Neuroendocrinology Letters ISSN 0172–780X

ORIGINAL ARTICLE

Circadian rhythm of melatonin in patients with colorectal carcinoma

Beata Kos-Kudla[1], Zofia Ostrowska[2], Andrzej Kozlowski[3], Bogdan Marek[1], Nelly Ciesielska-Kopacz[4], Marek Kudla[5], Dariusz Kajdaniuk[1], Janusz Strzelczyk[1] & Pawel Staszewicz[1]

1. Department of Pathophysiology and Endocrinology,
2. Department of Clinical Biochemistry,
3. 2nd Department of General and Vascular Surgery,
4. Department and Clinic of Internal and Allergic Diseases, Silesian Medical University, Zabrze, Poland
5. III Dept.& Clinic of Obstetrics and Gynaecology, Silesian Medical University, Katowice, Poland

Correspondence to: Beata Kos-Kudla, M.D., Ph.D.,
Department of Pathophysiology and Endocrinology
Silesian Medical University
Pl. Traugutta 2, 41-800 Zabrze, POLAND
PHONE/FAX: +48 32 2786126
E-MAIL: beatakos@ka.onet.pl

Submitted: *January 4, 2002*
Accepted: *March 6, 2002*

Key words: **melatonin; circadian rhythm; colorectal carcinoma**

*Neuroendocrinology Letters 2002; **23**:239–242 pii: NEL230302A06*

Abstract

OBJECTIVE: The aim of the study was pineal gland function assessment on the base of daily rhythm study and mean daily melatonin (MEL) concentrations in serum in patients with colorectal carcinoma.

MATERIAL AND METHODS: Studies were performed in 12 women at the age of 63.17±5.90 years and 21 men aged 58.95±11.32 years with large intestine adenocarcinoma. The control group consisted of 28 healthy volunteers at comparable age. During the circadian study blood samples for the measurement of melatonin (MEL) were collected every 4 hours during 12 h. MEL concentrations were assessed with the use of RIA methods. Statistical analysis of circadian rhythms of MEL was carried out with the use of cosinor method according to Halberg.

RESULTS: Existence of daily rhythm of MEL secretion was shown in all studied groups. A significant decrease of amplitude of rhythm and secretion of MEL at nocturnal hours in comparison with the control group was shown in the group of women with large intestine carcinoma. A significant decrease of mesor value and amplitude of MEL rhythm as a consequence of decrease of MEL secretion at nocturnal and morning hours was observed in the group of ill men.

CONCLUSIONS: Decrease in melatonin circadian rhythm amplitude as a consequence of its lowered nocturnal secretion occurred in all patients with colorectal carcinoma. Abnormalities in daily rhythm of melatonin secretion were more intensified in men with large intestine carcinoma, which leads to suppression of mean daily hormone concentration.

Introduction

Recent studies, both experimental and clinical indicate connections between the pineal gland and neoplastic growth. The pineal gland is called an oncostatic organ. The presence of melatonin (MEL) receptors in different types of malignant tumors has been shown, among others in patients with large intestine carcinoma [1,2,3]. It was suggested that melatonin may influence the human colonic functions through interaction with its receptors in the mucosa [3].

The mechanism of oncostatic action of MEL is still not precisely explained. MEL may act through the endocrine system, immune system, regulating interaction of the pineal gland and opioid system, and exerting direct antiproliferative action [5]. Cos and Blask [4] hypothesized that melatonin can inhibit action and/or release of growth factors stimulating development and/or growth of tumor cells. It was shown *in vitro* that MEL inhibits tumor growth stimulated by epidermal growth factors (EGF) and insulin-like growth factor (IGF-I). IGF-I takes part in the promotion of normal and neoplastic cells growth. Most probably it also plays a role in processes of neoplastic transformation, angiogenesis, and progression of neoplasm, in this in metastases forming [4]. The mechanism of oncostatic action of MEL can also be connected with antioxidant and scavenging free radicals properties of MEL. Free radicals are considered an essential factor in pathogenesis of neoplastic disease. It was shown that MEL is a scavenger of hydroxyl and peroxyl radicals [6–7].

