

The Science of Photobiology

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The U.S. National Committee for Photobiology (National Academy of Sciences/National Research Council), recognizing the need for a national organization for photobiology announced in June 1972 its intention of forming the American Society for Photobiology. The newsletter making this announcement contained the following statement:

The U.S. National Committee for Photobiology of the National Academy of Sciences/National Research Council recognizes that there is a growing general awareness of the unique importance of the effects of light (both beneficial and detrimental) on man and all other living organisms, that the science of photobiology is generated by scientists of diverse educational and practical experience and therefore needs a vehicle for enhanced communication and the dissemination of knowledge, and that current problems of national and international concern require an accurate and effective input of knowledge of photobiology and photochemistry. Therefore, while it will continue to serve as the U.S. liaison for international photobiology as the U.S. representative to the Comité International de Photobiologie, the U.S. National Committee for Photobiology (NAS/NRC) has decided to form an American Society for Photobiology and to delegate its national responsibilities to this Society.

Since this initial announcement, over 700 scientists have joined the American Society for Photobiology including many from outside the Western Hemisphere. The research interests of the members cover one or more of the 14 specialty subgroups of the Society, namely: bioluminescence, chronobiology, environmental photobiology, medicine, photochemistry, photomorphogenesis, photomovement, photoreception, photosensitization, photosynthesis, phototechnology, spectroscopy, ultraviolet radiation effects, and vision.

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The first annual scientific meeting of the Society was held in Sarasota, Fla., 10-14 June 1973. There were 240 scientists registered for this historic first meeting; some came from as far away as Europe and Brazil.

Sunlight is probably the most important single element of our environment, yet it has been largely ignored by the scientific community—perhaps because of its ubiquity. Previously, there have been no compelling reasons to study the biological effects of light except to satisfy the curiosity of a small number of scientists. This situation is now changing as people become more aware of the effects of light other than as an aid to vision. The publicity about the supersonic transports (the SST's) and the consequences of a possible SST-induced increase in solar ultraviolet radiation reaching the surface of the earth; publicity about the treatment of congenital jaundice in premature babies with light therapy, the treatment of Herpes infections (the common cold sores), and even of malignant tumors with dyes plus visible light; and, the concern over the energy crisis and its possible solution by harnessing solar energy have all helped to focus attention on the properties of sunlight.

The scientific program of the 1st annual meeting of the American Society for Photobiology was planned to highlight certain important effects of sunlight and of artificial light. Six lectures for general audiences were presented. A Photobiology School was instituted to introduce basic principles of photobiology to students and to serve as a refresher course for more senior researchers. In addition to 124 contributed papers, eight specialized symposia were presented.

The purpose of this report is to survey briefly the new insights in the science of photobiology that emerged from this first meeting of the American Society for Photobiology.

Detrimental Effects of Excessive Exposure to Sunlight. In a symposium entitled the Photobiology of Disease, the detrimental effects of excessive exposure to sunlight were discussed. To avoid the sun would be to exist without one of the great pleasures of life. But as with most enjoyable things, indiscriminate exposure and lack of understanding of the possible unpleasant consequences can result in unhappiness and even serious illness.

Among the known effects on the skin of man of natural and artificial UV radiation are sunburn, changes in skin which are interpreted as signs of "aging," and premalignant and malignant skin tumors. A better understanding of the cell constituents affected, the kinds of alterations produced, the type of repair mechanisms, and the effect of interactions among these is needed in order to combine the maximum benefits with the least damage from UV radiation (F. Urbach, Temple University).

Chemicals present in soaps, cosmetics, medicine, and environmental pollutants can sensitize people to sunlight, leading to intense sunburnlike reactions even in the absence of UV radiation (e.g. sunlight filtered through window glass). Two mechanisms exist for this type of photosensitization: (1) Phototoxic reactions are mediated by the absorption of radiation by the photosensitizer, and the transfer of this absorbed energy to biological molecules, leading to their chemical alteration (P. D. Forbes, Temple University). (2) If the absorption of light photochemically changes the structure of a molecule such that it is now recognized by the body as foreign, antibodies are produced leading to a photoallergic response (L. C. Harber, New York University).

