

**Naturally fluorescent field mouse, *Mus booduga* (Gray, 1837) and common house gecko, *Hemidactylus frenatus* (Schlegel, 1836) as model organisms for biomedical studies**

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**Abstract**

Since the discovery of the Green fluorescent protein from crystal jellyfish *Aequorea victoria*, many fluorescent organisms have been identified from different parts of the world. As a result, various fluorescent proteins such as GFP, RFP, and DsRed are discovered and widely used in genetic engineering studies to label a wide range of proteins and genes in mouse models. In bioimaging research, a variety of mouse models are used to visualize fluorescence-based phenotypic variations. In this study, the field mouse, *Mus booduga*, and common house gecko *Hemidactylus frenatus* displayed natural blue fluorescence (440-480 nm). The fur of *M. booduga* and bones of *H. frenatus* have displayed blue fluorescence. These organisms may serve as a naturally fluorescent models in bioimaging studies to study phenotypic changes triggered by physico-chemical factors. This study infers that implication of fluorescent terrestrial chordates in evolutionary studies would also delineate the origins (ocean to land) and phylogenetic spread pathways of fluorescent trait in eukaryotes.

**Keywords:** Fluorescent proteins; Fur; Muridae; Gekkonidae; Evolution.

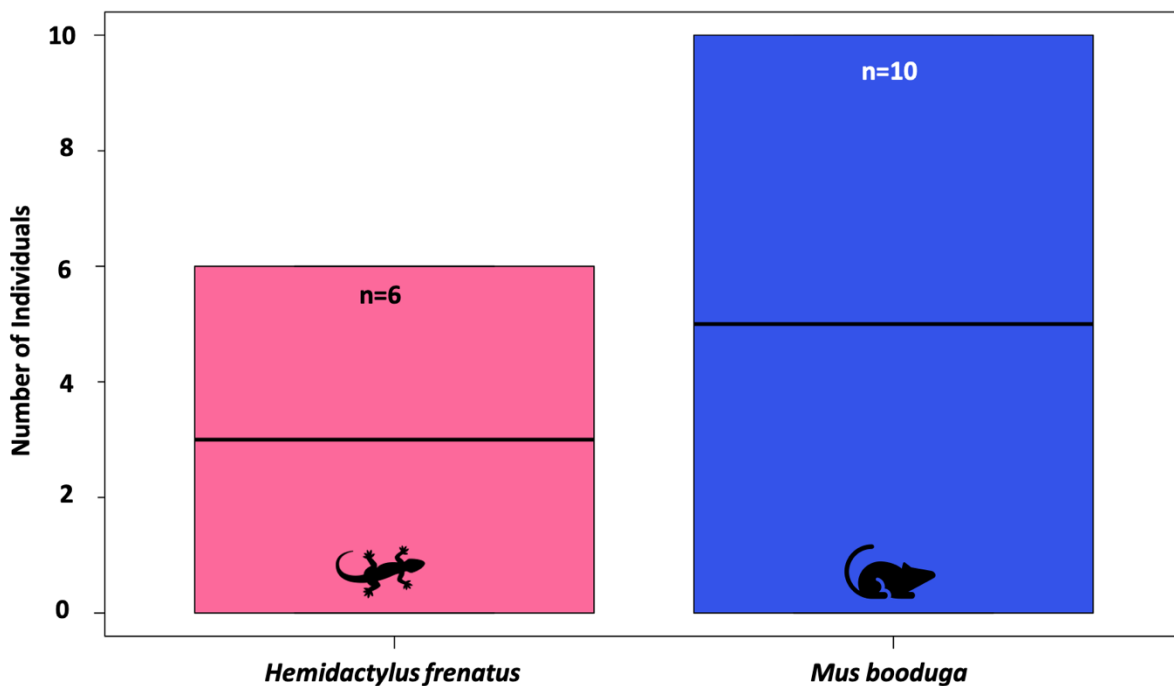
Fluorescence trait is a biological phenomenon where the specific fluorescent body tissues, bones, and fur of different organisms absorb the short-wavelength light and reemit it into a high wavelength [1]. This phenomenon is known to originate from the ocean and spread to environments like polar regions, terrestrial and marine milieus. Organisms with this spectacular trait are widespread in various phylogenetic lineages, but their occurrence is not commonly encountered. The first-ever known fluorescent protein was the green fluorescent protein (GFP) extracted from the bioluminescent jellyfish, *Aequorea victoria* [2]. Blue-green and red fluorescent organisms are very commonly observed in nature [1]. In general, based on the current literature, fluorescence trait is known to function as intraspecific signaling, camouflage, mate attraction, anti-predatory, UV reflectors [1], and evolutionary time signatures to study the fossils [3, 4]. Fluorescent organisms have ecological, evolutionary, and multifaceted biomedical importance. Therefore, current researchers have focused on discovering fluorescent organisms to develop fluorescence-based toxicological assays and biomedical applications.

