



Firefly: Its importance and an inspiration for innovations

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Abstract

Bioluminescence is a unique character of some unique organisms, including fireflies. There are around 2000 species of fireflies worldwide and are belong to the lampyridae family. Luciferase is a protein that is involved in the process of light emission. Firefly larvae exhibit predatory behaviour by eating soft-bodied species such as earthworms and wood lice, which is advantageous from an ecological standpoint. Using luciferase for reporter activities Bioluminescence Imaging Technique was developed. In recent years, firefly luciferase has been employed to produce carrier proteins in the realm of nanomedicine. The firefly's communication mechanism is also used in cutting-edge research domains such as mobile robotics and wireless network design. Various toxicological studies have been undertaken using the firefly luminescence method by various researchers, and these studies have proven to be incredibly useful in monitoring various contaminants and pesticides.

Keywords: bioluminescence, fireflies, lampyridae, luciferase, nanomedicine, carrier proteins, mobile robotics, contaminants, pesticides

Introduction

In nature, a wide range of living organisms with luminescence properties exist which belongs to 700 genera and 16 different phyla (Muthukumaran *et al.* 2014) ^[1]. These bioluminescent organisms commonly found in the sea, on land, and in the air (Campbell 1988; Muthukumaran *et al.* 2014) ^[2, 1]. Beetles are the most diversified and glowing creatures among the bioluminescent organisms (Viviani 2002; Muthukumaran *et al.* 2014) ^[1]. The firefly is a type of beetle that has luminescent behaviour, is nocturnal, and thrives in damp, warm environments (Muthukumaran *et al.* 2014) ^[1]. They can be found in a variety of places, including wetlands, sports and agricultural fields, parks, woodlands, and grasslands (Lewis *et al.* 2020) ^[4].

Around 297 million years ago, beetles evolved (Zhang *et al.* 2018; Lewis *et al.* 2020) ^[5, 4]. Beetles make about 38 percent of all known beetle species on the planet (Stork 2018; Lewis *et al.* 2020) ^[6, 4]. There are more than 2000 species of firefly (Ohba 2004, Lewis & Cratsely, 2008, Lewis 2016, Han *et al.* 2020) ^[7, 4] in the lampyridae family, which is classified into 100 genera (Lewis & Cratsely, 2008; Han *et al.* 2020) ^[4]. Fireflies are most charming insects hence many stories, poems and songs are inspired by them (Han *et al.* 2020). In many countries fireflies are becoming major tourist attraction hence fireflies got economic importance to some extent (Napompeth 2009; Lewis 2016; Lewis *et al.* 2020) ^[4].

Ecological Benefits

Firefly larvae exhibit predatory behaviour, feeding on snails, earthworms, and other soft-bodied organisms (Lewis *et al.* 2020) ^[4]. It was encountered by Nishijima *et al.* that the larvae of the firefly *Luciola parvula* fed wood lice (*Porcellio scaber*), land snails (*Euphaedusa tau tau* and *Pupinella rufa*) or earthworms (*Oligochaeta* spp.) when they molted to 2nd instar larva for about 80 weeks (Nishijima *et al.* 2010).

Understanding of Various Biological Processes Using Firefly Luciferase

Bioluminescence imaging technique (BIT) used firefly luciferase as reporters (Leclerc *et al.* 2000; Maguire *et al.* 2012). Enzymatic behaviour of luciferase is preferably use in various biological reactions due to its numerous beneficial properties such as non-invasive quantification method, rapidity and sensitivity (Choi *et al.* 2003). Leclerc *et al.* 2000 in his study follows gene expression monitoring techniques demonstrated by Castaño *et al.* 1998; Castaño *et al.* 1996; Frawley *et al.* 1994; Núñez *et al.* 1998; and Villalobos *et al.* 1999 to quantify the level of hormonal gene expression as well as monitoring the whole process in breast cancer cells, pituitary cells and neurons. With recent advancements in technology, it is now possible to integrate luciferase reporter activity with the digital imaging microscopy technique, which can aid in the collection of information about single cell gene expression (Leclerc *et al.* 2000).