Man's sensitivity to sunlight is controlled by heredity. This is exemplified by genetic deficiencies in melanin formation and thus the absence of tanning (as in Irish, Scottish, and Welsh peoples); deficiencies in cellular capacity to repair solar radiation damage, as in the inherited disorder xeroderma pigmentosum; metabolic over-production of porphyrin (a natural photosensitizer); and altered tryptophan metabolism (J. H. Epstein, University of California, San Francisco).

The wavelengths of sunlight below 320 nm, those modulated by the presence of ozone in the stratosphere, are the most detrimental to biological systems. Yet it is this same wavelength region of light that produces the essential vitamin in the skin of man—vitamin D. Thus, the situation is one of balance: sunlight is necessary for life, yet in excess, it is harmful.

Repair of UV Radiation-Induced Damage to Cells. Most organisms, with the notable exception of man, tend to shun sunlight unless they are well protected from the damaging effects of UV radiation by external shields such

as feathers, hair, shells, and pigments (A. C. Giese, Stanford University). When cells are exposed to radiation, their sensitivity, measured in terms of lethality, depends mainly upon their ability to repair radiation-induced damage in their deoxyribonucleic acid (DNA). This point is dramatically exemplified by the observation that mutant cells that are genetically deficient in DNA repair systems are much more easily killed by UV radiation.

Currently three major enzymatic pathways are known for the repair of UV-induced damage to DNA:

1. The damaged part of the deoxyribonucleic acid molecule is restored to its functional state in place. A large percentage of a culture of cells (except those from placental mammals that have been inactivated by UV radiation (254 nm) can be reactivated by a second irradiation with near UV radiation around 380 nm. This enzymatic process is called photoreactivation.

2. The damaged sections of a DNA molecule are cut out and replaced with undamaged nucleotides to restore the normal function of the DNA. This excision mode of repair does not require light for it to function, and appears to be ubiquitous in nature. Furthermore, it is not restricted to the repair of UV radiation-induced damage but also repairs certain types of chemical and X-ray-induced damage to DNA (R. D. Ley, Argonne National Laboratory). Several new branches of the excision repair process were described (D. A. Youngs and K. C. Smith, Stanford University).

3. The damaged section of DNA is not directly repaired but is bypassed during replication. When the damage in the parental strands of DNA are bypassed during normal replication, gaps are produced in the newly synthesized daughter-strands of DNA. These gaps are subsequently repaired, yielding normal daughter-strand DNA, but the damage remains in the parental strands. This post-replicative repair process is also a dark-repair process (W. D. Rupp, Yale University).

While UV radiation produces very few breaks directly in DNA molecules, in the process of repairing UV radiation-induced damage to DNA, enzyme-induced breaks are produced in the DNA. These breaks are then subsequently repaired. Near-UV radiation and visible light in the presence of photosensitizing dyes produce breaks in DNA molecules directly, as do X-rays. These direct radiation-induced breaks in DNA molecules are also repaired by three separate systems. The speed of the repair and the complexity of the repair process appear to depend upon the chemical complexity of the chain breaks. Two of the systems for the repair of X-ray-induced chain breaks in DNA have also been found to repair some of the enzyme-induced chain breaks produced after UV irradiation (D.A. Youngs, Stanford University).

The Roles of Light in the Human Environment. The development of varied and powerful sources of artificial light from electricity has led to sophisticated knowledge of illumination and its measurement—usually in terms of its perception by the eye. But visible

light, as much as UV or infrared radiation, has the ability to exert measurable biological effects. Medical uses of the visible spectrum have been virtually ignored by physicians for the past 90 years. However, there is a new appreciation of these uses by medical scientists, stimulated in great part by the advent of phototherapy of the jaundiced newborn.

Directly beneficial effects of light produced by artificial sources include the photorepair of UV damage, treatment of jaundice of the newborn infant with blue light, destruction of the virus of herpes simplex when stained by certain photosensitizing dyes (C. Wallis and J. L. Melnick, Baylor College), diagnosis of some hereditary diseases in utero by activation of fluorescent dyes in fetal cells (S. S. West, University of Alabama), and recently, the destruction of certain cancer cells by visible light irradiation after their incorporation of photosensitizing agents (I. Diamond et al., University of California, San Francisco).

Not all the effects of visible light are beneficial, however. Marked detrimental effects on the retina have been demonstrated under circumstances previously thought innocuous. Certain enzymes and other substances in blood and tissue will absorb light in vivo and thus undergo photochemical decomposition. Photosensitizing drugs given to pregnant women readily cross the placenta to the fetus. If, after delivery, the infant is exposed to light of high intensity, as in phototherapy, the chance of photosensitization is increased.

