Lightning Bugs

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Bioluminescence is the phenomenon of light emission by living organisms. This is well exhibited in many insects, and best understood in fireflies. Bioluminescence is the result of chemical reactions primarily involving luciferin, luciferase and oxygen. Luciferin is a heat-resistant substrate and the source of light; luciferase, an enzyme, is the trigger, and oxygen is the fuel. Luminescing insects utilize light as a mating signal, to attract their prey, or to defend themselves from enemies. This biological phenomenon has been exploited in space and medical research, insect pest management, and is also a useful tool in biotechnology.

Bioluminescence is the ability of certain animals to produce light, a phenomenon primarily seen in marine organisms. It is the predominant source of light in deep oceans. The light production is the result of chemical reactions and hence it is also called *'chemiluminescence'*. Bioluminescence is exhibited by bacteria, fungi, jellyfish, insects, algae, fish, clams, snails, crustaceans, etc. Bioluminescent bacteria have been found in marine, coastal and terrestrial environments. Some fungi can also emit light. Luminescent fungi such as *Armillaria mellea* and *Mycena* spp. produce a continuous (non-pulsing) light in their fruiting bodies and mycelium. It is believed that bioluminescent fungi use their light to attract insects that will spread the fungal spores, thus enhancing their reproduction. Some nematodes are luminescent due to the presence of symbiotic bacteria associated with them. Nematodes of the genus *Neoaplectana*, *Steinernema* and *Heterorhabditis* have a symbiotic association with luminescent bacteria like *Xenorhabdus luminescens* and thus exhibit luminescence. There are no luminous flowering plants, birds, reptiles, amphibians or mammals in nature. Though bioluminescence is generated by various organisms, it is highly

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developed in insects. The examples of true or self-luminescence are found in Collembola, Diptera, Coleoptera and Homoptera.

The order Coleoptera constitutes the largest bioluminescent group in which several hundred species are known to contain highly developed photogenic organs. The best understood luminous insects belong to the families Lampyridae, Elateridae and Phengodidae. The members of Lampyridae are called fireflies or lightning bugs. Their immature forms are commonly referred to as glowworms, while the adults are called fireflies. Similarly, the individuals of Elateridae are called wireworms or click beetles. Scientists have discovered that the brightest insect is the very large *Pyrophorus noctilucus* (Elateridae), with a brightness of 45 millilamberts. This insect is also known as the Jamaican click beetle and the 'cucujo' beetle of the West Indies. The immature forms of Phengodidae are called railroad worms.

Organs of Light Production in Insects

Bioluminescence observed in many organisms is mostly attributed to their association with luminous bacteria, but insects have evolved special photogenic organs that produce light. These organs may occur in both sexes, or may be restricted to females and immature forms, particularly in fireflies.

The light organs in insects are situated very close to the body surface behind a window of translucent cuticle. They may be found scattered anywhere from the head to the tip of abdomen, including the thorax. In adult insects, these organs are mostly on the ventral side of the thorax or abdomen. In *Photuris* (Lampyridae), males have two pairs of the light organ while females possess only one pair. Larvae and adult females of railroad worm have 11 pairs of light organs on the lateral sides of the thorax and abdomen and another pair on the head (*Figure* 1) In *Fulgora* (Homoptera) the light organ is situated only on the head. The light organs generally originate from fat bodies, except in *Arachnocampa* (Diptera) where these stem from the enlarged distal ends of malpighian tubules.

Figure 1. The railroad worm Phrixothrix hiatus produces red light from its head and green light along the rest of its body.
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Photogenic organs consist of a large number of specialized cells called *photocytes,* arranged cylindrically at right angles to the translucent cuticle, which permits light to pass through it (*Figure* 2). Behind the photocytes there is a reflecting surface chiefly consisting of urate granules. The photocytes receive oxygen through air tubes or tracheoles. They also contain a heavy aggregation of mitochondria that supplies adenosine triphosphate (ATP) required for the chemical reaction.