Among the numerous known transgenic and wild model organisms in genetic engineering studies [5, 6], the wild mice *Mus* are widely used in gene expression research to study the propagation of various diseases, phenotypic and genotypic variations, evolutionary aspects, animal behavior, and therapeutic applications [7, 8]. In the current research, visualization of cell-cell interactions has become easy by transfecting the targeted genes with combination of fluorescent proteins such as GFP and DsRed. In contrast, the use of Gecko's as a space model is gaining interest in space research [9]. Together, reptiles and rodents are recognized as an important biomedical and space research models to study the phenotypic variations caused by stress and drugs as well as genotypic changes through bioimaging studies. In the present study, while searching fluorescent organisms during nighttime, a blue fluorescent gecko and mouse were observed from coastal regions of India. These two organisms are the first-ever recorded fluorescent genera globally and are being reported for the first time from India.

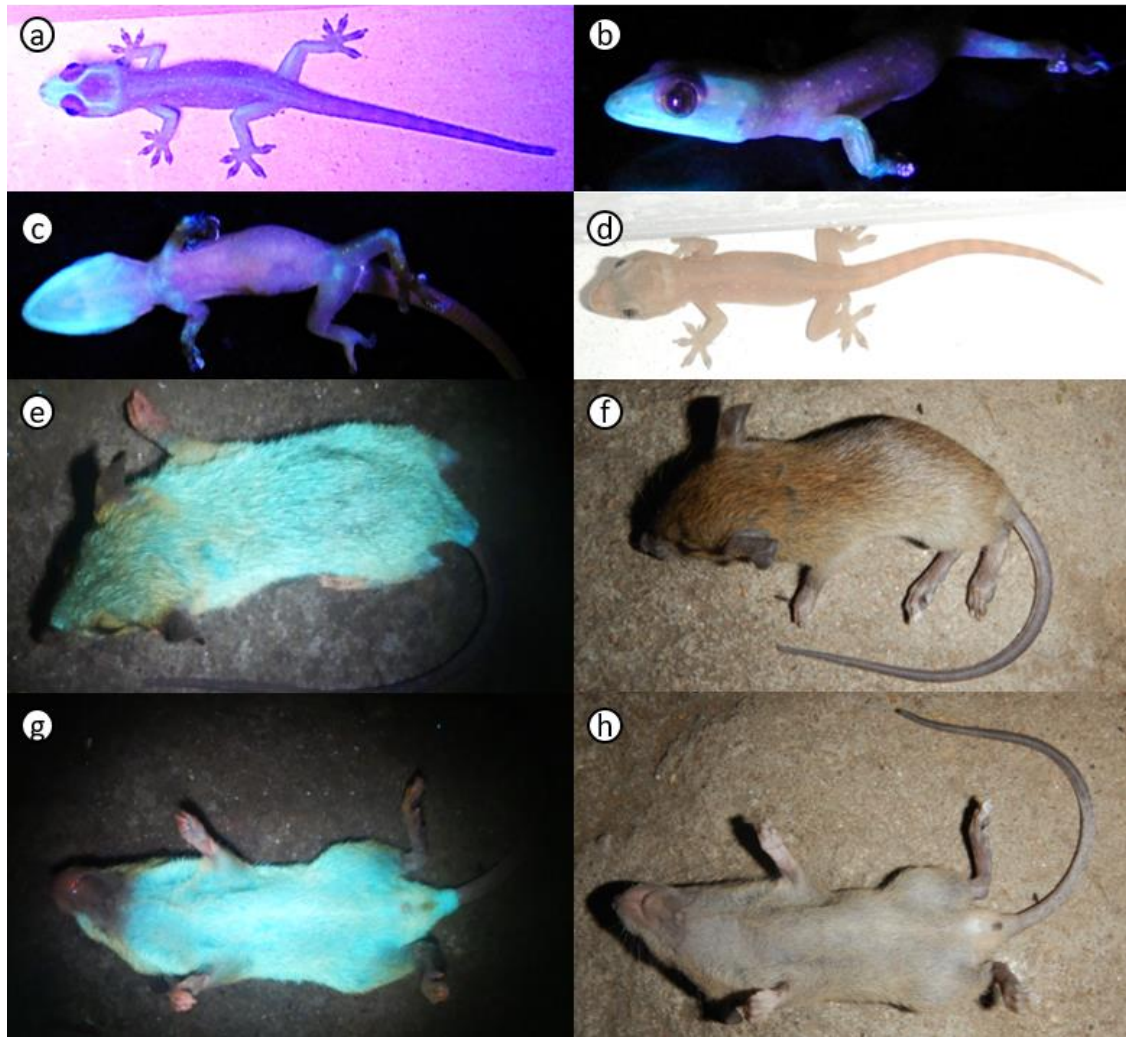
Live specimens of common house gecko *Hemidactylus frenatus* were investigated during night from Dona Paula (15°27'36"N; 73°48'30"E), Goa, the southwest coastal region and Barrankula (15°53'46"N; 80°57'22"E), Andhra Pradesh, southeast coastal region. Dead specimens of the little Indian field mouse, *Mus booduga* specimens were also observed in the nighttime near a rice grain storeroom in Barrankula (15°53'46"N; 80°57'22"E). A UV-Blue flashlight, amiciVision 21 LED UV Torch Light, was used to test the fluorescence from these specimens. Fluorescence of these specimens was captured in a dark conditions after exposing the specimens briefly (20 to 40 seconds) to a high-energy short-wavelength (395-400 nm). None of these specimens were harmed nor collected in this

