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Research Article

Sequences of Luciferase Enzyme from Extracted 25 Organisms to Create Database using WAMP Software

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Abstract

Luciferases are the enzymes that catalyze the reactions producing light. The oxidative mechanism which leads to light emission is similar for most luciferases. However, these enzymes and their substrates are evolutionarily unrelated. The firefly enzyme luciferase catalyzes the luminescent reaction of luciferin with ATP and oxygen. This substance is only slowly transported through cell membranes, in contrast to the aldehyde substrate in the bacterial reaction. Therefore the bacterial luciferase system seems more suitable to the study of environmental or developmental changes in gene expression in living cells.The property of bioluminescence identified in various bacteria, fireflies and insects has been illustrated in detail in this database. The sequences of luciferase enzyme from 25 organisms have been extracted from databases in Genbank and fasta format. Those sequences were analyzed for its physical and chemical parameters and also for identifying homologous sequences. The databases have been created using WAMP software which includes Windows, Apache, MySQL, PHP.

Keywords: Luciferase; Database; WAMP; Sequence; Enzyme; PHP.

Introduction

Luciferase is an enzyme that transforms chemical signal into light, therefore we suppose that high sensitivity of modern photomultipliers (PMT) and avalanche photodiodes (APD) allows detecting in principal a few molecules into the probe. But the realization of this limit into luciferase biosensor needs more smart electronic amplification [1-3]. Bioluminescence is the emission of visible light by biological systems, which arises from enzyme-catalyzed chemical reactions. Bioluminescence occurs widely in marine vertebrates and invertebrates, as well as in some fungi, microorganisms and terrestrial invertebrates. Some symbiotic organisms carried within larger organisms produce light [4,5].

Bioluminescence can be distinguished from chemiluminescence in that it occurs in living organisms and requires an enzyme catalyst. These chemical-dependent emissions of light differ from fluorescence and phosphorescence[6-8], which involve the absorption of light by a compound followed by emission of light at a lower energy (higher wavelength) from the excited state of the molecule [9-11]. The excited molecule produced during bioluminescence reactions, however, is analogous to that produced during fluorescence, and consequently the luminescence emission spectrum can often be related to the fluorescence emission spectrum [12-15]. As a general approach, light intensity of firefly bioluminescence is correlated to the chemical concentrations of the reaction components. When configured properly, the light intensity can be used to associate an observable parameter with a molecular process. About 2500 species are currently described to have luciferase many more are probably hidden in the rapidly shrinking tropical forests around the world [16].

The classification of luciferase consists of Bacterial Luciferase, Insect luciferase and other luciferase. Other luciferases that have been introduced as candidates for genetic reporters include click beetle, *Gaussia*, *Metridia*, and *Vargula* luciferases [17-20]. Previously, luciferases were available from a few different species; the most popular one was lucifers from Photinus pyralis. Today, one can choose from a variety of recombinant luciferases with different properties. The database can be even design your own luciferase meeting your specific specifications. You must then consider which

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properties are the most important for a particular application.

In this study luciferase enzyme screening of the organisms were carried out to database creation identify homologous sequences and modeling. The aim of the study was to database, generate physical and chemical parameters analyzed using ProtParam. ProtParam computes various physico-chemical properties that can be deduced from a protein sequence. The parameters computed by ProtParam include the molecular weight, theoretical pI, amino acid composition, atomic composition, extinction coefficient. You must then consider which properties are the most important for a particular application.

Materials and methods

Data collection

Figure 1 represents the different sources of data collection of luciferase enzyme.

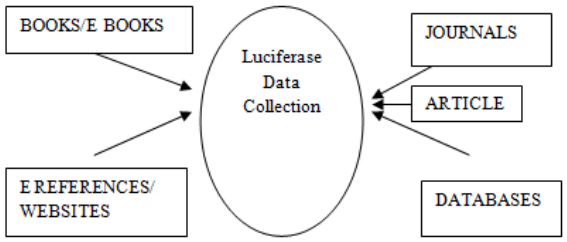


Figure 1. Luciferase enzyme sequences data collection

Retrieved of protein sequence

The retrieved of protein sequence extraction of the Genbank and fasta sequences of exclusively twenty five luciferase enzyme producing organisms from NCBI (National Centre for Biotechnology Information) database.

