The NCI Office of Cancer Complementary and Alternative Medicine

Invited Speaker Series

Melatonin, Chronobiology, and Cancer

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Introduction

The National Cancer Institute's Office of cancer Complementary and Alternative Medicine (OCCAM) hosted Melatonin, Chronobiology and Cancer, the second in its Invited Speakers Series, on February 28, 2003, in Lipsett Auditorium at the National Institutes of Health. A panel of international experts on chronobiology and the use of melatonin in cancer treatment presented the results from their research and clinical practice. The 3 hour seminar was videocast over the Internet and is available for viewing on the NIH Videocasting website and can be accessed through the OCCAM website at http://www3.cancer.gov/occam/conferences.html.

This topic was chosen for several reasons. First, it relates to the activities of the OCCAM which involve both complementary and alternative medicine (CAM) as well as certain aspects of cancer research which would not directly fall under a consensus definition of CAM such as the effect of human homeostasis mechanisms on factors involved in cancer prevention and treatment.

The hormone melatonin, is found in a great variety of organisms. The plant derived form, phytomelatonin, is sold as a dietary supplement in the United States and is used by some CAM practitioners in the treatment of cancer patients.

Though there is great excitement in oncology over the promise of new drugs directed a specific molecular targets, much work is also being done to investigate new ways to use old drugs or interventions. Stem cell or growth factor support and other approaches to cytoprotection (e.g. amifostine, mesna) permit the increase of chemotherapy doses (i.e. dose-intensity). Another strategy, also facilitated by the use of hematopoietic cytokines, is the shortening of chemotherapy cycles (i.e. increased dose-density) thus decreasing the tumor's ability to grow between cycles. This has recently shown promise in the adjuvant therapy of breast cancer.

Yet another approach to improving the effect of conventional cancer therapies is the use of chemotherapy or radiation modulation. Radiosensitizers and radioprotectants, such as amifostine, and biochemical modulators (e.g. folinic acid with 5-FU) increase the

therapeutic ratio of radiation or chemotherapy, again allowing higher doses or more frequent treatment, potentially increasing the anticancer effect without increasing the damage to normal tissues.

Hormones also have been used to modulate the effect of chemotherapy. Estrogen-induced synchronization strategies were conceived in the 1980's and eventually taken into randomized clinical trials with some evidence of increased efficacy. Work continued into the 1990's but the approach has not been broadly adopted in clinical practice. Other hormones, such as growth hormone, have also been examined for their effects on cell cycle and potential for combined effects with chemotherapy.

The research findings summarized in these talks indicate that both melatonin and chronotherapy or circadian-timed chemotherapy address all of these issues in cancer therapy. Information is also reviewed regarding the potential direct anticancer activity of melatonin. These approaches also apply to areas beyond cancer therapeutics, including cancer prevention and symptom and side effect management.

Both melatonin and chronotherapy have been studied for many years but, despite largely positive findings, have not been brought into mainstream cancer therapy. We hope these presentations will contribute to re-invigorating activities focused on the examination of these and related approaches to cancer management.

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Successful Cancer Therapy Development: Beating the Odds by Respecting Human Circadian Organization

William J. M. Hrushesky, M.D., director, Research Service Line, and VA Research and Development, Columbia, S.C.

Summary

Circadian rhythms traverse all realms of biological organization. Circadian organization of the host-cancer balance is important to cancer prevention, screening, diagnosis, and treatment. Melatonin, a hormone produced by the pineal gland, is a primary tuner, synchronizer, and chronobiotic that helps gate normal and cancer cells on a circadian basis.

Chronobiotics, synchronization, and tuning are different words for the ability to sharpen melatonin's amplitude and phase relationships through appropriately timed circadian interventions. This ability makes melatonin useful as a therapy for cancer prevention and treatment. Chronobiology and chronotherapy use timing to take advantage of robust endogenous synchronization.

Cellular proliferation and apoptosis, effects of hormones like melatonin, immune functions, and all other biological events are organized within circadian time. Selectivity that results from the timing of stimulatory, suppressive, cytotoxic, or anti-metabolite therapy can affect the efficacy and side effects of chemotherapy and all other therapies.

Clinical trials in patients with widespread ovarian cancer benefited from optimal circadian doxorubicin/cisplatin scheduling by allowing safer administration of higher doses of each drug and a 4-fold, 5-year survival advantage.