The mechanism of oncostatic action of MEL may also be connected with immunopotentiating action of the hormone [8]. MEL stimulates activated lymphocytes T to synthesis and release of endogenic opioids, interleukin –2 and γ- interferone [9]. Lissoni et al. [10] have observed that MEL administration causes inhibition of tumor development in patients with neoplasm of large intestine, liver, stomach, pancreas and lungs with simultaneous increase of the relation of lymphocytes T4 to T8.

MEL exerts an inhibiting influence on development and/or growth of many types of tumors induced experimentally or transplanted in animals [5,11]. There are only a few studies concerning the assessment of MEL concentrations in different types of malignant tumors in humans [12–17].

Trials to use therapy with MEL in advanced neoplastic disease were also undertaken, obtaining encouraging effects. There is a suggestion that MEL, except oncostatic action, can be useful in the improvement of the quality of life in patients with incurable, advanced malignant tumors [10,18–20].

The aim of the study was estimation of circadian rhythm and mean daily serum MEL concentrations in patients with colorectal carcinoma.

Material and methods

Studies were performed in 12 women aged 63.2±5.9 years and 21 men aged 59.0±11.3 years with colorectal carcinoma (histopatologically: adenocarcinoma; degree of neoplasm differentiation: G II–III, degree of progression according to Dukes: C 1–2). The control group consisted of 28 healthy volunteers: 13 women aged 59.5±9.3 years and 15 men aged 60.1±8.1 years.

The study was conducted with the permission of the Ethics Committee at the Silesian Medical University in Katowice, and all patients gave their written consent to participate in the study.

All subjects were hospitalised during the study and their daily activities, meal times and nocturnal rest were synchronized.

During the circadian study blood samples for the measurement of MEL were collected every 4 hours during 24 h beginning at 06 h. The studies were performed before operation. During the dark period a weak (25 W) red light was lit immediately prior to blood sampling and was switched off within 2 minutes. Serum samples were obtained with the use of a centrifuge (450 g, 10 min.) and stored frozen (–75°C) until assays.

MEL concentrations were measured with the use of commercially available RIA kits from DRG Instruments GmbH (USA). Sensitivity of the assay as well as intra- and interassay coefficients of variations were 1 pg/sample, 8.6%, and 9.2%, respectively.

The results were statistically analysed with the use of variance analysis for Kruskal-Wallis nonparametric tests. After rejecting the variance uniformity hypothesis, further analysis of statistical significance was conducted using the Mann-Whitney U test.

Statistical analysis of circadian rhythms of MEL was carried out with the use of the cosinor method according to Halberg et al. [21]. Cosinor analysis was carried out for a fixed average time group value by fitting the main cosinor function $f(T) = M + A \cos(t\omega + \varphi)$, where f (T) is the average hormone concentration at the given time point; M is the mesor, arithmetic average of actual values describing oscillations within the cycle; A is the amplitude, difference between maximum (or minimum) value and the sinusoidal average; φ is the acrophase, angle (360°=24h) corresponding to maximum value of a given hormone concentration within 24 hours; ω is angular frequency. The appearance of a rhythm was deducted following rejection of zero amplitude hypothesis.

Results

The existence of daily rhythm of MEL secretion was shown in all the studied groups .

Women with large intestine carcinoma showed a significant decrease of MEL concentration at night (at 18:00, 22:00, and 02:00 h) in comparison with values in the control group (Fig. 1). Analysis of chronobiological parameters of daily MEL rhythm showed a significant

decrease of amplitude in relation to the control group.

In the group of men with large intestine carcinoma a decreased MEL concentration at nocturnal and morning hours (at 18:00, 22:00, 02:00, and 06:00 h) in comparison to the control group was shown (Fig. 2). A significant decrease of mesor value and amplitude of MEL rhythm was also observed.