Computation of physico-chemical properties

EXPASY is a Bioinformatics resource portal operated by the Swiss Institute of Bioinformatics (SIB) and in particular the SIB Web Team. It is an extensible and integrative portal accessing many scientific resources, databases and software tools in different areas of life sciences. EXPASY under the ProtParam computes various physico-chemical properties that can be deduced from a protein sequence.

BLAST analysis

In Bioinformatics, Basic Local Alignment Search Tool, or BLAST, is an algorithm for comparing primary biological sequence information, such as the amino-acid sequences of different proteins or the nucleotides of DNA sequences. A BLAST search enables a researcher to compare a query sequence to a library or database of sequences, and identify library sequences that resemble the query sequence above a certain threshold.

Creation of database

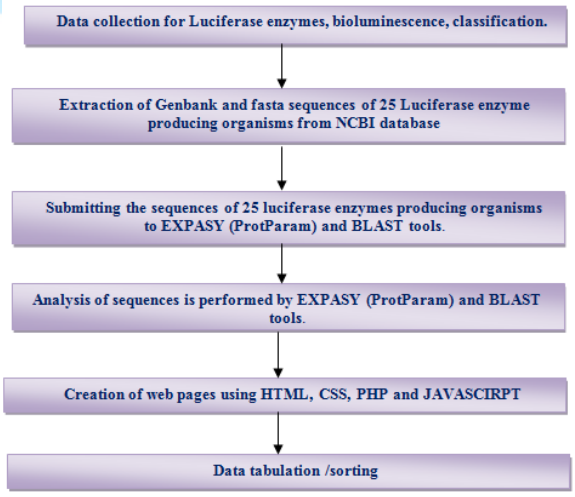
The databases created by using WAMP (Windows, Apache, MySQL, PHP). WAMP is a form of mini-server that can run on almost any Windows Operating System. The following materials required are,

1. SOFTWARE
   a) WAMP (Used version 2.4)
      Windows Xp
      APACHE (WAMP uses APACHE version 2.4.4 and is Windows compatible)
      MYSQL (WAMP uses MYSQL version 5.6.12 and is Windows compatible)
      PHP (WAMP uses PHP version 5.4.16 and is Windows compatible)
      Php MyAdmin (WAMP uses PHP version 4.0.4 and is Windows compatible)

PHP is a mainly widely-used general-purpose scripting language that is especially suited for web development and can be embedded into HTML. It's composed of data collection documents written in Hypertext Markup Language which is a huge repository of formless data.

Methodology

Multistep approach used for the database development on luciferase enzyme is described in scheme 1.



Scheme 1. Multistep approach in the database development on luciferase enzyme

Result and discussions

Luciferase has the user access page where the existing users can Sign in using his/her EMAIL ID and password. New users can Sign up by clicking on the "Sign Up link" provided as shown in figure 2-4.

Figure 2. Database of luciferase

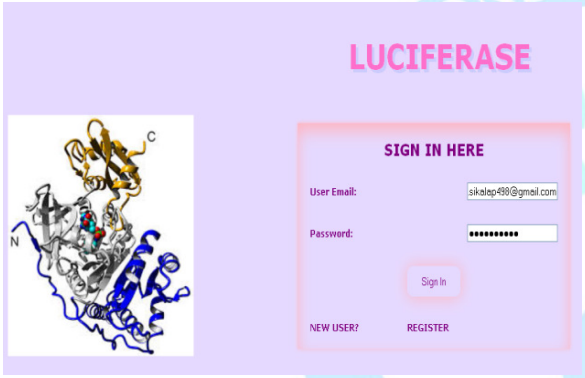


Figure 3. Sign in page

Figure 4. Sign up page

PHP codes are,

```
<?php
mysql_connect{"localhost","root",""};
mysql_select_db{"users_db"} or die
{mysql_error{}};
if {isset{ $_post{'submit'{}}}
$user name =$_post{'name'};
$user pass =$_post{'pass'};
$user email =$_post{'email'};
```