Randomized clinical studies of fluoropyrimidine anti-metabolites (FUDR, 5FU) showed that a circadian dosing schedule was superior to standard flat-rate continuous infusion, allowing nearly 50% more drug to be given with substantially less toxicity than lower doses given by constant-rate infusion. Recent French multicenter studies doubled the objective-

response frequency of a 5FU leukovorin-oxaliplatin combination by optimally timing the dosing schedules of these agents within the day. Circadian 5FU studies showed rhythmic circadian 5FU concentrations in the serum of patients receiving a constant-rate intravenous 5FU continuous infusion. A new 3-drug fluoropyrimidine-based chronotherapy for colorectal cancer markedly diminished all drug toxicities and doubled objective cancer response frequency.

Optimal circadian timing of growth factors (granulocyte colony stimulating factor, erythropoietin) is responsible for a 1.0–1.5 order-of-magnitude improvement in therapeutic activity. Circadian timing of short-half-life biological response modifiers such as the interferons, anticancer cytokines, and tumor necrosis factor can diminish or enhance tumor growth.

A group of circadian-clock control genes that time events in the central circadian clock (suprachiasmatic nucleus) and every cell in the body were recently discovered and cloned. In mouse studies, mutations in one of these genes, mouse period 2, result in the spontaneous development of cancer.

Such clinical and preclinical observations indicate that cancers maintain a circadian dialog with their host locations; clock-control genes time the circadian organization of normal and cancer cell function, proliferation, and apoptosis; and all molecular cancer-control strategies may be useful at certain times of day or useless or damaging at other times of day.

Melatonin: An Integrative Chronobiotic Anticancer Therapy Whose Time has Come

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Summary

All living organisms are organized in biological time, and melatonin, a hormone secreted mainly by the pineal gland, is an important part of this scheme. In the human light-dark cycle, melatonin peaks every 24 hours, between 2 a.m. and 3 a.m.

The light-dark cycle helps synchronize the activity of the central biological clock—a group of cells in the hypothalamus called the suprachiasmatic nucleus (SCN)—which drives the rhythm of melatonin production and secretion. Melatonin duration is regulated by the length of the night, so the SCN-pineal gland also acts as a calendar.

Melatonin is involved in circadian rhythm regulation, sleep, hormonal expression of darkness, seasonal reproduction, retinal physiology, antioxidant free-radical scavenging, cardiovascular regulation, immune activity, cancer control, and lipid and glucose metabolism. It is also a new member of an expanding group of regulatory factors that control cell proliferation and loss and is the only known chronobiotic hormonal regulator of neoplastic cell growth.

At physiological concentrations, melatonin suppresses cell growth and multiplication and inhibits cancer cell proliferation *in vitro* through specific cell-cycle effects. At pharmacological concentrations, melatonin suppresses cancer cell growth and multiplication. At physiological and pharmacological concentrations, melatonin acts as a differentiating agent in some cancer cells and lowers their invasive and metastatic status by altering adhesion molecules and maintaining gap-junction intercellular communication. In other cancer cell types, melatonin, alone or with other agents, induces programmed cell death.

Biochemical and molecular mechanisms of melatonin's oncostatic action include regulation of estrogen receptor expression and

transactivation, calcium/calmodulin activity, protein kinase C activity, cell structure and function, intracellular oxidation-reduction status, melatonin-receptor-mediated signal transduction cascades, and fatty acid transport and metabolism.

One of the main ways melatonin inhibits tumor growth at certain stages in the circadian cycle is by suppressing the activity of epidermal growth factor receptor (EGFR)/mitogen-activated protein kinase (MAPK). This effect occurs via melatonin-receptor-mediated blockade of tumor linoleic acid uptake and its conversion to 13-hydroxyoctadecadienoic acid (13-HODE), which normally activates EGFR/MAPK mitogenic signaling.

This is a potentially unifying model for melatonin's chronobiological inhibitory regulation of cancer growth in maintaining host-cancer balance. It is also the first biological explanation of how melatonin enhances the efficacy and reduces the toxicity of chemotherapy and radiotherapy in cancer patients.

Melatonin, Chronobiology and Cancer: Clinical Experience with Melatonin Alone or Combined with Standard Anticancer Therapies in Medical Oncology

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Summary

After more than 20 years of investigation into the pineal gland's role in cancer and clinical studies of its hormone, melatonin, in treating human neoplasms, medical oncology has begun to accept the fact that the pineal gland modulates biological characteristics of tumor cells and the psychoneuroimmune status of cancer patients.

