BIOLUMINESCENCE AND ITS APPLICATIONS
Sumeet Singh, Sandhya Upreti, Piyali Sarkar
Department of Electrical and Electronics Engineering
PVPIT, Budhgaon

Abstract: Bioluminescence is a chemical reaction caused by an enzyme-catalyzed process: The enzymes that produce light are called luciferin, and those that are the catalyst are called luciferase. The reporter is responsible for this enzymatic activity, which releases bioluminescence energy and gives the bacteria its color. We won’t see this bacteria’s blue-green glow in the ocean though; its reporter protein is visible under a microscope or with a special light recording device. The sensor is how the bacteria is able to continually check for the presence of certain chemicals, determined by the scientists who bioengineer the organisms. It also allows the bacteria to detect the concentration and distribution of pollutants and whether or not they are spreading. Bioluminescence has emerged as an extremely useful and versatile reporter technology. It provides a sensitive, non-destructive, and real-time assay that allows for temporal and spatial measurement. The ability to emit light is dependent on the reducing power of the organism; hence, only metabolically active cells can produce light. The direct relationship between viability and light emission allows the use of bioluminescent bacteria to assess the effect of various chemical, biological, and physical signals. This paper will review about bioluminescence scientific discoveries on bacterial bioluminescence and the current applications of bioluminescence in environmental studies.

Index Terms – Enzymes, Luciferase, Metabolically, Viability

I. Introduction

During Bioluminescence is the production and emission of light by a living organism. It is a form of chemiluminescence. Bioluminescence occurs widely in marine vertebrates and invertebrates, as well as in some fungi, microorganisms including some bioluminescent bacteria and terrestrial invertebrates such as fireflies. In some animals, the light is produced by symbiotic organisms such as Vibrio bacteria. The principal chemical reaction in bioluminescence involves the light-emitting pigment luciferin and the enzyme luciferase, assisted by other proteins such as aequorin in some species. The enzyme catalyzes the oxidation of luciferin. In some species, the type of luciferin requires cofactors such as calcium or magnesium ions, and sometimes also the energy-carrying molecule adenosine triphosphate (ATP). In evolution, luciferins vary little: one in particular, coelenterazine, is found in nine different animal (phyla), though in some of these, the animals obtain it through their diet. Conversely, luciferases vary widely in different species. Bioluminescence has arisen over forty times in evolutionary history.
The chemical reaction that results in bioluminescence requires two unique chemicals: luciferin and either luciferase or photoprotein. Luciferin is the compound that actually produces light. In a chemical reaction, luciferin is called the substrate. The bioluminescent color (yellow in fireflies, greenish in lanternfish) is a result of the arrangement of luciferin molecules.

Some bioluminescent organisms produce (synthesize) luciferin on their own. Dinoflagellates, for instance, bioluminesce in a bluish-green color. Bioluminescent dinoflagellates are a type of plankton—tiny marine organisms that can sometimes cause the surface of the ocean to sparkle at night.

Some bioluminescent organisms do not synthesize luciferin. Instead, they absorb it through other organisms, either as food or in a symbiotic relationship. Some species of midshipman fish, for instance, obtain luciferin through the “seed shrimp” they consume. Many marine animals, such as squid, house bioluminescent bacteria in their light organs. The bacteria and squid have a symbiotic relationship.

Luciferase is an enzyme. An enzyme is a chemical (called a catalyst) that interacts with a substrate to affect the rate of a chemical reaction. The interaction of the luciferase with oxidized (oxygen-added) luciferin creates a byproduct, called oxyluciferin. More importantly, the chemical reaction creates light. Most bioluminescent reactions involve luciferin and luciferase. Some reactions, however, do not involve an enzyme (luciferase). These reactions involve a chemical called a photoprotein. Photoproteins combine with luciferins and oxygen, but need another agent, often anion of the element calcium, to produce light.

Environmental biosensors using bioluminescent bacteria

Environmental biosensors to assess the toxicity of environmental media such as water, soil, and atmosphere have been developed using various recombinant bioluminescent bacteria. Those bacteria were constructed based on specific stress-responsive promoters in bacterial cells. They are thus activated by different groups of toxicity. For continuous monitoring of water toxicity, a multichannel system
having different stress-responsive strains in each channel, and composed of two-stage mini-bioreactors, was successfully developed. Soil toxicity was assessed using a soil biosensor based upon immobilization of recombinant bioluminescent bacteria that worked with the addition of rhamnolipids biosurfactant. An example of phenanthrene toxicity is shown. For the assessment of gas toxicity, an immobilization technique has been set up to allow the biosensor to come in direct contact with the toxic gas in the sensing chamber. An example of benzene toxicity is shown. This mini review will show how the recombinant bioluminescent bacteria can be utilized as environmental biosensors. With further findings and developments of new non-specific stress promoters, the potency and extensiveness of the information that can be obtained using these environmental biosensors is immense.

3. Bacterial Biosensors for Measuring Availability of Environmental Pollutants

The appearance of bioluminescent light varies greatly, depending on the habitat and organism in which it is found. Most marine bioluminescence, for instance, is expressed in the blue-green part of the visible light spectrum. These colors are more easily visible in the deep ocean. Also, most marine organisms are sensitive only to blue-green colors. They are physically unable to process yellow, red, or violet colors.

Most land organisms also exhibit blue-green bioluminescence. However, many glow in the yellow spectrum, including fireflies and the only known land snail to bioluminesce, native to the tropics of Southeast Asia. Few organisms can glow in more than one color. The so-called railroad worm (actually the larva of a beetle) may be the most familiar. The head of the railroad worm glows red, while its body glows green. Different luciferases cause the bioluminescence to be expressed differently.