Biochemistry of Light Production

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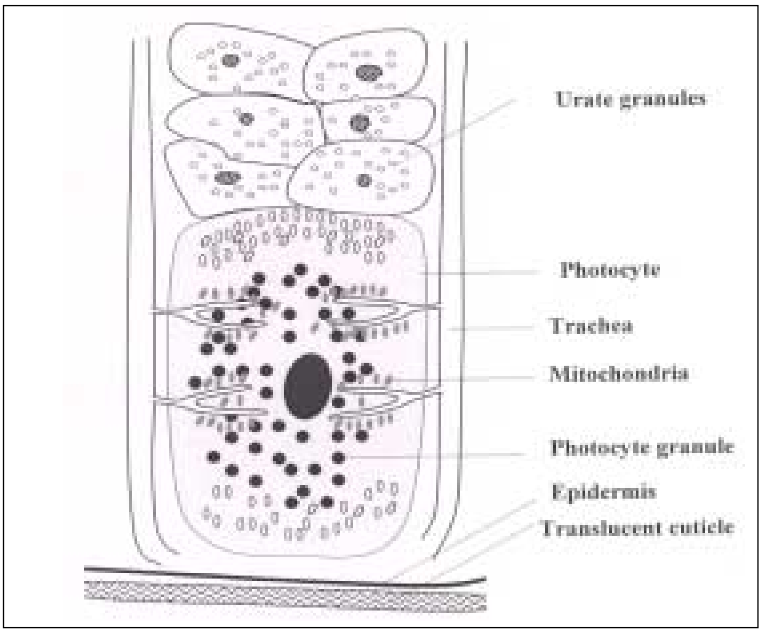

Figure 2. A simplified cross-section of the light organ in insects.

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Raphael Dubois, a French physiologist, demonstrated that three substances are involved in bioluminescence: *luciferin, luciferase* and *molecular oxygen*. Bioluminescence is due to the substrate-enzyme complex of luciferin-luciferase within the cellular cytoplasm. Luciferin refers to any light-emitting compound (*lucifer* means 'light-bringing' in latin). These luciferin-luciferase complexes differ in structure among species. Luciferin and luciferase of the American firefly, *Photinus pyralis*, was isolated in pure crystalline form by McElroy and Seliger at John Hopkins Institute in the early 1960s.

Luciferin, which is secreted by photocytes, is a low molecular weight compound that may be an aldehyde, a polypeptide complex or a protein. Light is produced by the oxidation of luciferin in the presence of the enzyme luciferase (molecular weight 62 kDa) (*Figure* 3). ATP first activates luciferin in the presence of magnesium (Mg^{2+}) and luciferase to produce adenylluciferin, which is then oxidized to form excited oxyluciferin. During the enzymatic oxidation of luciferin large amounts of energy, 40-80 KCal per mole, become available in a single step, so that the product of the reaction is left in a highly excited state. This product emits light when the excited state returns to the ground

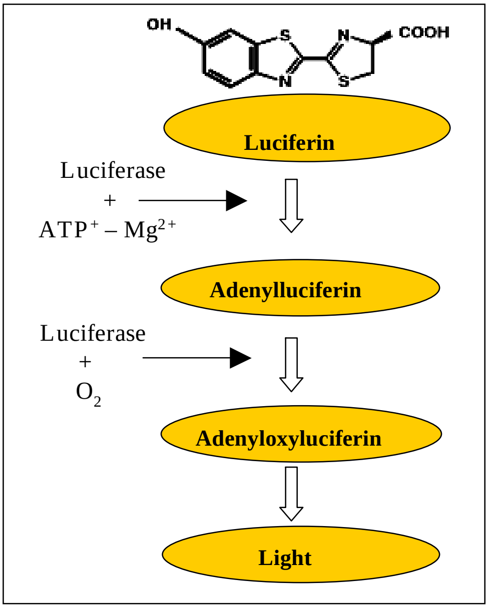

Figure 3. Sequence of events involved in light production in insects.

state. The reaction is very efficient, some 98% of the energy involved being released as light without production of heat. A synaptic fluid near nerve endings, which are chemical intermediaries between nerve and the light organ, triggers the light production.

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Variation in Colour of Light

In most insects the light produced is yellow-green as in *Photinus* and *Lampyris* (Coleoptera). In larval and adult female railroad worms, the light organs on the thorax and abdomen produce green to orange light, while that on the head produces red light (*Figure* 1). Light produced by *Arachnocampa* is blue-green while that of *Fulgora* is white. Thus, the colour of light emitted varies with species and the variation may be due to environmental factors or differences in the structure of luciferase.