Several studies have demonstrated that progressive decline in pineal function is the most frequent endocrine alteration in cancer patients. Given the well-documented physiological anticancer activity of melatonin and other pineal indoles, such an effect could play a role in cancer prognosis.

The 15-year clinical history of melatonin therapy of human neoplasms includes palliative therapy of untreatable solid neoplasms; cancer neuroimmunotherapy with melatonin plus anticancer cytokines (IL-2) to enhance efficacy in tumors resistant to IL-2 alone; chemoneuroendocrine therapy with melatonin plus chemotherapy, and radioneuroendocrine therapy with melatonin plus radiotherapy to reduce chemotherapy toxicity and enhance therapeutic efficacy; and neuroendocrine therapy with melatonin plus classical cancer endocrine therapies to enhance tumor endocrine dependence.

In all studies, melatonin was given orally once a day in the dark phase of the photoperiod at doses of 20–40 mg/day. In randomized studies of 1,440 patients with untreatable advanced solid-tumors who received melatonin or supportive care alone, melatonin prolonged survival time and prevented neoplastic cachexia, even though objective tumor regression was seen in only 2% of patients.

Cancer neuroimmunotherapy produced more promising tumor regression results. Melatonin-T plus subcutaneous low-dose IL-2 (3 milli

international units/day) showed a tumor regression rate of 16% in 400 patients with untreatable advanced solid tumors, with particular efficacy in hepatocarcinoma, gastrointestinal tumors, non-small-cell lung cancer, and mesothelioma.

In a randomized study of 450 advanced cancer patients with poor clinical status or chemotherapy-resistant tumors, melatonin seemed to enhance tumor response rate, prolong survival time, and prevent chemotherapy-induced thrombocytopenia, cardiotoxicity, neurotoxicity, and asthenia. No protection was seen against anemia, leukopenia, or alopecia.

In association with tamoxifen, melatonin seemed to induce a response rate of 29% in 14 metastatic breast cancer patients progressing on tamoxifen alone and 12% in 25 patients with advanced neoplasms other than classical endocrine-dependent tumors.

Melatonin may therefore be successfully used in medical oncology alone or to biologically modulate conventional anticancer therapies, including chemotherapy, radiotherapy, immunotherapy, and endocrine therapy. Successive studies are needed to establish the efficacy of melatonin therapy in relation to melatonin receptor expression by cancer cells and endogenous melatonin production.

Using melatonin to treat human neoplasms could transform medical oncology in a biomodulatory way. Biomodulation—or biological response modification—extends biological therapy beyond conventional immunotherapy. It includes increasing host tolerability to cytotoxic treatment; modifying tumor cell membrane characteristics to alter their immunogenicity, metastatic propensity, or susceptibility to killing; and preventing or reversing transformation or promoting maturation of the primitive cancer cell.

Such a transformation could help re-establish the psychobiological unity of patients and reconstitute psychoendocrine and immunobiological responses against cancer growth.

Speaker Biographies and Abstracts

William J.M. Hrushesky, M.D.
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William J.M. Hrushesky is the WJB Dorn Department of Veterans Affairs Medical Center director of research, an associate director of the South Carolina Cancer, and a professor in the University of South Carolina School of Medicine Department of Developmental Biology and Anatomy and the Norman J. Arnold School of Public Health Department of Epidemiology and Biostatistics at Columbia, S.C.

Dr. Hrushesky received a bachelor's degree with high honors in philosophy from Syracuse University Honors College in 1969, and an M.D. from the University of Buffalo Medical School in 1973, where he did experimental cancer research at Roswell Park Cancer Institute. He has trained at the Johns Hopkins University, the National Cancer Institute, and the University of Minnesota, where he was a tenured faculty member in the Departments of Medicine, Laboratory Medicine, and Pathology and in the Pathobiology and Biomedical Engineering graduate programs.

He has been DVA Network Two and Stratton Department of Veterans Affairs Medical Center senior clinician investigator and professor of medicine at Albany Medical College, a faculty member in the Rensselaer Polytechnic Institute Department of Chemical Engineering, and the Albany College of Pharmacy Department of Pharmaceutics.

Dr. Hrushesky is a fellow of the American College of Physicians and a member of many scientific societies; a reviewer for many scientific and medical journals; an editorial board member of several scientific journals; and consultant to a variety of drug delivery, pharmaceutical, biotechnology, and venture capital firms and financial houses.