Firefly luciferase has been extensively employed in research to better understand a variety of biological phenomena (Maguire *et al.* 2012). The use of luciferase in BIT provided critical information on a variety of biological functions, including transcription factor activation, tumorigenesis, and bacterial pathogenicity (Klerk *et al.* 2007; Badr *et al.* 2009; Andreu *et al.* 2010; Maguire *et al.* 2012). Maguire *et al.* discovered a thermo-stable

luciferase from the Italian firefly *Luciola italica*, which produces red and green light and is important *in vivo* mammalian gene expression studies (Maguire *et al.* 2012).

Importance of Firefly in the Field of Nano Medicine

In the field of nanomedicine one of the major achievements is the establishment of carriers which are able to supply sufficient amount of therapeutic payloads to the specific locations (Ali-Boucetta *et al.* 2008; Choudhuri *et al.* 2009; El-Sayed *et al.* 2013). In the whole process of delivering therapeutic payloads, the major task is to monitor the delivery process (El-Sayed *et al.* 2013). Thermo-stable luciferase from *Luciola cruciata* was used to develop a new system for *in vivo* imaging of nanotubules and along with that it was also proved that maintenance of protein activity is possible by appropriate chemical conjugation of luciferase of *Luciola cruciata* to nanotubules (El-Sayed *et al.* 2013). It was also proved by El-Sayed *et al.* that doxorubicin which is an anticancer agent when loaded with luciferase of *Luciola cruciata* and nanotubules did not alter the property of luciferase of *Luciola cruciata* (El-Sayed *et al.* 2013). This newly developed luciferase- nanotubule consisting generic *in vivo* imaging technique shows many advantages for monitoring various drug delivery systems related to nanomedicine.

Use of Firefly Communication Technique in Technological Innovations

Intelligence of large flying group of insects which are known as swarm is very high and this intelligence of swarms provides a wide range of ideas to solve various problems (Banu *et al.* 2017). Glowing phenomenon of firefly is considered as continuous research field which has various aspects *viz.*, biochemical and social (Lewis and Cratsley, 2008; Fraga, 2008; Banu *et al.* 2017) [8]. Following the mechanism of communication between/among the fireflies using luminescent light, similar techniques are implemented in some highly advanced fields of research like mobile robotics (Krishnanand and Ghose, 2006; Banu *et al.* 2017) and wireless network design (Leidenfrost and Elmenrieche, 2008; Banu *et al.* 2017). Inspired by behaviour of firefly, fuzzy-firefly clustering application was developed to solve the gene clustering problems which follows the algorithm of optimization-based platform and this application was used to study pattern of gene expression (Banu *et al.* 2017).

Use of Firefly Luciferase in Toxicological Studies

Various studies on polyhalogenated diphenyls, polycyclic aromatic hydrocarbons, dioxins, phenols, and pesticides in microorganisms were conducted using the bioluminescence method (Reid *et al.* 1998; Min *et al.* 2003; Valat *et al.* 2003; Kudryasheva *et al.* 2002; Farré *et al.* 2002; Trajkovska *et al.* 2005). The most important enzyme in the firefly bioluminescence method is luciferase, and this firefly luminescence method can be used to monitor pollutant influence (e.g., pesticides) (Trajkovska *et al.* 2005). Trajkovska *et al.* investigated the effects of fenoxaprop-p-ethyl, diclofop-methyl, and metsulfuron methyl, as well as the impact of their degradation products on the firefly luciferase enzyme and the assessment of variation based on the bioluminescence vs. time graph (Trajkovska *et al.* 2005).

Conclusion

There are still many unexplored areas in firefly research. The Indian firefly, *Luciola praeusta*, was similarly given little attention by academics working in the same subject. As a result, there are many more opportunities to characterise the diverse functionalities of numerous unknown firefly species. Fireflies are undoubtedly intelligent organisms, and their intelligence has already been used to develop various advanced accessories in the fields of science and technology, despite the fact that the majority of firefly species are still in the research stage or have yet to enter the research circle. The population distribution of this magnificent organism all over the world is significantly influenced by increasing urbanisation and numerous development activities, so protecting this species from extinction necessitated certain immediate actions. If no actions are taken now to preserve such an organism, it will exist only in photos in the future.

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