

FOURIER ANALYSIS OF FIREFLY PULSES AT LOW TEMPERATURE AND UNDER A STRONG STATIC MAGNETIC FIELD

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Abstract: Quite a few scientific investigations have been carried out on the flashing of the firefly till now. Here we analyse the Fourier spectra of three species of fireflies—two Japanese species *Luciola cruciata* and *Luciola lateralis*, and one Indian species *Luciola praeusta*—in control and under a strong static magnetic field. The number of harmonic contents on the Fourier transform plots is found to decrease under the high magnetic field, which points towards a certain type of filtering of pulses. This filtering can be compared with a typical bandstop filter called notch filter. In order to compare with a similar type of phenomenon, a Fourier spectrum of the flashes from the Indian firefly-species at a low temperature is presented.

Keywords: Firefly Flash, Strong static magnetic field, Fourier spectra, Harmonics, Low temperature.

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1 Introduction

Bioluminescence is an interesting phenomenon utilised by certain living organisms such as the firefly where luciferin starts the chemical reaction catalysed by luciferase, an enzyme present inside the firefly lantern, in the presence of O_2 , ATP and Mg^{2+} to yield an electronically excited oxyluciferin species. Visible light is emitted as the oxyluciferin decays to the ground state. This reaction, taking place in the ventral layer of the firefly light-producing organ, is called the chemiluminescence reaction. The importance of firefly bioluminescence lies in the high quantum yield value.

Throughout the world, especially in the tropics, there are more than two thousand species of the glow-worm family *Lampyridae* to which fireflies belong. Numerous scientific studies have been carried out on the flashing of fireflies. Some of the aspects of firefly flashing studied in recent times are: influences by calcium [1], nitric oxide vapours [2], gating of oxygen to light-emitting cells [3], geographic locations [4, 5], temperature variations [5]-[7], and static and pulsed magnetic fields [8]-[10]. The flash duration of *Luciola parvula* female fireflies is measured to be longer than that of males of the species [11], and *Photinus carolinus* female's response to the conspecific male flash pattern is noticed to be a two-peaked (doublet) courtship flash [12]. It is shown that flashes from the Indian species *Luciola praeusta* are composed of a large number of microsecond-duration pulses [13], and that these pulses are manifestations of an oscillating chemical reaction, like the Belousov-Zhabotinsky reaction [14]. The amplitude of the continuous train of triangular pulses is apparently altered in accordance with the instantaneous values of a hypothetical signal [15]. Inspired by the flashing behaviour of fireflies, a metaheuristic algorithm, called the firefly algorithm (FFA), has been developed [16]. Using the FFA, solutions to harmonic minimisation and total harmonic distortion problems for the multilevel inverter, a power electronic device, have been proposed [17, 18].





Figure 1: Schematic diagram of the arrangement for recording firefly flashes at low temperatures.

Investigations of *in vivo* flashes of three species of fireflies, two Japanese species *Luciola cruciata*, *Luciola lateralis* and one Indian species *Luciola praeusta*, positioned under a strong static magnetic field of strength 10 Tesla for a long time, showed that the flashes in general became more rapid, and occasionally overlapped to produce broad compound flashes [10]. It was proposed that the Lorentz force, induced by the strong static magnetic field, decelerated the velocity of the nerve action potential when both the directions of the nerve conduction and magnetic field were crossed, and a slight decrease in the velocity of nerve conduction might be amplified in the bioluminescence process. As it was observed that a firefly emits flashes a little bit more rapidly when stimulated by stirring the sample-fixing plate, terms like 'magnetic stimulation', 'magnetic sense' and 'oxidative stress' were put forward for this phenomenon [10]. It could be mentioned here that in an earlier study, static magnetic fields up to 14 T were found to affect the emission intensity and spectrum of the bioluminescence of the luciferin-luciferase system both *in vivo* and *in vitro* when a firefly was in a steady emission state [8]. Very recently, it was reported that the flashes obtained from specimens of firefly *L. praeusta* at temperatures below 20 °C resemble those under a strong static magnetic field [7].