He has given more than 200 invited lectures, many of them international; published more than 500 scientific articles, chapters or abstracts; holds several patents; is the founder of two corporations; is editor of several books and monographs. He has been consultant to the FDA, the National Academy of Sciences Institute of Medicine, the Congressional Office of Technology Assessment, and the President's Cancer Panel in the areas of biological rhythms, women's health, and women and drug

development. He has received basic research grants from the Department of Veterans Affairs, the National Cancer Institute, the National Heart, Lung and Blood Institute, and various medical device and pharmaceutical firms.

Dr. Hrushesky is a founder of Medical Chronotherapeutics (timing medical treatments relative to endogenous human time structure). He is also a member of the Cosmos Club and his biography has been included in several editions of *Who's Who*. His research interests include the study of several important biological rhythms and how these rhythms interact, drug delivery systems and their temporal control, and solid tumor oncology.

Discoveries and ongoing projects include the invention, development and reduction to practice of a quick noninvasive urinary test to predict kidney transplant rejection; characterization of the sole useful murine kidney cancer model (RENCA); development of the only reproducibly effective chemotherapy program for widespread kidney cancer; discovery that the toxicities, pharmacokinetics, maximum safely achievable dose intensities, and efficacy of several cancer therapies differ depending on when in cancer patients' circadian cycles they are administered; completion of the first, pivotal trial of implantable programmable drug-delivery technology for cancer therapy; improving understanding of the biological consequences of physical changes induced in bioactive proteins by contact with biomedical device surfaces; a method and diagnostic device for noninvasively and precisely measuring aerobic cardiopulmonary performance and risk through compartmentalizing heart-rate variability; that the optimal menstrual cycle timing of breast cancer resection confers substantial potential survival advantage; that the timing of cytotoxic chemotherapy in the mammalian fertility cycle determines the likelihood of subsequent fertility maintenance and the bone-marrow damage resulting from that therapy; that uterine cervical epithelial abnormalities discovered by pap smear occur and resolve rhythmically throughout the year; and that the host-cancer-treatment balance is meaningfully modulated throughout each year.

Successful Cancer Therapy Development: Beating the Odds by Respecting Human Circadian Organization

William J.M. Hrushesky, M.D.

Abstract

Three decades ago it was reported that the arrangement within the day of 3 hourly doses of the antimetabolite cytosine arabinoside had a pronounced effect upon the survival probability of mice previously inoculated with L1210 leukemia cells.[1] About 20 years ago a series of studies reported that circadian timings of the xenobiotic doxorubicin and reactive metal compound cisplatin impacted all of the toxicities of each drug as well as the ability of each agent separately and together to control and cure a solid tumor in rats.[2] Subsequent clinical trials in patients with widespread ovarian cancer demonstrated benefit from optimal circadian doxorubicin/cisplatin scheduling by allowing safer administration of higher doses of each drug and a 4-fold 5-year survival advantage for women who received the circadian schedule predicted to be best by preceding rat studies.[3]

During the last decade, chronopharmacodynamic attention has been focused on the fluoropyrimidine antimetabolites fluorodeoxyuridine (FUDR) and 5-fluorouracil (5FU). Continuous intravenous infusions of FUDR that peaked at different times within the day were given to rats bearing a mammary adenocarcinoma and demonstrated lower toxicity and greater efficacy when most of the daily dose was infused in the 6 hours late in the animal's daily activity span and during the first half of its daily sleep span.[4]

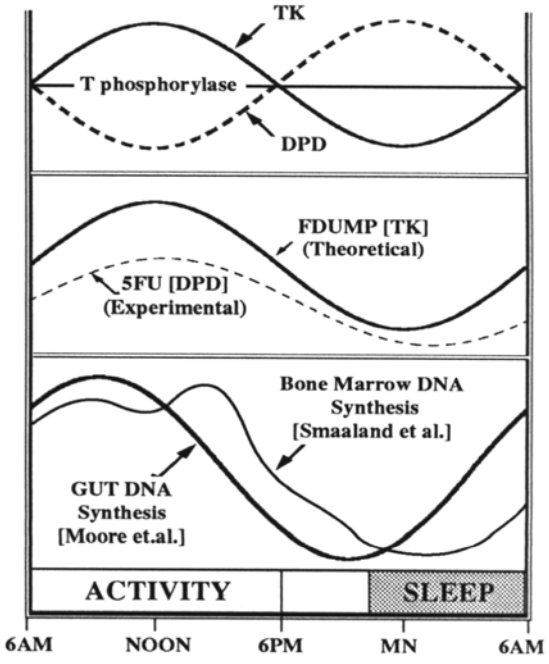
Randomized clinical studies demonstrated superiority of a similar circadian schedule to the standard flat-rate continuous infusion. The circadian schedule was far less toxic, allowing almost 50% more drug to be safely given with substantially less toxicity than lower doses given by constant-rate infusion.[5] This regimen was also found to have unexpected activity in renal cell carcinoma, a disease in which no other cytotoxic therapy has proven useful. Recent French multicenter studies doubled the objective response frequency of a combination of 5FU leukovorin and oxaliplatin by optimally timing these agents within the day.[6]

