CHRONOBIOLOGY: IT'S ABOUT TIME

By Roberta Lee, MD, and Michael J. Balick, PhD

As the stars cross the sky on a clear night, the uncountable points of light weave a brilliant tapestry across the sky. Long ago, people began to observe the patterns and formations of the stars and planets, as well as how the sky differs during different times of the year. The Maya people of Central America were known for their precise astronomical observations and were guided by some of these observations in their everyday life. One example of this is the Maya belief that certain times of the day or month are best for the harvest of medicinal and other useful plants. They based this on observations, for example, of how sap flow differed during the phases of the moon.

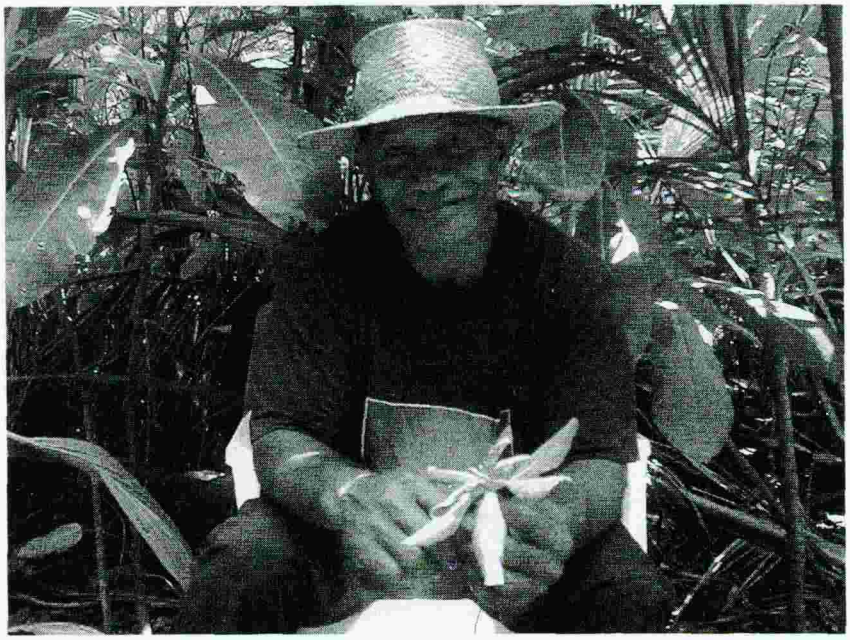
Today, in Central America, farmers note the phases of the moon, and this information guides their planting and harvest of useful plants. Arvigo and Balick[1] explained how important it is to recognize the phases of the moon when, for example, harvesting thatch from palm trees for constructing a house. They tell the story of a newcomer to the region, a North American, who contracted with a local person to build a traditional thatched house. The job had to be completed within a certain period, but, when the builder ran out of seasoned palm leaves, he advised the owner that he would have to wait until the following month before more leaves could be cut and dried. Only then would the moon phase be right–the builder would only cut leaves, and for that matter lumber as well, on the full-moon phase. After a long discussion best described as an argument, the owner insisted that the roof of the house be finished using newly harvested leaves, collected that day, dried in the sun, and woven into the traditional thatch. The builder agreed to do what his client insisted, and thatched one third of the roof with the leaves that, according to his tradition, were not harvested at the right lunar cycle. Within three months of moving into the lovely new dwelling, of course, one third of the thatched roof had decayed and fallen to pieces. The newcomer learned a lesson, at some cost, and the roof was then properly completed with leaves harvested on the proper moon phase.

Curiously enough, on a recent trip to the island of Kosrae, in the Federated States of Micronesia, one of us (M.B.) speaking with an elder heard virtually the same story regarding the use of the *Fahsuc* palm (*Nypa fruticans* Wurmb.) for thatch. According to Tadao Wakuk, local Kosrae historian and legend teller, the leaves of *Fahsuc* should only be harvested during the period that follows the full moon, until the moon is "gone." When past that period, the palm must never be harvested. If harvested too early in the moon phase, the termites and other insects will destroy the leaves, reducing the useful life of the thatch from six to 10 years down to two years at best. This was explained in a room full of young people, all of whom had built *Fahsuc*-thatched houses, but none of whom had ever heard about the proper way of harvesting the thatch. How is it that such similar information is preached by elders, in two isolated spots of the world, many thousands of miles apart? Ethnobotanical theory holds that, when similar beliefs about plant use are gathered from several isolated sites not known to have previous contact, the probability that the belief has validity is significantly greater.