The psychological effects of light, particularly of colored light, are well known but not well understood. These effects may bear a causal relationship to purely biological processes in the brain induced by light, which in turn will affect psychic behavior. Light intensity as well as wavelength specificity may alter productivity and mood. In the infant, sensory overload by prolonged exposure to highly intense illumination may produce undesirable effects on development. Indeed, the manipulation of the lighting environment of adults as well as of infants can have consequences of which we may be quite unaware.

The penetrance of visible light in tissues deeper than the skin has not been adequately measured. The penetrance of UV radiation has been measured with some success, so that we do know much about the photochemical actions within these superficial cells, but the photochemical action of visible light upon deep tissues is an area of study that needs much further work.

It is obvious that we must consider the types and sources of artificial light, their intensities and spectral characteristics, and the chemical, physiological, and psychological effects of the lighting environment upon man—not from the standpoint of illumination but from that of specific photobiological consequences of its use (T. R. C. Sisson, Temple University).

Photochemistry in Photobiology. Since all photobiological responses to light are the consequence of photochemical changes produced in biological systems, it is important

to stimulate more chemists to work on the molecular basis of photobiological problems. In a general lecture, A. A. Lamola (Bell Laboratories) discussed how, in addition to identifying photoproducts, chemists can contribute to the understanding of photobiological phenomena by studying details of the photochemical steps. Once the photochemical mechanism is known it is usually possible to learn how to modify the photochemistry, which is quite useful for relating molecular events to photobiological phenomena.

The area of photobiology that Lamola believes is presently best suited for significant input from photochemists is that of photosensitization of biological systems by small molecules. Photosensitized reactions are induced by the absorption of light by a photosensitizer molecule. The excited photosensitizer molecule can then act in a variety of ways which lead to an altered target molecule, eventually resulting in a biological effect. Many of these photosensitized reactions can be modeled in a useful way that also allows scrutiny by the techniques of modern photochemistry.

Because many natural and synthetic chemicals can be altered by sunlight to produce compounds toxic to man and other animals, and to plants, it is important to study the photochemistry of all chemicals produced by man which may become exposed to sunlight. In addition, a national registry of cases of phototoxicity would speed the removal of such agents from commerce.

Effects of Near UV Light (320-400nm). Near UV light has been considered by many to be a harmless form of radiant energy. A typical source of this type of radiation is the so-called "black light" bulb used to display fluorescent posters, etc. However, in vitro studies have shown that light between 320 and 400 nm can photooxidize aromatic amino acids into toxic compounds (S. Zigman, University of Rochester; F. Landa and A. Eisenstark, University of Missouri). This light also inhibits the growth of bacteria and destroys ubiquinone within bacterial cells (H. Werbin et al., University of Texas, Dallas). It causes the drug psoralen to combine with the DNA of mammalian cells (E. Ben-Hur and M. M. Elkind, Brookhaven National Laboratory; L. Musajo et al., University of Padua; M. A. Ashwood-Smith and E. Grant, University of Victoria), and it inactivates transforming DNA (J. G. Peak et al., Argonne National Laboratory).

The wavelengths of sunlight which are modulated by the ozone in the stratosphere, i.e., 280-320 nm, are the wavelengths of light that produce sunburn and skin cancer. Wavelengths longer than 320 nm are very ineffective in this regard. However, if both types of light are used together they produce a greatly enhanced sunburn reaction in human skin (M. Pathak et al., Harvard University) and an increased incidence of skin cancer in mice (P.D. Forbes, Temple University).

Photosynthesis. C. C. Black (University of Georgia) reported on different mechanisms by which plants take carbon dioxide from the air to form sugars, which are then converted

into other forms of plant food. The first process, discovered and described in the 1950's by Nobel prize winner Melvin Calvin, is the classical "pentose cycle." Black described the newly discovered "four-carbon cycle" which operates in grasses and sugar cane. This process is much more efficient, since it takes place at low carbon dioxide concentrations and at higher light intensities, and such plants do not perform photorespiration. The success of the four-carbon pathway depends upon compartmentalization of enzymes within the plant cell. With the intense need for increased food production in the world, information concerning the natural mechanism by which certain plants use sunlight more efficiently in photosynthesis would be of great importance in helping scientists plan for increased food production throughout the world.