Circadian studies have also been carried out with 5FU. Based on early studies at the University of Minnesota that demonstrated the importance of dihydropyridinedehydrogenase (DPD) in the catabolism of 5FU and

studies from our lab that demonstrated prominent circadian time structure to DPD activity in lymphocytes from human volunteers, others have demonstrated rhythmic circadian 5FU concentrations in the serum of patients receiving a constant-rate intravenous 5FU continuous infusion. Serum levels of 5FU peak when (in the day) these patients' mononuclear cell DPD activities are lowest. Additional clinical studies have demonstrated up to several-fold safe increases in 5FU and 5FU leukovorin dose intensity by delivering most of each day's infusion during cancer patients' late activity and/or early sleep span.[7]

Human bone marrow cells and human gut epithelial cells synthesize DNA nonrandomly within the day. The most DNA synthesis occurs in both tissues between early morning and late afternoon. In addition to this coordinate DNA synthetic capacity, biochemical pathways responsible for fluoropyrimidine catabolism and anabolism are also coordinated in circadian timing within the liver and other cells, including toxicity targets such as bone marrow and gut.

During the past year, the circadian dynamics of many enzymatic pathways critical to understanding fluoropyrimidine chronopharmacodynamics have been elucidated and a coherent picture is emerging.[8]



The top panel of this figure illustrates experimentally determined circadian rhythmic patterns of the activities of three key enzymes in fluoropyrimidine metabolism. DPD catabolizes 5FU to noncytotoxic metabolites; thymidine phosphorylase (TP) converts 5FU to its nucleotide fluorodeoxyuridine (FUDR); thymidine kinase (TK) converts FUDR to fluordeoxyuridyl monophosphate FDUMP, which binds to thymidylate synthase (TS), blocks DNA synthesis by starving the cell for thymidine; DPD activity peaks about midnight; TP activity does not vary during the day; TK activity peaks around noon.

The middle panel demonstrates the result of these rhythmic enzyme activities on the pharmacokinetics of constant-rate 5FU infusion (5FU levels) and the final activation product FDUMP. DPD removes 5FU rhythmically during the day, making much more 5FU available in the early daytime hours. TP converts 5FU to FUDR at a constant rate but the higher morning substrate levels result in high morning FUDR levels. TK activity is higher in the morning, converting more of the higher FUDR levels to FDUMP at that time of day.

The bottom panel illustrates experimentally determined circadian patterns of gut and bone marrow DNA synthesis in human beings. Much more DNA synthesis occurs at the time of day associated with highest FDUMP levels. This coincidence of enzyme activities and gut and bone marrow DNA synthesis results in the experimentally observed high-amplitude reproducible circadian rhythm in myeloid and gastrointestinal susceptibility to 5FU. Giving a zero-order 5FU infusion thereby results in nonzero order and a reproducibly very unfavorable pharmacodynamic pattern from the perspective of toxicity to normal tissues. Furthermore, changing the circadian pattern of the 5FU infusion so most of the daily continuous infusion is given in the evening would give the resultant fluoropyrimidine pharmacokinetics and pharmacodynamics a more favorable toxic therapeutic profile.

A great deal of excellent work by EORTC and Dr. Lewis' group has resulted in the development of three-drug fluoropyrimidine-based chronotherapy for colorectal cancer.[9,10] This therapy was a chronotherapeutic continuous infusion of 5FU leukovorin and oxaliplatin to markedly diminish all drug toxicities and concurrently double objective cancer response frequency.[10]

In people with cancer, the circadian timing of the oxygen reactive, long-half-life xenobiotic doxorubicin and the highly nucleophylic metal-based cis diamminedichloroplatinum determine to a substantial extent the toxic therapeutic ratios of each agent, even though they each have multiple subcellular targets. Nonreactive short-half-life antimetabolites such as 5FU and FUDR, with more focused and fewer sites of action, demonstrate a greater circadian coordination of toxic therapeutic ratio.