Cultural beliefs such as these, tested over millennia, can be examined with contemporary scientific tools and judged as to their veracity. In the case of the above, we now know that the moon influences the movement of water (and its contents) between different parts of a plant. Zürcher et al[2] showed that the volume and movement of fluid in spruce trees (*Picea abies* L. Karst.) measurably changed the stem diameter of the trees, correlated to tidal strength and timing, on a daily cycle. Others have shown direct relationships between growth of plants and moon phase.[3] However, as Arvigo and Balick[1] have noted, "the effects of the moon on the earth's oceans are well-known and have given rise to conjecture about its impact on other water-related aspects of our lives as well," including ". . .childbirth, menstruation, and human behavior."

In addition to the response to lunar cycles, plants have many internal cycles, which make their behavior appear somewhat similar to that of animals. They can tell time–anticipating the maximal period for flowering, determine germination, and detect unfavorable seasonal cycles of harsh weather. They can also respond to environmental changes. These responses make sense from an evolutionary perspective because there is much to lose from an ill-timed response. What could be worse timing than flowering in the dead of winter when insects may be dormant and unavailable for pollination, or, worse still, temperatures may be so cold that the flowers would be vulnerable to freezing?

Circadian rhythms are cycles that occur in intervals of approximately 24 hours under relatively constant environmental conditions.[4] Plants, animals, and insects have a mechanism responsible for this phenomenon. This is an independent internal timing mechanism often identified as an organisms' "biological clock." The factors responsible for keeping this free-running rhythm going are genetic in origin, and the external influence that entrains this rhythm is the environment. For plants, photoperiodism serves as the major environmental entrainment. *Photoperiodism* is defined as the response to changing 24-hour cycles of light and dark. This measurement is crucial in determining the onset of flowering in many plants. Plants fall into various flowering categories determined by the day length. Plants that flower when the light exceeds a critical length are labeled as long-day plants, and others that flower only when the day length is less than a critical period are labeled as short-day plants. There are also day-neutral flowering plants. Subsequent experiments have shown that the dark pe-



Tadao Wakuk in the historical site of Lelu, Kosrae, FSM (Photo taken by Roberta Lee, 1999).

riod actually is the most critical determinant of flowering phenomena.

The molecule responsible for photoreception, also identified as a *phytochrome*, is composed of a light-absorbing portion and a large protein moiety–some plants have many phytochromes, and cyanobacteria also have similar phytochromatic molecules. This molecule can exist in two forms: Pr and Pfr. Exposure to a particular UV wave length (660 nanometers identified as red light) determines the transition from Pr to Pfr, and, conversely, another UV wavelength exposure (730 nanometers identified as a far red light) is required for its reversion back to the original form. Pfr is the active form of the phytoreceptor promoting flowering in long-day plants and inhibiting flowering in short-day plants. Pfr also promotes normal seedling growth. Interestingly, photoperiodism is perceived in the leaves, but the actual responses occur in the bud, and, although it has not been identified, there seems to be a chemical substance that flows through the plant's circulatory system from the leaves to the bud that generates this response.

Dormancy, or arrested growth in plants, is also determined by day length cycles. This response enables plants to survive extreme temperatures or predictable periods of water shortage. There are many other cyclic phenomena within plants such as solar tracking and shoot growth of roots that are determined by photoreception as well.