Once the user enters the database homepage shown in figure 5, a list of menus is provided with a navigation bar namely,

1. HISTORY
2. ORGANISM
3. BIOLUMINESCENCE
4. CLASSIFICATION

HISTORY menu provides a history of luciferase and structure determination of the bacterial and firefly luciferase shown in figure 6. Bacterial luciferase is the enzyme that catalyzes light emission at the heart of bacterial bioluminescence. However, the catalytic machinery involved in continuous light production in luminous bacteria includes not only bacterial luciferase, but also the enzymes that supply and regenerate the substrates of bacterial luciferase.

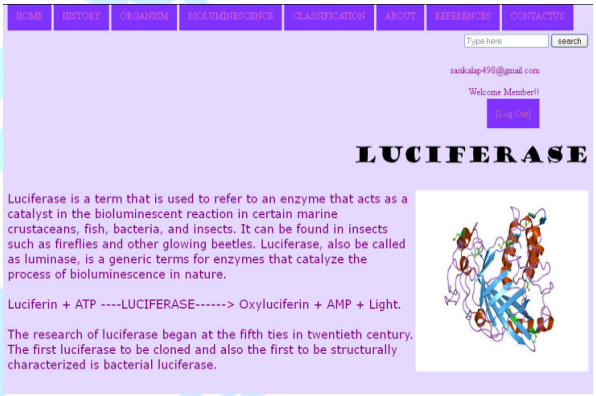


Figure 5. Luciferase-Homepage

Figure 6. Luciferase-History

ORGANISM menu provides 25 luciferase enzyme producing organisms created in the table Organism Name, Structure, Amino Acid, Accession Number, Fasta Sequence and Sequence Analysis are presented in figure 7-9. The Link pages of Luciferase enzyme producing organisms listed are shown in figure 10-12.