Newer results with growth factors (GCSF and EPO) that have very short half lives and nearly unitary receptor-mediated activity targets demonstrate that the optimal circadian timing of these agents is reproducibly responsible for 1–1.5 orders of magnitude improvement in therapeutic activity. Finally, short-half-life biological response modifiers such as the interferons, IL-2, and tumor necrosis factor can reproducibly diminish or reproducibly enhance tumor growth, depending on circadian timing. The development of optimal temporal schedules relative to internal biological rhythms all agents capable of modulating the host-cancer balance is essential to more steadily improving our ability to control cancer.

We have further shown that spontaneous human cancer cell proliferation is organized within each day and that temporal organization faithfully reflects the temporal organization of the proliferation of that cancer's cells of origin.[11] This means that any molecular cancer treatment aimed at targets related to cell proliferation can be expected to exert its therapeutic effect to a reproducibly different extent at different times of day.[12]

A group of circadian-clock control genes that time events in the central circadian clock, the suprachiasmatic nucleus (SCN), and every cell in the body[11] have recently been discovered and cloned. Interestingly, one of these, mouse period 2 (m per2) has been shown to be essential in the temporal organization of p53-gated apoptosis and cyclin-controlled cell proliferation in the liver. Even more relevant—mutations of this clock gene results in the spontaneous development of cancer.[13]

The clinical and preclinical observations outlined above indicate that cancers of human beings and mice maintain a circadian dialogue with their host; that clock genes time the circadian organization of normal

and cancer cell function, proliferation, and apoptosis; and that all molecular strategies applied to control cancer may be useful at certain times of day and useless or damaging at other times of day.

All the data described above, with rare exceptions, have been uncovered by transverse observation of cancer growth, drug toxicity, and efficacy in groups of mice or people. The availability of an imaging system for small animals will allow us to extend observations of tumor growth, new blood vessel formation, and capillary permeability changes to individual mice over an entire circadian cycle. Such studies will allow us to test the efficacy of hitting specific molecular therapeutic targets at optimal times of day in individual tumor-bearing mice. Further exploration of functional MRI in the brains of these animals in the area of the circadian clock (SCN) will also allow us to observe central and peripheral coordination of clock function and cancer growth, new vessel function, and capillary permeability.

Finally, anticancer drug development is a complex, protracted, expensive, high-risk venture. Compounds are screened for cytotoxicity, analogues are developed, or agents are designed and synthesized to target certain critical cellular events or pathways. These compounds move through a "decision network" drug development process with many steps. At each step, most candidate agents are discarded for reasons related to toxicology, specificity or efficacy. The few agents that graduate from *in vitro* screening to whole animal systems have high subsequent failure probability as they move through *in vivo* toxicology trials in mouse, rat, dog, and primate, or during concurrent efficacy evaluation in human tumor cell lines and mouse tumor model systems.

If the target of drug action in host cells is virtually present or absent at specific times of the day, the results of screening studies will be divergent depending on when in the day they are performed. An example might be the development of a new S-phase active agent whose subcellular target is thymidylate synthase, the enzyme required for *de novo* synthesis of thymidine, which is necessary for DNA replication.

If the agent's toxicology is evaluated at the time of day associated with little gut or bone marrow DNA synthesis activity, the agent will have an excellent toxic-therapeutic ratio. In fact, the time of day that these studies are usually performed is in the first half of the working day. This

generally corresponds to the first half of the mouse's daily sleep span, when relatively low levels of DNA synthetic activity occur in gut and bone marrow.

Phase I clinical trials are also carried out in the first half of the working day, during the cancer patient's diurnal activity span, when human gut and bone marrow each exhibit the *highest level of daily DNA synthetic activity.* The resultant apparent poor therapeutic ration index might well cause this agent to be discarded as too toxic.

This scenario might have been different, however, if preclinical studies demonstrated the optimum circadian time for therapy and if subsequent phase I trials were performed at that time of day. Because efficacy is also nonlinearly related to treatment timing, the circadian timing of drug administration in phase II and phase III studies is likewise relevant to maximizing drug efficacy.