There are several types of bacteria which carry out a primitive photosynthesis. Because of its simpler nature, bacterial photosynthesis has been an excellent system for scientific investigation. Roderick Clayton (Cornell University), who was instrumental in the isolation and characterization of the reaction centers from photosynthetic bacteria, presented current views of how this reaction center works in photosynthetic bacteria. Paul A. Loach (Northwestern University) and Kenneth Sauer (University of California, Berkeley) reported results of their studies designed to show how all of the many components of the membrane are put together to allow photosynthesis to proceed. There is general agreement that photosynthesis in the membrane proceeds through photosynthetic units which contain a cluster of protein molecules embedded in a membrane matrix which consists of lipids, or fats. Each of these photosynthetic units carries out a sequence of reactions initiated by the absorption of light leading to the formation of sugar.

Lawrence Bogorad (Harvard University) described how the plant forms the machinery of photosynthesis in the chloroplast. Plants in the dark do not have chlorophyll, nor do they have the fully organized chloroplasts. They do contain a body known as the etioplast, which contains some of the proteins found in the mature chloroplast. In some as-yet-unknown fashion, exposure of a dark-grown plant to light initiates a series of reactions leading to the formation of chlorophyll and new proteins, and their inclusion into the membrane system of the chloroplast to produce the small photosynthetic unit that traps sunlight and produces sugars.

Bioluminescence. Biological reactions that yield light are actively studied since they provide important clues as to the mechanism of interconversion of light and biological energy at the molecular level. In the sea pansy, several enzymes which give rise to a sequence of reactions leading to bioluminescence are packaged together in a subcellular particle which has been isolated and named the "lumisome" (M. J. Cormier et al., University of Georgia). In certain jellyfish, however, a single protein has been isolated which con-

tains both luciferase and a calcium activated luminescence activity (S. J. Girsch and J. W. Hastings, Harvard University) although others have been able to resolve this system into five components (H. H. Seliger, Johns Hopkins University).

Bacterial bioluminescence comes from FMNH⁺, since the emission spectra from various types of bacteria can be precisely matched by the fluorescence of FMNH⁺ in an appropriate environment, and certain chemical shifts and the quantum yields also correspond (J. Lee et al., University of Georgia).

Light Perception Without Eyes. Light is perceived by structures other than the eyes in all classes of non-mammalian vertebrates. Most of the experimental evidence supporting this statement comes from studies on the role of light in controlling biological rhythms and seasonal reproductive cycles (e.g., testicular weight in birds). While the photoreceptive structures involved have not been precisely localized or identified, it is clear that, in some cases, they are located in the brain but are not the eyes nor the pineal gland. The interrelationships and adaptive significance of these extraocular photoreceptors present a challenge to the ingenuity of photobiologists (M. Menaker, University of Texas).

Photomotion. Plants can orient themselves with respect to light so that they are in an optimum position for carrying out photosynthesis. Motile one-celled plants share this ability with the more familiar flowering plants. Some concentrate in a light beam while others are able to move toward or away from a light source. Mary Ella Feinleib (Tufts University) discussed photomotion in microorganisms. The intriguing question is: How does a microbial cell detect light direction? Basically there are two possible mechanisms: (1) It may compare light absorbed in two regions of the cell at one point in time, or (2) It may compare light absorbed in one region of the cell at two points of time. The second mechanism appears to operate in *Euglena*. While information is minimal as to the nature of these photoreceptors in microorganisms, almost nothing is known about the transmission of photomotion signals from receptor to effector.

Chronobiology. The ability to distinguish time of day without reference to external light and darkness is found in plants and animals of all sizes and levels of complexity. This was discovered by study of rhythmic changes in activity or other physiological functions which can continue in constant light and temperature, a discipline now called chronobiology. One example of such a rhythm is the nocturnal activity of cockroaches which continues in cycles when these insects are kept in constant darkness.

Light has a number of important effects on this time sense or circadian clock, as it is sometimes called. Light keeps the timing cycle synchronous with environmental day and night and adjusts it to long or short days, even stops or starts it under certain conditions.

In the symposium on Light and Biological Rhythms the topics discussed were: how light

interacts with circadian clocks, the relationship of this interaction with photoperiodism, and the response of animals and plants to season through detection of day length (A. T. Winfree, Purdue University; Ruth Halaban, State University of New York; J. W. Truman, Harvard University).