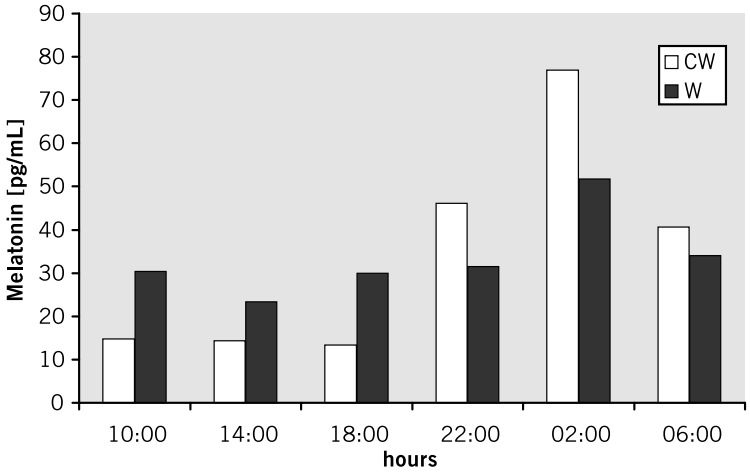

Figure 1. Mean melatonin circadian serum concentrations (MEL; pg/ml±SD) and chronobiological parameters of MEL rhythm in women with colorectal carcinoma (W) and in the control group (CW).

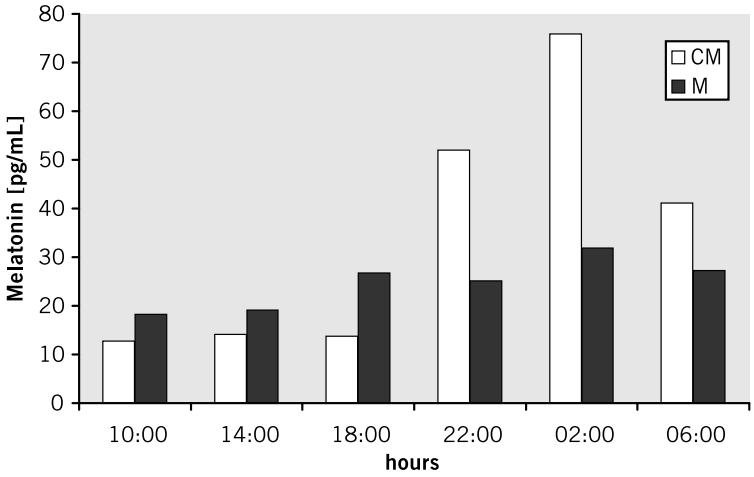

Figure 2. Mean melatonin circadian serum concentrations (MEL; pg/ml±SD) and chronobiological parameters of MEL rhythm in men with colorectal carcinoma (M) and in the control group (CM).

Discussion

Our studies have shown the existence of daily rhythm of MEL secretion both in women and men with colorectal carcinoma. Existence of daily rhythm of MEL secretion in patients with large intestine carcinoma was also confirmed by Vician's et al. [22] studies. However, in studies on an animal model – rats with experimentally induced large intestine tumors – existence of daily rhythm of MEL secretion was not shown [23].

There are a lot of data confirming pineal gland function disturbances in patients with neoplastic disease. A hypothesis was even put forward that a lack of sufficient synthesis and secretion of MEL may lead to development of some types of tumor [24,25].

Decreased, increased as well as normal MEL concentrations in patients with neoplastic disease in comparison with healthy people were described in the literature [see 26]. These differences can depend on different types of neoplasm, different degrees of the disease's progression, used drugs, the number, and different age of studied persons.

In the presented study we have observed disturbances of daily MEL concentrations in a form of decrease of its amplitude as a result of lowered secretion at nocturnal hours. Similar observations were made by Kvetnoy and Levin [27]. They studied diurnal excretion of MEL in 37 cases of gastric and rectal cancer. In cancer patients nocturnal excretion was lower than in daytime while in healthy subjects the situation was reversed [27]. Khoory and Stemme [16] have shown large individual variability in absolute MEL levels in patients with colorectal carcinoma. However, during the night, MEL concentration in cancer patients was significantly lower than in controls.

Lowered MEL concentrations are also observed in other types of malignant tumors, for example in patients with primary breast and prostate cancer. This decrease does not seem to depend on increased liver metabolism of MEL [28,29]. Similarly to our studies, in most patients with breast, prostate, colon or endometrial cancer, a decrease of nocturnal secretion of MEL was shown [13,14,16,17]. In patients with breast cancer blurring of nocturnal peaks of MEL secretion was connected with a decrease of the rhythm's amplitude of this hormone secretion by 65% [28–30]. It was also shown that intensification of MEL secretion disturbances in patients with cancer depends on the degree of progression of the disease – the more advanced degree of tumor development, the more suppressed the daily MEL rhythm [30]. A growing tumor can modulate MEL biosynthesis – in the early stage stimulates, and in advanced stage inhibits the secretion [29]. Lissoni et al. [31] also paid attention to changes of pineal gland function in patients with cancer. They have shown a decrease of MEL concentrations, among others in patients with large intestine cancer, which have changed after chemotherapy. They have even suggested the possibility of the use of MEL's assays for prognosing effects of chemotherapy. They showed that, irrespectively of the type of tumor and chemotherapeutic regimen, 75% of patients whose MEL markedly enhanced after chemotherapy had an objective regression. The percentage of objective responses was significantly higher in patients with a chemo-