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David Blask is senior research scientist in the Laboratory of Chrononeuroendocrine Oncology at the Bassett Research Institute, Cooperstown, N.Y., and research scientist in the Department of Medicine, Columbia University College of Physicians and Surgeons, New York, N.Y. He received a B.S. in biology at Utica College of Syracuse University in 1969, a Ph.D. in neuroendocrinology in 1974, and an M.D. in1978 at the University of Texas Health Sciences Center, San Antonio, TX.

He was also a postdoctoral fellow in neuroendocrinology from 1974 to 1978 while in medical school, and held the rank of instructor of anatomy. From 1978 to 1989 he rose from the rank of assistant professor to full professor of anatomy at the University of Arizona Health Sciences Center in Tucson, AZ. He has been with the Bassett Research Institute since 1991.

Dr. Blask's research focus is the chrononeuroendocrine regulation of cancer development and growth by the pineal gland hormone melatonin and its role in the host-cancer balance. His research team is dedicated to discovering and understanding the mechanisms—from cellular/molecular to organismal levels and in the context of biological timing—by which melatonin exerts its actions on cancer and adipose tissue cachexia. Dr. Blask's team is seeking a better understanding of the circadian pathobiology of cancer and cancer cachexia in relation to the host-cancer balance, and to translate that information into clinical trials of melatonin as a chronobiotic preventive and therapeutic agent.

He has authored or coauthored more than 250 research articles, book chapters, and abstracts and has delivered numerous invited lectures and seminars nationally and internationally. He is currently on the editorial boards of *Journal of Pineal Research*, *Neuroendocrinology Letters,* and *Integrative Cancer Therapies*. Dr. Blask's research has been supported by grants from the National Institute of Child Health and Human Development, the National Cancer Institute, the American Institute for Cancer Research, and several private research foundations.

Dr. Blask is also an accomplished and active jazz trumpet player who has played with and backed up many nationally and internationally known

musicians and entertainers, including Sammy Davis Jr., Bob Hope, Natalie Cole, Diane Schurr, Marilyn MaCoo, Debbie Reynolds, the Temptations, Frankie Vallie, Gladys Knight and the Pips, Bobby Vinton, Manhattan Transfer, Steve Allen, Anne Murray, Frank Sinatra Jr., Aretha Franklin, and Olivia Newton-John, to name just a few.

Melatonin: An Integrative Chronobiotic Anticancer Therapy Whose Time Has Come

David E. Blask, M.D., Ph.D.

Abstract

Melatonin, as a new member of an expanding group of regulatory factors that control cell proliferation and loss, is the only known chronobiotic, hormonal regulator of neoplastic cell growth. At physiological circulating concentrations, this indoleamine is cytostatic and inhibits cancer cell proliferation *in vitro* via specific cell cycle effects. At pharmacological concentrations, melatonin exhibits cytotoxic activity in cancer cells. At physiological and pharmacological concentrations, melatonin acts as a differentiating agent in some cancer cells and lowers their invasive and metastatic status through alterations in adhesion molecules and maintenance of gap junctional intercellular communication.

In other cancer cell types, melatonin, either alone or in combination with other agents, induces apoptotic cell death. Biochemical and molecular mechanisms of melatonin's oncostatic action include regulation of estrogen receptor expression and transactivation, calcium/calmodulin activity, protein kinase C activity, cytoskeletal architecture and function, intracellular redox status, melatonin receptor-mediated signal transduction cascades, and fatty acid transport and metabolism.

A major mechanism mediating melatonin's circadian-stage-dependent tumor growth inhibitory action is suppression of epidermal growth factor receptor (EGFR)/mitogen-activated protein kinase (MAPK) activity. This occurs via melatonin receptor-mediated blockade of tumor linoleic acid uptake and its conversion to 13-hydroxyoctadecadienoic acid (13-HODE), which normally activates EGFR/MAPK mitogenic signaling. This represents a potentially unifying model for the chronobiological inhibitory regulation of cancer growth by melatonin in maintaining the host-cancer balance. It also provides the first biological explanation of melatonin-induced enhancement of the efficacy and reduced toxicity of chemotherapy and radiotherapy in cancer patients.

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EDUCATION
1968-1973: Classic Lyceum in Milan.
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1984-1985: M.D., Centre for Addiction, Monza, Mila, Italy.
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AREAS OF INTEREST IN CLINICAL RESEARCH
Immunotherapy and endocrine therapy of solid tumors and clinical neuroimmunomodulation.

PUBLICATIONS
More than 500 publications in national and international scientific journals.