The Ultraviolet World of Insects. For man the visible spectrum ends at about 380-400 nm, but for many insect species vision extends to 300 nm in the ultraviolet region. Moreover, near UV light is a distinct color for many species of insects and has special significance in influencing the behavior of this large and ecologically important group of animals. For example, because near UV light is the most effective in attracting insects, insect traps are fitted with UV lamps. Conversely, because lamps that are poor in blue and UV light offer much less stimulation to insects, yellow bulbs are frequently used to illuminate porches and patios. There are other examples, less obvious but vastly more important ecologically. Flower colors frequently involve patterns of differential UV light reflectance that can be appreciated by insect pollinators but not by the unaided human eye. The wings of butterflies also contain patches of high UV reflectance which flag prospective mates. In flight these signals can be quite conspicuous to other members of the species but remain unseen by vertebrates (T. H. Goldsmith, Yale University).

Vision. Considerable attention was given at this meeting to a problem which has long plagued the scientist studying photobiological processes such as vision; namely, how light absorbed by an organism is eventually converted into metabolic or sensory information for the organism. In vision science, this problem centers on attempts to describe how light impinging on the retina is eventually translated into a signal that goes to the brain. E. A. Dratz (University of California, Santa Cruz) described biochemical and electrochemical models for the rod photoreceptor cells of vertebrate retina. E. W. Abrahamson and co-workers (Case Western Reserve) reported that upon light exposure, calcium released within the photoreceptor cells of the retina is partially responsible for producing the electrical signals which eventually reach the brain.

Several groups reported explicit studies designed to determine how light-sensitive parts of the retina are constructed (C. R. Worthington, Carnegie-Mellon University; W. Stoeckenius, University of California, San Francisco; R. B. Park, University of California, Berkeley; B. J. Litman, University of Virginia; P. J. O'Brien, National Institutes of Health; and S. L. Bonting, University of Nijmegen, The Netherlands).

The isolated perfused eye coupled with electroretinography and time-lapse photography was demonstrated as a useful tool in photobiology and pharmacology (A. L. Marchese and A. H. Friedman, Loyola University).

Solar Energy Conversion. In a symposium in honor of Farrington Daniels, Sr., and chaired by A. Hollaender (Oak Ridge National Laboratory), R. K. Clayton (Cornell University),

L. O. Krampitz (Case Western Reserve University), and L. Herwig (National Science Foundation, RANN) surveyed various schemes for solar energy conversion, some involving biological processes: (1) Direct heating. (2) Growth of grain and hence of cattle and chickens. Growth of algae, aided by nutrient wastes. (3) Photosynthetic production of hydrogen from water. (4) Solar batteries: Inorganic, organic, cells patterned on photosynthetic models.

In terms of the conversion of solar energy, the most promising mechanisms appear to be solar powerhouses; i.e., the trapping of heat to run conventional turbines or to run chemical reactions leading to the splitting of water thus yielding hydrogen gas. Certain types of solar batteries also look promising (e.g., silicon cells and cadmium sulfide). Energy conversion based upon photosynthesis or chemical models of photosynthesis may require many years to develop. Solar energy conversion, however, is probably the only ecologically acceptable source of power.

Summary. One cannot help but be impressed by the great number of ways that plants and animals are affected, both beneficially and detrimentally, by light. Yet, in most scientific experiments using animals and cells, the quality and quantity of light and its cyclicity are totally ignored. Clearly, because of the unique physiological importance of light to all living things, the light environment in experiments must be accurately controlled in the same way that, for example, temperature and pH are controlled.

The future of the science of photobiology seems bright. Its goals can be roughly divided into four categories: (1) The development of ways to protect organisms, including man, from the detrimental effects of light; (2) The development of ways to control the beneficial effects of light upon our environment; (3) The continued development of photochemical tools for use in studies of life processes; and (4) The development of photochemical therapies in medicine. The science of photobiology appears to have come of age as a major new scientific frontier.

Acknowledgment

I wish to thank R. K. Clayton, J. Jagger, A. A. Lamola, J. Lee, H. Shichi, B. M. Sweeney, and L. P. Vernon for their helpful comments on this manuscript.

The symposia at the Sarasota meeting were supported in part by a contract from the Climatic Impact Assessment Program of the Department of Transportation.