In this paper, Fourier spectra of the flashes under a strong static magnetic field vis-á-vis those in control are presented. An analogy is drawn with the frequency domain-flashes obtained at a low temperature of 16 °C. The simple philosophy behind Fourier transform is that almost every imaginable signal can be broken down into a combination of simple waves. A real signal is often made up of many frequencies, and the Fourier transform is a tool that tells us which frequency a signal consists of and to what intensity.







Figure 2: Flashing of the firefly species Luciola cruciata. (a) Spectrum in the time domain. The average duration of a flash of this species has been reported to be 1.6 s. (b) Fourier transform of the plot in (a). (c) Rapid flashing under the static magnetic field of strength 10 T. (d) Fourier transform of the plot in (c).

2 Materials and Methods

Experimental details of the study in the magnetic field have been given elsewhere [10]. Those are as follows: The firefly was kept fixed with a transparent tape at the end of a fiber of length approximately 5 m. The lantern of the firefly faced the entrance face of the fiber. This face was put inside a black polythene cover, so that no light could enter the fiber. The other end of the fiber was kept in front of a Hamamatsu photomultiplier tube (E10679), which received and amplified the optical signal. A parallel combination of resistor-capacitor, having values 1 k Ω -0.1 μ F, was connected between the anode and ground of the PMT. The output waveforms were displayed by an Agilent digital storage oscilloscope (1024A). For recording flashes under the magnetic field, the firefly-attached face of the fiber is attached to a meter-long wooden scale which was inserted at the centre of an OXFORD superconducting magnet. The magnet, 480 mm long with a bore of 50 mm in diameter, produced 10 T at its centre. The insect was positioned at the center of the magnetic field, which was homogeneous at that location. The angle between the firefly and the strong magnetic field was about 20°. Temperature and relative humidity in the magnet were 25 °C and 40–50%, respectively. Experimental details for the low-temperature dependence study could be found in the paper by Sharma et. al. [7]. Figure 1 shows the schematic diagram of the arrangement. A USB device recorded the waveforms in both cases. Origin 8.0 was used for our analysis. A total of twenty-five Fourier plots for the flashes under the magnetic field, and twenty plots at the low temperature were analysed.





Figure 3: Flashing of the firefly species Luciola lateralis. (a) Spectrum in the time domain. The average duration of a flash from this species has been reported to be 400 ms. (b) Fourier transform of the plot in (a). (c) Spectra under the static magnetic field of strength 10 T. (d) Fourier transform of the plot in (c).

3 Results and Discussion

A spectrum of the firefly species *L. cruciata* in control, along with its Fourier transform, are shown in Figures 2 (a, b), while that of the same under the 10 T magnetic field with its Fourier transform are shown in Figures 2 (c, d). Figures 3 (a, b, c, d) and 4 (a, b, c, d) present these plots for the species *L. lateralis* and *L. praeusta*, respectively. It is evident that under the static magnetic field a few harmonics, which were present in the control state, disappear. Disappearances of the harmonics are found to occur in regular intervals, which can be assigned as particular bands. Fourier analysis of the spectra in control and under the 10 T magnetic field is presented in Table 1. Thus, under the magnetic field, a stopband appears for all firefly species. The bandwidth of this stopband is different for different species. Hence the result shows analogy with multiple notch filters which is required to get rid of more than one frequency. A notch filter highly attenuates/eliminates a particular frequency component from the input signal spectrum while leaving the amplitude of the other frequencies relatively unchanged [19]. Flashes generated at 16 °C from a specimen of the species *L. praeusta* with the Fourier transform are presented in figure 5. Disappearance of harmonics is discernible in this figure.