Melatonin, Chronobiology and Cancer: Clinical Experience with Melatonin Alone or Combined with Standard Anticancer Therapies in Medical Oncology

Paolo Lissoni, M.D.

Abstract

After more than 20 years of investigations on the pineal gland's role in cancer and of clinical studies on its hormone melatonin (MLT) in the treatment of human neoplasms, it seems justified to affirm that acceptance of the pineal gland's importance in neoplastic diseases requires a great modification in the consciousness of human sciences. The aim is to realize a metaphysical refoundation of medical oncology by considering the biological characteristics of tumor cells and the psychoneuroimmune status of cancer patients, which is fundamentally modulated by the pineal gland.

Investigation of the pineal gland may contribute to a new cooperation between philosophy and the biological sciences. Several studies have demonstrated that the progressive decline in pineal function with cancer represents the most frequent cancer-related endocrine alteration. This could play a role in the prognosis of cancer itself, because of the well-documented physiological anticancer activity of melatonin and other pineal indoles.

Melatonin therapy of human tumors is justified for at least three reasons:

1) Endocrine substitution therapy to correct cancer-related pineal deficiency.

2) Melatonin anticancer activity due to antiproliferative action (by modulating oncogene and antioncogene expression and apoptotic processes), activation of anticancer immunity (in particular enhancing IL-2 activity and release by TH1 lymphocytes and stimulation of IL-12 secretion by dendritic cells) and antioxidant property.

3) Palliative therapy, namely cancer-related cachexia and asthenia and thrombocytopenia, because of melatonin's anticachectic and thrombopoietic properties.

The 15-year-old clinical history of melatonin therapy in human neoplasms may be summarized, as follows:

1) Palliative therapy of untreatable solid neoplasms.

2) Cancer neuroimmunotherapy with melatonin plus anticancer cytokines, namely IL-2 to enhance its efficacy in tumors generally resistant to IL-2 alone.

3) Chemoneuroendocrine therapy with melatonin plus chemotherapy and radioneuroendocrine therapy with melatonin plus radiotherapy to reduce toxicity through melatonin's antioxidant property, and to enhance therapeutic efficacy by preventing chemotherapy- and radiotherapy-induced lymphocyte damage and by increasing their cytotoxic power.

4) Neuroendocrine therapy with melatonin plus the classical endocrine therapies of cancer on the basis of melatonin's ability to enhance tumor endocrine dependence. In all studies, melatonin was given orally one/day in the dark phase of the photoperiod at doses ranging from 20 to 40 mg/day.

In randomized studies on 1440 untreatable advanced solid tumor patients receiving melatonin or supportive care alone, melatonin prolonged survival time and prevented neoplastic cachexia, even though objective tumor regression was observed only in 2% of patients.

More promising tumor regression results have been achieved by cancer neuroimmunotherapy with melatonin plus subcutaneous low-dose IL-2 (3 MIU/day), with a tumor regression rate of 16% in 400 untreatable advanced solid tumor patients, with particular efficacy in hepatocarcinoma, gastrointestinal tumors, non-small-cell lung cancer, and mesothelioma.

In a randomized study of 450 advanced cancer patients with poor clinical status or chemotherapy-resistant tumors, melatonin appears to enhance tumor response rate, prolong the survival time with respect to chemotherapy alone, and prevent chemotherapy-induced

thrombocytopenia, cardiotoxicity, neurotoxicity, and asthenia. No effect was seen in protection from anemia, leukopenia, and alopecia. Melatonin in association with tamoxifen appeared to induce a response rate of 29% in 14 metastatic breast cancer patients on tamoxifen alone, and a response rate of 12% in 25 patients with advanced neoplasms other than classical endocrine-dependent tumors.

Melatonin therefore may be successfully used in medical oncology either alone or to biologically modulate conventional anticancer therapies, including chemotherapy, radiotherapy, immunotherapy, and endocrine therapy. Applying melatonin in the treatment of human neoplasms could completely modify medical oncology in a biomodulatory way, re-establish the psychobiological unity of patients, and reconstitute the psychoendocrine and immunobiological responses against cancer growth. Successive studies will be required to establish the efficacy of melatonin in relation to melatonin receptor expression by cancer cells and to the endogenous production of melatonin itself.

Cancer is a biological war that destroys the unity of life, and melatonin therapy may help restore harmony in the biology of cancer patients. This is the contribution of the pineal gland to peace, by involving all people and traditions in a common effort to win the fight against cancer on the basis of a new philosophic interpretation of the human biology.