Recently, melatonin, an indoleamine neurohormone well-known in human and mammal physiology, has been identified in plants.[5] Melatonin appears to be in high concentration in actively growing plants–among the plants identified with significant amounts of this hormone are St. John's wort (*Hypericum perforatum L.*), feverfew (*Tanacetum parthenium L.* Schultz Biop.), and Huang-quin (*Scutellaria biacalensis*).[6] Serotonin, another familiar neurohormone, has also been identified in plants.[7] These neurohormones are thought to influence the light/dark signaling mechanisms and root organogenesis. Because the discovery is so recent, exact mechanisms of their roles is still to be determined.[8] In musing about the recent effects of neurohormones in plants, experts have wondered if the plants with higher amounts of melatonin/serotonin used in indigenous treatments of syndromes related to perturbations of the nervous system could be, in part, successful because of these newly identified plant neurohormones.[6] In other words, is it possible that St. John's wort is modestly effective for modulating mood disorders because melatonin is synergizing with other identified "active ingredients?" Future research clarifying the role of these constituents in plant physiology and the implications of their role in botanical pharmacology continue to be investigated.

There are a myriad of cycles influencing human physiology. Melatonin rises in the night and disappears in the day.[9] Cortisol, secreted by our adrenal glands, rises in the morning and peaks at 9 AM but is low as we begin to sleep in the evening.[10] Similarly, blood pressure has a diurnal circadian pattern–it's worst in the early morning and late afternoon and is at its lowest in the evening before sleep.[11] Exacerbations of asthma are typically greatest between the hours of 2 AM and 4 AM.[12] However, migraine headaches and allergic rhinitis are most likely to begin between 6 AM and noon,[13] and heart attacks occur with a 30% to 40% higher probability from 6 AM to noon.[14] Despite these facts, a study surveying 320 physicians on their knowledge of the body's chronobiological cycles revealed that only 5% of physicians identified themselves as familiar with chronobiological cycles, and 44% said they were somewhat familiar with the concept.[15] However, as a physician, I (R.L.) can recall very few protocols that address this issue in conventional medicine to maximize therapeutic outcomes.

There are a growing number of researchers specialized in different biological disciplines who feel that chronobiological patterns may be important in maximizing a drug's safety and efficacy profile. Utilizing chronobiological information in clinical medicine may even reduce a drug's toxicity.[16,17] Authors Russell Foster and Leon Kreitzman, in their book *The Rhythms of Life: The Biological Clocks that Control the Lives of Every Living Thing*, noted that "Rats or mice are commonly used as experimental animals in screening

Fahsuc palm (Nypa fruticans Wurmb.) in mangrove forest taken in Kosrae, FSM (Photo by Michael Balick, June 2006).

the carcinogenic activity of drugs and additives. But rats are nocturnal animals." They suggested that results from animals tested in a resting diurnal are physiologically different than "diurnally active humans."[18] Thus, in testing the potential of carcinogenic substances during the day, a time when rodent metabolism is in a resting state and DNA replication is at its lowest level of activity, findings would reveal a falsely higher threshold of susceptibility–suggesting that a higher concentration of the tested substance would be "safe." Conversely, if a chemical is screened in mice at night, when rodent metabolism is active and DNA replication is at its highest, findings would result in a chemical safety profile that had a much lower threshold of susceptibility for toxicity. E. Robert Burns, a neurobiologist and senior researcher in cell kinetics and chronochemotherapy at the University of Arkansas for Medical Sciences, has estimated that the difference in cross species testing omitting chronobiological differences could result in findings that lower toxic susceptibility by 40%.[19] In other words, safety data appropriately accommodating for circadian changes across animal models could result in the lowering of a "safe dose" for a carcinogenic compound by almost half its purported nontoxic concentration.