Figure 7. Luciferase enzyme producing organisms - list 1

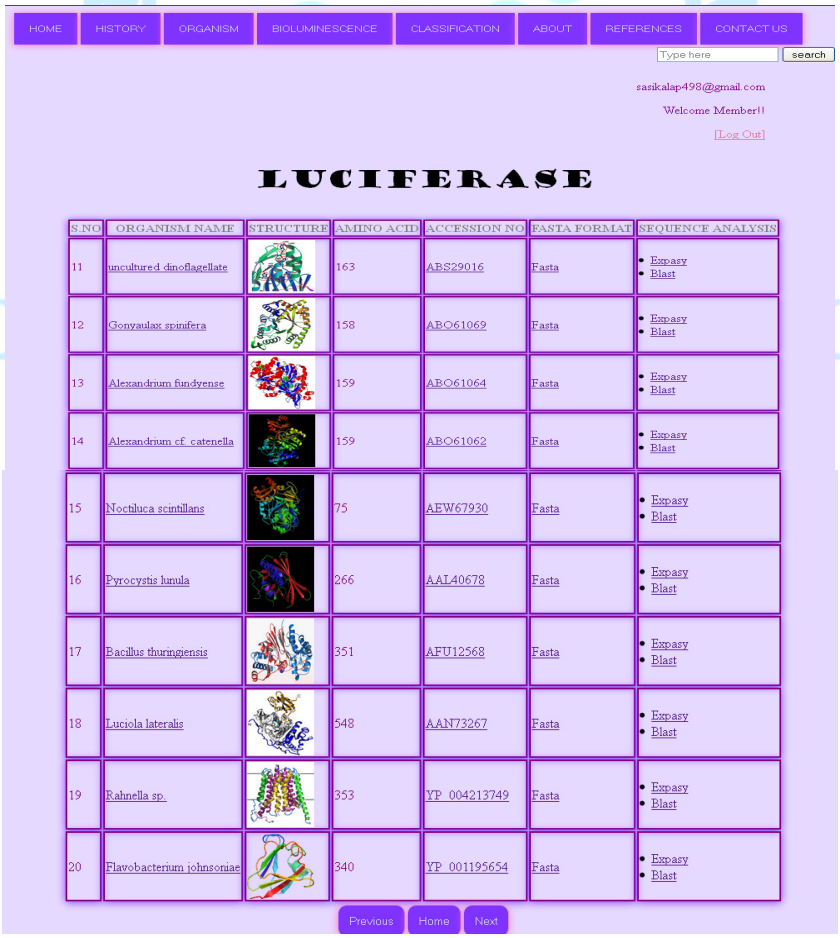


Figure 8. Luciferase enzyme producing organisms - list 2

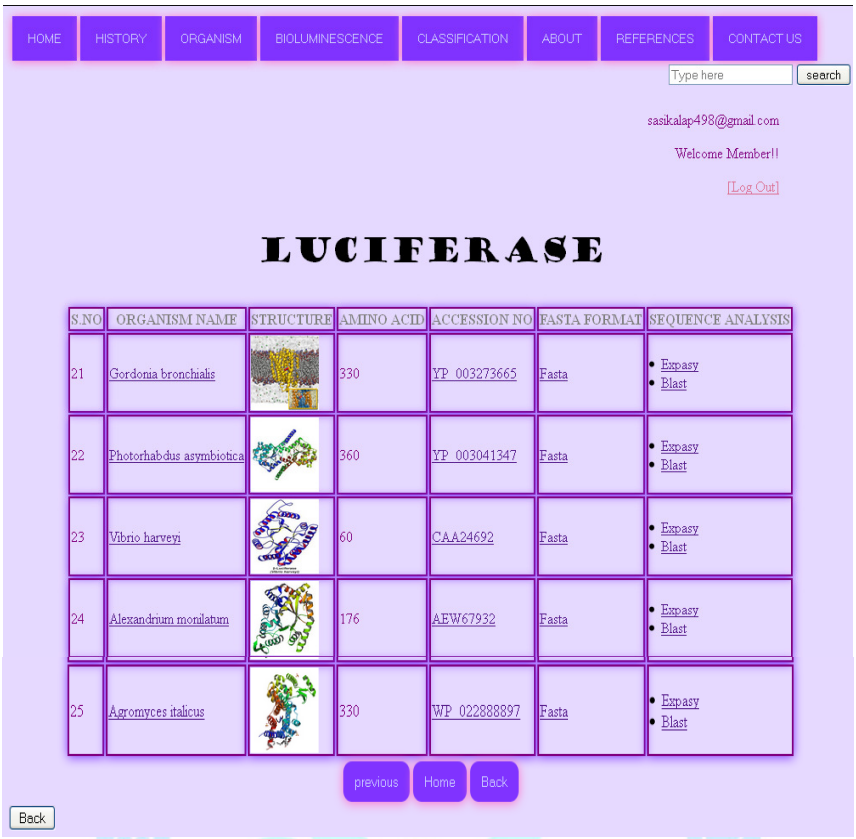


Figure 9. Luciferase enzyme producing organisms - list 3

Organism name: Luciferase enzyme producing organism's activities.
Structure: Luciferase enzyme producing organism's structure.
Amino acid: Luciferase enzyme producing organism number of amino acid presented.
Accession Number: Accession Number provides a Genbank sequence of Luciferase enzyme producing organisms.
Fasta sequence: This link provides a fasta sequence of Luciferase enzyme producing organisms.
Sequence analysis: Here the DNA, RNA or peptide sequences are subject to a wide range of analytical methods to understand its features, function, structure, or evolution using EXPASY (Protparam) AND BLAST.

BIOLUMINESCENCE menu provides an introduction to bioluminescence and pages linking to the types of bioluminescence are shown in figure 13-14. Bioluminecence is light produced by a chemical reaction with an organism. The bacterial bioluminescences are the most widely distributed light-emitting organisms with the majority existing in seawater and the remainder living in the terrestrial or freshwater environment.

Figure 10. Link pages of Luciferase enzyme producing organisms

CLASSIFICATION menu provides classification of luciferase and details about bacterial and insect luciferase link pages are shown in figure 15-18. The bacterial and insect

luciferase contains occurring in a remarkably diverse set of organisms including bioluminescence.