therapy-induced MEL increase than in those with no MEL increase [31].

Contrary to our observations Vician et al. [22] showed that nocturnal MEL concentrations did not differ in a group of patients with cancer as compared to control group, and diurnal concentrations in cancer patients were even higher than those in the controls. Moreover, surgical treatment in patients with large intestine cancer caused a significant increase of MEL concentrations in serum both at night and during the day. MEL concentration during the day in tissues of the large intestine was over 10-times higher than in serum. On the base of these observations, authors [22] hypothesized that increased levels of MEL in the gastrointestinal tract may play an important protective role against the development of colorectal cancer via stimulation of the immune system.

Anisimov's et al. [23] studies can confirm these observations. They have shown in animals with large intestine colorectal cancer experimentally induced by 1,2-dimethylhydrazine an increase in MEL concentration in comparison to the control group.

The observation coming from our studies that changes in daily MEL secretion are more marked in men than in women with large intestine cancer is difficult to interpret.

More precise determination of the relation between pineal gland and neoplastic process in the large intestine needs further studies.

REFERENCES

1 Blask DE, Cos S, Hill SM, Burns DM, Lemus-Wilson A, Grosso DS. Melatonin action on oncogenesis. In: Fraschini F, Reiter RJ, editors. Role of melatonin and pineal peptides in neuroimmunomodulation. New York: Plenum Press; 1991. p. 233–40.

2 Stankov B, Lucici V, Scaglione F, Cozzi B, Righl M, Canti G, et al. 2-[^{125}]Iodomelatonin binding sites in normal and neoplastic tissues. In: Fraschini F, Reiter RJ, editors. Role of melatonin and pineal peptides in neuroimmunomodulation. New York: Plenum Press; 1991. p. 117–25.

3 Poon AM, Mak AS, Luk HT. Melatonin and 2[125I]iodomelatonin binding sites in the human colon. Endocr Res 1996; **22**:77–94.

4 Cos S, Blask DE. Melatonin modulates growth factor activity in MCF – 7 human breast cancer cells. J Pineal Res 1994; **17**:25–32.

5 Pawlikowski M, Winczyk K, Karasek M. Oncostatic action of melatonin: facts and question marks. Neuroendocrinol Lett 2002; **23**(suppl 1):24–29.

6 Pierrefiche G, Laborit H. Oxygen free radicals, melatonin and aging. Exp Gerontol 1995; **30**:213–27.

7 Reiter RJ, Tan DX, Allegra M. Melatonin: reducing molecular pathology and dysfunction due to free radicals and associated reactans. Neuroendocrinol Lett 2002; **23**(suppl 1):3–8.

8 Maestroni GJM, Hertens E, Galli P. Mechanisms of action of melatonin on the human immune system. Front Horm Res 1997; **23**:62–71.

9 Maestroni GJM: The immunoendocrine role of melatonin. J Pineal Res 1993; **14**:1–10.

10 Lissoni P, Barni S, Crisoino S, Tancini G, Fraschini F. Endocrine and immune effects of melatonin therapy in metastatic cancer patients. Eur J Cancer Clin Oncol 1989; **25**:789–95.

11 Karasek M. Relationship between the pineal gland and experimentally induced malignant tumors. Front Horm Res 1997; **23**:99–106.

12 Tarquini B, Cornelissen G, Tarquini R, Perfetto F, Halberg F. General and unspecific damping by malignancy of the circadian amplitude of circulating human melatonin? Neuroendocrinol Lett 1999; **20**:25–8.

13 Bartsch C, Bartsch H, Fuchs U, Lippert TH, Bellman O, Gupta D. Stage-dependent depression of melatonin in humans with primary breast cancer. Cancer 1989; **64**:426–33.