4. Environmental Biosensors Using Bioluminescent Bacteria

Environmental biosensors to assess the toxicity of environmental media such as water, soil, and atmosphere have been developed using various recombinant bioluminescent bacteria. Those bacteria were constructed based on specific stress-responsive promoters in bacterial cells. They are thus activated by different groups of toxicity. For continuous monitoring of water toxicity, a multichannel system having different stress-responsive strains in each channel, and composed of two-stage mini-bioreactors, was successfully developed. Soil toxicity was assessed using a soil biosensor based upon immobilization of recombinant bioluminescent bacteria that worked with the addition of rhamnolipids biosurfactant. An example of phenanthrene toxicity is shown. For the assessment of gas toxicity, an immobilization technique has been set up to allow the biosensor to come in direct contact with the toxic gas in the sensing chamber. An example of benzene toxicity is shown. This mini review will show how the recombinant bioluminescent bacteria can be utilized as environmental biosensors. With further findings and developments of new non-specific stress promoters, the potency and extensiveness of the information that can be obtained using these environmental biosensors is immense.

5. Bacterial Biosensors for Measuring Availability of Environmental Pollutants

Traditionally, pollution risk assessment is based on the measurement of a pollutant’s total concentration in a sample. The toxicity of a given pollutant in the environment, however, is tightly linked to its bioavailability, which may differ significantly from the total amount. Physico-chemical and biological parameters strongly influence pollutant fate in terms of leaching, sequestration and biodegradation. Bacterial sensor-reporters, which consist of living micro-organisms genetically engineered to produce specific output in response to target chemicals, offer an interesting alternative to monitoring approaches. Bacterial sensor-reporters detect bioavailable and/or bioaccessible compound fractions in samples. Currently, a variety of environmental pollutants can be targeted by specific biosensor-reporters. Although most of such strains are still confined to the lab, several recent reports have demonstrated utility of bacterial sensing-reporting in the field, with method detection limits in the nanomolar range. This review illustrates the
general design principles for bacterial sensor-reporters, presents an overview of the existing biosensor-reporter strains with emphasis on organic compound detection. A specific focus throughout is on the concepts of bioavailability and bioaccessibility, and how bacteria-based sensing-reporting systems can help to improve our basic understanding of the different processes at work.

6. Applications of Bioluminescence

I. 6.1 Biology and medicine

Bioluminescent organisms are a target for many areas of research. Luciferase systems are widely used in genetic engineering as reporter genes, each producing a different colour by fluorescence, and for biomedical research using bioluminescence imaging. For example, the firefly luciferase gene was used as early as 1986 for research using transgenic tobacco plants. Vibrio bacteria symbiose with marine invertebrates such as the Hawaiian bobtail squid are key experimental models for bioluminescence. Bioluminescent destruction is an experimental cancer treatment.

6.2 Green fluorescent protein (GFP)

Green fluorescent protein (GFP), for instance, is a valuable “reporter gene.” Reporter genes are chemicals (genes) that biologists attach to other genes they are studying. GFP reporter genes are easily identified and measured, usually by their fluorescence. This allows scientists to trace and monitor the activity of the studied gene—its expression in a cell, or its interaction with other chemicals.

6.3 Bioluminescence (BRET)

Bioluminescence (BRET)- and fluorescence resonance energy transfer (FRET) techniques have become integral approaches in studies of protein–protein interactions in living cells. They rely on non-radiative transfer of energy between donor and acceptor species that can be appended to the proteins of interest. These techniques display exquisite dependence on distance and orientation between the energy transfer partners. This means they are well suited to measure both small conformational changes in response to ligand binding between partner proteins that remain within a complex or more extensive translocations of proteins between cellular compartments that occur in response to cellular challenge. Introduction of both energy donor and acceptor into a single polypeptide can also allow the detection of ligand-induced conformational switches in monomeric proteins in the millisecond time scale. Many of these approaches are amenable to high throughput screening and the drug discovery process. G protein-coupled receptors (GPCRs) represent a key drug target class. Specific applications of resonance energy transfer techniques to the identification of ligands for this class of protein are highlighted to illustrate general principles.

6.4 Bioluminescence imaging (BLI)

Bioluminescence imaging (BLI) has emerged as a powerful new modality for studies of viral infection and therapy in small animal models. BLI technology captures the light emitted from different luciferase enzymes to detect sites of viral infection and quantify viral replication in the context of a living animal. In this review, we discuss the biochemical features of various luciferase enzymes and modifications to these enzymes that can greatly enhance their ability to image viral infection, host responses and the effects of therapy. We also describe BLI instrumentation and technical aspects of BLI needed to optimize imaging data. Examples of BLI for quantitative analysis of viral infection and in vivo monitoring of antiviral and antibacterial therapy are presented to highlight the potential for BLI to accelerate discovery of new antiviral agents and determine efficacy of antiviral compounds. Ongoing research to use multiple luciferase enzymes to image viral infection, host immune signaling pathways, and cell trafficking in the same animal will continue to advance BLI for longitudinal, real-time quantification and analyses of viral infection and pre-clinical testing of promising therapeutic agents.

7. Conclusions

Bioluminescence have a wide range of applications ranging from clinical through to environmental and agricultural. Biosensors can provide cost-effective, easy-to-use, sensitive and highly accurate detection by using bioluminescent biosensors. This research paper describes the bioluminescence and its variety of research and commercial applications. Lot of research and development is going in this field for instance, could help light city streets and highways. This would reduce the need for electricity. Bioluminescent crops and other plants could luminesce when they needed water or other nutrients,
or when they were ready to be harvested. This would reduce costs for farmers and agribusiness thus increasing the efficiency and quality.

8. References
[1] A course in Electrical and Electronic Measurements and Instrumentation by A.K. Sawhney