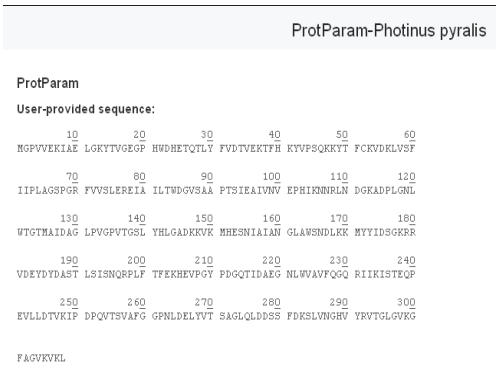


Figure 11. Expasy (protparam) result page

Figure 12. Blast result page

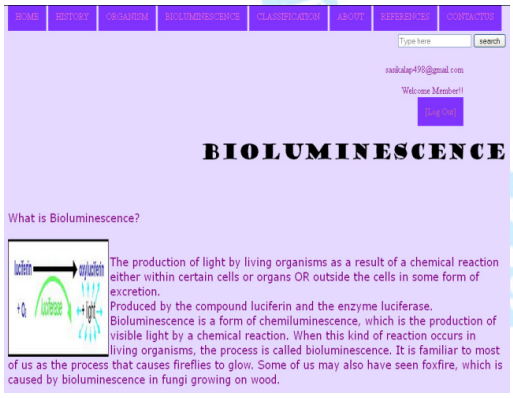


Figure 13. Luciferase-Bioluminescence

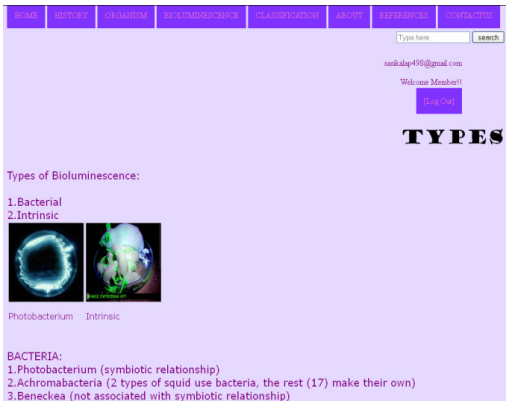


Figure 14. Types of Bioluminescence

Figure 15.Luciferase-Classfication

Figure 16. About luciferase

Figure 17. Bacterial luciferase

Figure 18. Firefly luciferase

The finally in this article provide develop a own search engine using PHP are shown in figure19, Mysql and Html codes are,

```
<html>
<head>
<base target="blank"></base>
</head>
<form id="searchbox" action="re.php" method="get">
<input type="text" name="value" placeholder="Type here">
 <input type="submit" name = "search" value="Search Now">
</form>
<hr>
<div style = "background-color:plum">
<?php
mysql_connect("localhost","root","")or die("cannot connect");
mysql_select_db("bioinformatics") or die(mysql_error());
echo "server connected!!";
if(isset($_GET['search'])) {
$search_value = $_GET['value'];
$query = "select * from informatics where KEYWORDS like '%$search_value%' ";
$run = mysql_query($query);
while($row=mysql_fetch_array($run)){
  $title =$row['TITLE'];
  $desc = $row['DESC'];
  $link = $row['LINK'];
   echo "<h2> $title</h2> <a href = '$link'>$link</a> <p> $desc</p>";
  }
}
?>
```

Figure 19. Luciferase-search results page

This database created with the help of WAMP holds good for the upcoming researches for their easy retrieval of the protein information about the luciferase enzyme producing organisms**.**

Conclusions

As of today web consists of millions of web pages. Data represented in Html, CSS, and JavaScript pages. Hence it is inefficient of meaningful information extraction.This paper focuses on ways to enhance the search results by using PHP.WAMP server is used to extract information from a PHP MyAdmin store where in MySQL query is embedded in PHP programs. This work can be further enriched inbuilt analysis tools deeper insights of luciferase enzyme producing organism's data updates.

Conflict of interest

The authors declare that there is no conflict of interests regarding the publication of this paper. Also, they declare that this paper or part of it has not been published elsewhere.

Acknowledgements

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