study. Natural illumination and fluorescent photographs from live specimens of *H. frenatus* and dead specimens of *M. booduga* were captured using the NIKON Coolpix camera with yellow filter UV glasses. Dorsal, lateral, and ventral images of live *H. frenatus* specimens were captured at 60° angle while crossing a glass platform.

Blue fluorescent organisms in the family Muridae have not been known before. In this study, fluorescent blue fur was observed in ten specimens of the little Indian field mouse, *Mus booduga*. Also, a putative bone-based blue fluorescence was observed in both juvenile and adult specimens of common house gecko *Hemidactylus frenatus* (a total of six individuals). These two fluorescent species are being reported for the first time in this study. The fur of *M. booduga* showed blue-green fluorescence and foot pale orange-pink fluorescence. The skull of *H. frenatus* displayed strong blue fluorescence than other body bones (Fig. 1&2). Eggs of *H. frenatus* did not show fluorescence but the remnants of egg fluids displayed blue-green fluorescence. Epidermis and other body tissues of *M. booduga* and *H. frenatus*, did not show any fluorescence. Juveniles and adult specimens of *M. booduga* and *H. frenatus* displayed fluorescence. Blue fluorescence in these animals is likely to fall between the spectral wavelength range of 440 to 480 nm. Both the specimens are commonly found in the coastal areas as well as mainland regions.



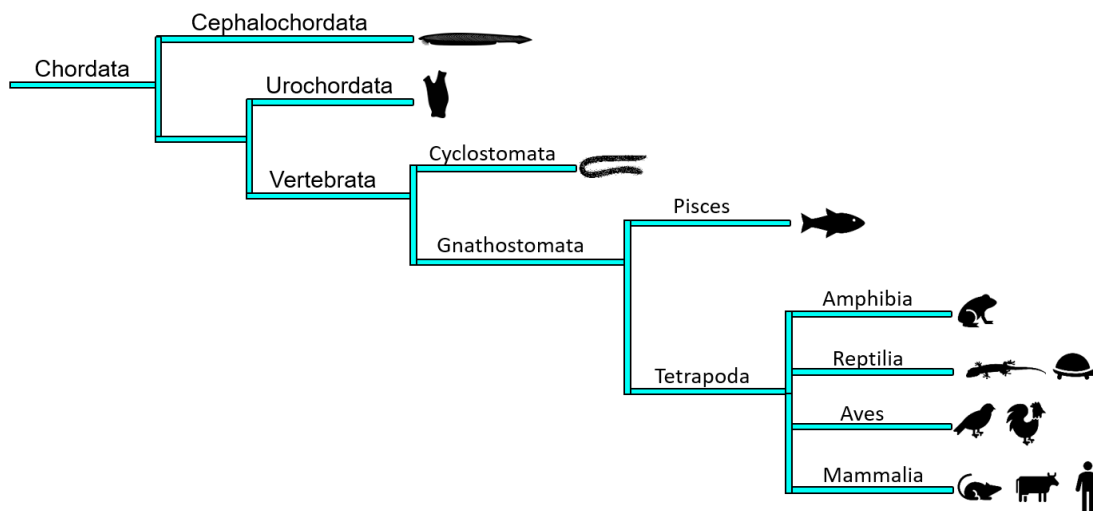
**Fig. 1.** The number of fluorescent specimens investigated for each species.



**Fig. 2.** Blue fluorescence observed in a gecko and a mouse. **a)** dorsal, **b)** lateral, and **c)** ventral views of bone-based fluorescence observed in *H. frenatus*; **d)** *H. frenatus* under visible light. Fluorescent fur of *Mus booduga* in dorsal (**e**) and ventral views (**g**), and the same specimens under visible light (**f** & **h**).