In clinical oncology, an area in which the benefits and risks of optimal dosing efficacy and maximal reduction of side effects and drug toxicity are extremely significant, chronotherapy seems to have the most potential. At this time, approximately eight "clock genes" have been identified that interface and generate biological rhythms. These findings have resulted in the development of chronotherapy.[20] Chronotherapy is the administration of chemotherapeutic agents timed to optimize trends in biological cycles related to immunity, kidney function, tumor tissue replication, and other processes for cancer treatment. Although investigated as a potential therapeutic treatment for over four decades, its application in clinical oncology in the United States remains scarce and is categorized as "cutting edge."[21] William Hrushesky, a senior clinical investigator specializing in chronobiology at the Dorn VA Medical Center in South Carolina, suggests that "chronotherapy allows for larger [chemotherapy] doses to be delivered more frequently, with a higher efficacy and a lower toxicity."[21] However, chronobiology experts acknowledge that, despite promising results, significant logistical hurdles exist in bringing this type of approach to cancer treatment in clinical medicine. Keith Block, MD, founder and medical director of the Block Center for Integrative Cancer Care in Evanston, Illinois, agrees that chronotherapy in clinical oncology is challenging. However, in providing this type of care for a decade, he also observes that chronotherapy has fewer oncological complications (mainly drug toxicity and renal failure) and other side effects (nausea and fatigue; personal communication; June 15, 2006; Roberta Lee). However, only one center in the United States offers chronobiologicaly administered chemotherapy: the Block Center. The other US location that has chronobiology available offers this therapy in the context of clinical research only under the direction of William Hrushkey at the Dorn VA Medical Center in South Carolina. Block noted that, "Patients who receive chemotherapy in this manner have a completely different chemotherapy experience." When his patients receive chemotherapy, they can take a walk, meditate, practice Tai Chi, or even rollerblade. Furthermore, in some cases Block suggested "that using chronotherapy with well-known conventional chemotherapeutic agents extended progression-free survival and overall survival rates in specific cancer types that were metastatic. Furthermore, the results were significantly more favorable than the nonchronobiologic administration using the same protocol (same chemotherapy agents)."[22]

Drug toxicity in oncology is minimized by balancing excessive toxicity to the patient and antitumor activity. Sometimes, the unintended consequences for complications using particular chemotherapy agents such as cisplatin can result in kidney failure, despite rigorous dosing schedules. This event appears to be dramatically minimized by chronotherapeutic administration.[23,24] In his clinical population, Block confirmed this observation (personal communication; June 15, 2006; Roberta Lee).

We live in a world that demands immediate response. Our work hours are significantly shifted from ancient patterns of rising at dawn and resting at dusk. Some of us spend years working in schedules that are completely reversed from the normal day/night patterns. And worse still, it is becoming increasingly difficult to completely remove ourselves from our instant communication devices. Do we need this much stimulation, and what are the long-range effects for our health? We have touched upon the importance of cycles in plants–noting significant tidal responses to lunar cycles, recently discovered neurohormones in plants, and recognized the potential of innate circadian rhythms in maintaining health. Our ancestors–shamans, traditional healers, and common folk alike–all knew the optimal cycles of the weather, seasons, and periods of the day for harvesting plants to build dwellings and prepare medicines. With painstaking and persistent effort, elders in many indigenous cultures pass this knowledge to their young. In hindsight, the observations of elders that have seemed interesting, even novel but somewhat anecdotal, now appear with more research to be once again intriguing–the ancient lore of harvesting plants perhaps alluding to very complex biological relationships yet to be investigated. In 1675, Sir Isaac Newton said, "If I have seen farther than certain other men, it is by standing upon the shoulders of giants." Recognize that by supporting ourselves on the traditional wisdom of the ancients, we might just gain a better view of our world and how it works. Perhaps it is time to give more respect to nature's pendulum.

REFERENCES

1. Arvigo R, Balick M. *Rainforest Remedies: One Hundred Healing Herbs of Belize*. Twin Lakes, WI: Lotus Press; 1998.
2. Zürcher E, Cantiani MG, Sorbetti-Guerri F, Michel D. Tree stem diameters fluctuate with tide. *Nature* 1998;192:665.
3. Endres K.-P., Schad W. *Biologie des Mondes, Mondperiodik und Lebensrhythmen*. Stuttgart-Leipzig: S. Hirzel Verlag; 1997.
4. Raven P, Evert R, Eichhorn S. *Biology of Plants*. 6th edition. New York: W.H. Freeman and Company; 1999:702–725.
5. Hattori A Migitaka H, Iigo M, et al. Identification of melatonin in plants and its effects on plasma melatonin levels and binding to melatonin receptors in vertebrates. *Biochem Mol Biol Int* 1995;35:627-634.
6. Murch S, Simmons C, Saxena P. Melatonin in feverfew and other medicinal plants. *Lancet* 1998;350:1598-1599.

