury. *Curr. Med. Chem.* Mar. 9(6), 639-662.

Van Eldik, L.J., J.G. Zendegui, D.R. Marshak and D.M. Watterson. 1992. Calcium binding proteins and the molecular basis of calcium action. *Inter. Rev. Cyt.* 77, 1-61.

Wasserman, R. H. and C.S. Fullmer. 1981. Calcium transport proteins, calcium absorption and vitamin D. *Ann. Rev. Physiol.*, 43, 369-375.

Zawilska, J. B. and J.Z. Nowak. 1997. Receptory melatoninowe. *Wydawnictwo Naukowe PWN.* Warszawa.