Fluorescence trait is a widespread phenomenon in evolution but uncommon in many organisms. Chordate groups are known to comprise at least a single fluorescent organism in their lineages, indicating the evolutionary importance of fluorescence trait (Fig. 3). Fur-based natural fluorescence has been reported in mammals such as Didelphidae marsupials [10], squirrel, *Glaucomys* spp. [11], platypus, *Ornithorhynchus anatinus* [12], and springhare, *Pedetes surdaster* [13]. However, the function of fluorescent fur in mammals is remain unknown. In springhare, *P. surdaster*, fluorescence is hypothesized to involve in camouflage function [13]. In the case of squirrel *Sciurus niger*, fluorescence is observed in bones [3] but not fur [11]. The squirrel, *Glaucomys* spp., was hypothesized to utilize fluorescence for intraspecies specific communication, anti-predatory, and mate selection functions [11]. The little Indian field mouse *Mus booduga* is a widespread rodent found abundantly during the post-monsoon season [14]. *M. booduga* live specimens are active during the night for

foraging, and very often, they are found in grain storerooms. However, the function of the fluorescent fur of *M. booduga* is not yet known. In India ten species of the genus *Mus* were known [15], but the occurrence of fluorescence in other species has not yet been reported. Thus, further field studies and laboratory experiments are required to understand fluorescence phenomenon in Rodentia. The origin of fluorescence in the family Muridae might have arisen either independently or through dietary acquisition of terrestrial or coastal organisms. It is easier to track independently originated fluorescence trait in these organisms through molecular approach. While, tracking the dietary originated fluorescence is not an easier task in many cases. A recent study gives some hypothetical clues that the omnivore Keen’s mouse, *Peromyscus keeni* diet was comprised of 35–37% beach dwelling arthropods [16]. Unfortunately, Davidson et al. (2021) did not investigate the presence of fluorescence in this species. Thus, it is a missing piece of detail to understand fluorescence occurrence in mouse specimens feeding on marine organisms. Majority of the Arthropods bear fluorescence and their trophic interactions would give many new insights on the dietary flow fluorescent molecules. Thus, to confirm the dietary originated path, an arduous trophic interactions study need to be conducted.



**Fig. 3.** Fluorescence in Chordata. At least few species from each chordate groups are known to be fluorescent. Blue color indicates the evolutionary spread of fluorescence in various chordate groups (Image created based on the literature reviewed by Ramesh, 2021).

Fluorescence in reptiles occurs through two main mechanisms: (1) bone-based fluorescence [17, 18] and (2) dermal fluorescence [19]. Bone-based fluorescence have been recorded in recent times from terrestrial reptiles includes house gecko, *Hemidactylus parvimaclulatus* [20], bibron’s gecko, *Chondrodactylus bibronii* [18] and bent-toed Gecko, *Cyrtodactylus quadrivirgatus* [21]. A recent study observed the iridophores mediated and guanine crystals dependent neon-green dermal

fluorescence in the desert gecko *Pachydactylus rangei*, which is postulated to function as an intraspecific visual signaling mechanism under moonlight [19]. In all the fluorescent gecko specimens, fluorescence is presumed to act as UV reflectors or signaling trait as anti-predator and mate attractants [22]. Another possible function of fluorescence is prey attractant, as observed in an experiment on the marine hydromedusa [23].

However, except the known and presumed functions, the actual ecological function of dorsoventral fluorescence in *H. frenatus* and *M. booduga* remains unknown. The persistence of blue fluorescence throughout their life stages needs to be investigated through histological aspects. The lack of experimental studies on various fluorescent organisms has limited our knowledge of fluorescence functions in different organisms. Thus, further studies on these animals' behavior in presence and absence of fluorescence bait or predator and their interactions with other fluorescent and non-fluorescent organisms would give many clues on prey-predatory interactions and mate attraction.

The first identification of fluorescent mouse *M. booduga* might serve as a potential wild fluorescent model in bioimaging research to study phenotypic variations (e.g., hair morphology, epidermal changes) during clinical trials. Bone based fluorescence may also help to understand gradual development of bone. Likewise, both, mouse and geckos may also be utilized in space research to study the adaptations during spaceflight. Therefore, further histological aspects, protein analysis, and genetic studies are required to understand the evolution of fluorescent proteins in Muridae and Gekkonidae. Extending research on fluorescent organisms from marine and terrestrial organisms would help us to understand the convergent and divergent evolution processes. Further studies on biogeographic, ecological, anatomical, and molecular mechanisms of fluorescent mouse and gecko are needed to address the evolutionary origin and functions of fluorescence in these organisms.

### **Conflict of interest**

The author declares no competing interests.

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