14 Bartsch C, Bartsch H, Schmidt A, Ilg G, Bichler KH, Fluchter SH. Melatonin and 6-sulfatoxymelatonin circadian rhythms in serum and urine of primary prostate cancer patients: evidence for reduced pineal activity and relevance of urinary determinations. Clin Chim Acta 1992; **209**:153–67.

15 Lissoni P, Rovelli F, Malugani F, Bucovec R, Conti A, Maestroni GJ. Anti-angiogenic activity of melatonin in advanced cancer patients. Neuroendocrinol Lett 2001; **22**:45–7.

16 Khoory R, Stemme D. Plasma melatonin levels in patients suffering from colorectal carcinoma. J Pineal Res 1988; **5**:251–8.

17 Karasek M, Kowalski AJ, Zylinska K. Serum melatonin circadian profile in women suffering from the genital tract cancer. Neuroendocrinol Lett 2000; **21**:109–13.

18 Lissoni P, Barni S, Tancini G, Fraschini F. Clinical study of melatonin in untreatable advanced cancer patients. Tumori 1987; **73**:475–80.

19 Barni S, Lissoni P, Paolorossi F, Crispino S, Archieli C, Tancini C. A study of the pineal hormone melatonin as a second-line therapy in metastatic colorectal cancer resistant to fluorouracil plus folates. Tumori 1990; **76**:58–60.

20 Karasek M, Kuzdak K, Cywinski J, Zylinska K, Smialowska A, Pluzanska A. Effects of melatonin administration in advanced breast cancer patients – preliminary report. Neuroendocrinol Lett 1998; **19**:15–9.

21 Halberg F, Tong YI, Johnson EA. Circadian system phase as aspect of temporal morphology: procedure and illustrative examples. In: von Mayerbach H, editor. The cellular aspects of biorhythms. Berlin: Springer Verlag; 1967. p. 20–48.

22 Vician M, Zeman M, Herichova I, Jurani M, Blazicek P, Matis P. Melatonin content in plasma and large intestine of patients with colorectal carcinoma before and after surgery. J Pineal Res 1999; **27**:164–9.

23 Anisimov VN, Kvetnoy IM, Chumakova NK, Kvetnaya TV, Molotkov AO, Pogudina NA, et al. Melatonin and colon carcinogenesis. II. Intestinal melatonin-containing cells and serum melatonin level in rats with 1,2-dimethylhydrazine-induced colon tumors. Exp Toxicol Pathol 1999; **51**:47–52.

24 Hofbauer LC, Heufelder AE. Endocrinology meets immunology: T lymphocytes as novel targets for melatonin. Eur J Endocrinol 1996; **134**:424–5.

25 Weeb SM, Puig-Domingo M. Role of melatonin in health and disease. Clin Endocrinol 1995; **42**:221–34.

26 Karasek M. Malignant tumors and the pineal gland. In: Gupta D, Wollmann HA, Fedor-Freyberg PG, editors. Pathophysiology of immune-neuroendocrine communication circuit. Heidelberg: Mattes Verlag; 1994. p. 225–44.

27 Kvetnoy IM, Levin IM. Daily excretion of melatonin in patients with cancer of the stomach and large intestine. Vopr Onkol 1987; **33**:29–32.

28 Bartsch C, Bartsch H, Bellmann O, Lippert TH. Depression of serum melatonin with primary breast cancer is not due to an increased peripheral metabolism. Cancer 1991; **67**:1681–4.

29 Bartsch C, Bartsch H, Flochter SH, Mecke D, Lippert TH. Diminished pineal function coincides with disturbed circadian endocrine rhythmicity in untreated primary cancer patients. Ann N Y Acad Sci 1994; **719**:502–25.

30 Bartsch C, Bartsch H, Buchberger A, Rokos H, Mecke D, Lippert TH. Serial transplants of DMBA-induced mammary tumors in fisher rats as model system for human breast cancer. Oncology 1995; **52**:278–83.

31 Lissoni P, Tancini G, Barni S, Crispino S, Paolorossi F, Rovelli F, et al. Melatonin increase as predictor for tumor objective response to chemotherapy in advanced cancer patients. Tumori 1988; **74**:339–45.