Figure 4: Flashing of the firefly species Luciola praeusta. (a) Spectrum in the time domain. The average duration of a flash from this species has been reported to be 100 ms. (b) Fourier transform of the plot in (a). (c) Spectra under the static magnetic field of strength 10 T. (d) Fourier transform of the plot in (c).





Figure 5: Flashing of the firefly species *Luciola praeusta* at 16 °C. (a) Spectrum in the time domain shows marked increase in the flash duration. (b) Fourier transform of the plot in (a) reveals similarity to the ones in Figures 2(d), 3(d), 4(d).

It was already observed that low temperatures had nearly similar effects on the firefly flashes as that of the high-intensity static magnetic field, and hence the inference that the strong static magnetic field possibly affected the body temperature of the firefly [7]. In fact, it was reported that strong static magnetic fields decrease the skin temperature of an animal [20]. It is worth mentioning here that among the techniques developed for modulation and control for multilevel converters, selective harmonic elimination has been the most widely used one.

It is well known that the firefly lantern is composed of two layers out of which the ventral layer is the main storehouse of the light-producing element. The ventral layer consists of photocytes where mitochondria are believed to act as 'gatekeepers', absorbing oxygen from the tracheal system to prevent the light reaction from occurring in excess. Mitochondria are the main source of ultra-weak chemiluminescence generated by reactive oxygen species, which are continuously formed during the mitochondrial oxidative metabolism. It has been speculated that mitochondria could be responsible for producing triangular pulses emitted by fireflies [15]. This capacitor-like action of mitochondria could also be responsible for filtering certain pulses, as we know that any filter consists of a capacitor-resistor combination. Once a lantern nerve fires, a series of chemical events occurs that allow oxygen to reach the peroxisomes and initiate the light reaction. The magnetic field affects this oxygen molecule transport in/around the firefly's light-emitting organ, resulting in the filtering action. It was inferred that diamagnetic torque forces, which were induced by the 10 T order of the static magnetic field, had an inhibitory effect on the biophotochemical process of the firefly light organ [9]. The stopband at the wavelength of 270–290 nm with almost unity reflectance ($R \approx 100\%$) and no amplification (a = 0), showed by an assumed filamentous mitochondria in the respiring state [21], also supports our analysis.

PANE Journal of Physics



4 Conclusion

It was speculated that the rapid flashing of the firefly probably occurred due to the influence of the strong static magnetic field on the neural activity of the spent-up fireflies. A similar argument was put forward for flashing at low temperatures. Hence we could conclude that bursts of neural activity releasing octopamine - widely believed to be the neurotransmitter responsible for the induction of luminescence in the firefly light organ - produce blocking of certain frequencies and allowing of other frequencies. Similar investigations on a few other species of fireflies would consolidate this conclusion. Effects of the magnetic field and low temperature on mitochondrial action should be further studied in the context of the design of notch filters.

Species	Harmonics in	Harmonics under
	control (Hz)	10 T field (Hz)
Luciola	0.42	0.47±0.02
cruciata	0.89±0.01	
(n=10)	1.36±0.01	
	1.75±0.01	1.75±0.01
	2.76±0.04	
	3.12±0.06	
Luciola	0.73±0.02	
lateralis	1.49±0.03	
(n=10)	2.01±0.04	
	2.77±0.03	2.75±0.04
	3.46±0.07	
	4.13±0.06	
	4.91±0.09	
	5.58±0.10	5.5±0.05
	6.10±0.11	
	7.12±0.13	
Luciola	2.04±0.03	
praeusta	4.80±0.20	
(n=5)	6.93±0.30	6.51±0.28
	9.70±0.43	
	11.83±0.52	
	13.83±0.40	13.11±0.31

Table 1: Average values of Fourier frequencies at which harmonics are present in the cases of the three species of fireflies in control and under the strong static magnetic field. Values imply deviations from the peak frequency assigned as 0 Hz. Deviations less than 0.01 Hz from the mean values are not tabulated.

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