7. Murch S, Campbell S, Praveen S. The role of serotonin and melatonin in plant morphogenesis: regulation of auxin-induced root organogenesis in in vitro-cultured explants of St. John's wort (*Hypericum perforatum* L.). *In Vitro Cell Dev Bio* 2001;37:786-793.
8. Murch S, Saxena P. Invited review: melatonin: a potential regulator of plant growth and development? *In Vitro Cell Dev Biol* 2002;38:358-536.
9. Swick T. The neurology of sleep. *Neurol Clin* 2005;23:967-989.
10. Cutolo M, Masi A. Circadian rhythms and arthritis. *Rheum Dis Clin N Am* 2005;31: 115-119.
11. Ozdemir R, Sezgin A, Topal E, et al. Findings of ambulatory blood presure monitoring and heart rate variability in patients with Bechet's disease. *Am J Cardiol* 2003; 92:646-648.
12. Sutherland ER. Nocturnal asthma: current reviews of allergy and clinical immunology. *J Allergy Clin Immunol* 2005;116:1179-1186.
13. Gelfand E. Inflammtory mediators in allergic rhinitis. *J Allergy Clin Immuno.* 2004; 114:S135-s138.
14. Guo YF, Stein P. Circadian rhythm in the cardiovascular system: chronocardiology. *Am Heart J.* 2003;145:779-786.
15. Smolensky MH. Knowledge and attitudes of American physicians and public about medical chonobiology and chronotherapeutics. Findings of two 1996 Gallup surveys. *Chronobiol Int* 1998;15:377-394.
16. Ohodo S. Changes in toxicity and effectiveness with timing of drug administration: implications for drug safety. *Drug Saf* 2003;26:999-1010.
17. Chassard R, Allaouchiche B, Boselli M. Timing is everything: the pendulum swings on. *Anesthesiology* 2005;103:454-456.
18. Foster R, Kreitzman L. *Rhythms of Life: The Biological Clocks that Control the Daily Lives of Every Living Thing.* New Haven: Yale University Press; 2004:214–216.
19. Burns ER. Biological time and in vivo research: a field guide to pitfalls. *Anat Rec* 2000;261:141-152.
20. Eruguchi M, Levi F, Hisa T, et al. Chronotherapy for cancer. *Biomed Pharmacother.* 2003;57:S92-S95.
21. Derdorff J. Qualities of life: put time on your side. Treatments synchronized with your body clock might make all the difference. March 12, 2006. Available at: www.chicagotribune.com. Accessed June 22, 2006.
22. Levi F, Misset JL Brienza S. A chronopharmacologic phase II clinical trial with 5-Florouracil, folinic acid and oxaliplatin using an ambulatory multichannel programmable pump. High antitumor effectiveness against metastatic colorectal cancer. *Cancer* 1992;69:893-900.
23. Von Reomeling R. The therapeutic index of cytotoxic chemotherapy depends upon circadian drug timing. *Ann N Y Acad Sci* 1991;618:292-311.
24. Li XM, Tanaka K, Sun J, et al. Preclinical relevance of dosing time for the therapeutic index of gemcitabine-cisplatin. *Br J Cancer* 2005;92:1684-1689.

Michael J Balick, PhD, *is vice president and chair of botanical research and training, philecology curator and director, Institute of Economic Botany, The New York Botanical Garden. An ethnobotanist, he studies the use of plants by traditional cultures.*

Roberta Lee, MD, *is medical director at the Continuum Center for Health and Healing at Beth Israel Medical Center, New York. She is co-director of the Integrative Medicine Fellowship. Her research involves ethnomedicine, the study of healing systems used by traditional cultures.*