Significance of Bioluminescence in Insects

In simple forms of life as in bacteria, bioluminescence is thought to have no functional significance as the light produced is a by-product of certain chemical activity within the cell. However, in many animals, light production has known functional significance. For example, in many arthropods, the light from bioluminescence is used to lure the opposite sex for mating, or it may be used to attract prey, or for defense.

i) *Mating signal:* Light is known to act as a mating signal in fireflies. In certain species the bioluminescence attracts individuals of the same species to aggregate, thus indirectly improving the chances of mating. In some species of Lampyridae, the females are wingless and sedentary; light production is therefore important for them to attract the winged males. Flash patterns in bioluminescent insects vary between species and between sexes. On a relatively cool night, some species wait 5.5 seconds then emit a single short flash. Other species may wait one second and then hold the flash for a full second. Some tropical species congregate in large numbers and flash in unison. Male and female fireflies of *Photuris pyralis* emerge at dusk, In railroad worms the continuous glow of the head region when the larvae are walking, suggests a possible illumination function, whereas the circumstances under which the lateral light organs are switched on suggest a defense function.

emitting a single short flash at regular intervals. The flashes are usually from male fireflies seeking mates. Males outnumber the females fifty to one. Females climb a blade of grass, flashing when males flash within 10–12 feet of the females. Exchange of signals is repeated 5 to 10 times until they start mating.

ii) *Predation:* The most unique example of light acting as a lure for prey is found in the New Zealand glowworm fly, *Arachnocampa luminosa*. The female fly deposits eggs on the ceiling of dark caves. Upon hatching, the larvae hang down by a sticky thread and produce light. During night, the entire cave may glow with this light, attracting other insect species. These attracted insects get entangled in the sticky threads and are preyed upon by the larvae. The caves inhabited by flies are popularly known as 'luminous caves' and are tourist attraction spots in New Zealand.

iii) *Defence:* In railroad worms the continuous glow of the head region when the larvae are walking, suggests a possible illumination function, whereas the circumstances under which the lateral light organs are switched on suggest a defense function. Sudden flashes can repel potential predators. The railroad worm larvae live at high densities, confined to small areas, and may use simultaneous emission to frighten potential enemies, or they may also use the light to intimate the mated females about to lay eggs about over crowding and competition for food sources.

Applications of Bioluminescence

The phenomenon of bioluminescence is utilized as a valuable scientific tool in many biological experiments.

i) *Space Research:* Luciferin and luciferase system can be used in spacecrafts sent to Mars or other planets to explore the existence of lifeforms. The idea is that a special electronic device would pick up soil from the alien surface and mix it with water, oxygen, luciferin and luciferase. Then if a glow were televised back to earth, we could know that ATP, the fifth requirement for light production, occurs there. Even as little as one quadrillionth of a gram of ATP is enough to be sensed by the electronic detectors. The presence of ATP would suggest, in turn the existence of some kind of life in that alien soil that is similar to lifeforms on Earth.

ii) ***Medical research:*** The presence of ATP in every living organism has been exploited in medical research too. Injection of luciferin and luciferase exhibit different reactions in a normal and cancerous cell, and can aid in detecting energy problems in human cells. This technique is now used to study heart ailments, muscular dystrophy, urological problems, etc.

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iii) ***Pest management:*** Bioluminescence is used as a tool for mapping organism distribution patterns. Recently, in 2001, scientists in USA have modified the genetic material of the pink bollworm, an insect pest of cotton, with green fluorescent protein (GFP) derived from the jelly fish, *Aequora victoria*. The GFP transgenic pink bollworm strain fluoresces strongly green when viewed in its larval stage. The objective of this research is two fold; the first is to develop a GFP marked strain of pink bollworm for field performance studies and to map the distribution of the pest. It may also serve as an additional tool for field managers. Their future objective is to eventually add a temperature-sensitive lethal gene along with the GFP gene into the pink bollworm that could be used for its management.

iv) ***Fluorescent marker gene:*** A marker gene is a short sequence of DNA that acts as a label and it is inserted along with a gene of interest into cells. Fluorescent marker genes make the transformed cells glow under light. Genetic markers enable scientists to choose only the cells that have taken up the target genes and discard others.

Suggested Reading

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