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**Database** 

### Actinobase: Database on molecular diversity, phylogeny and biocatalytic potential of salt tolerant alkaliphilic actinomycetes

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#### Abstract:

Actinobase is a relational database of molecular diversity, phylogeny and biocatalytic potential of haloalkaliphilic actinomycetes. The main objective of this data base is to provide easy access to range of information, data storage, comparison and analysis apart from reduced data redundancy, data entry, storage, retrieval costs and improve data security. Information related to habitat, cell morphology, Gram reaction, biochemical characterization and molecular features would allow researchers in understanding identification and stress adaptation of the existing and new candidates belonging to salt tolerant alkaliphilic actinomycetes. The PHP front end helps to add nucleotides and protein sequence of reported entries which directly help researchers to obtain the required details. Analysis of the genus wise status of the salt tolerant alkaliphilic actinomycetes indicated 6 different genera among the 40 classified entries of the salt tolerant alkaliphilic actinomycetes. The results represented wide spread occurrence of salt tolerant alkaliphilic actinomycetes belonging to diverse taxonomic positions. Entries and information related to actinomycetes in the database are publicly accessible at http://www.actinobase.in. On clustalW/X multiple sequence alignment of the alkaline protease gene sequences, different clusters emerged among the groups. The narrow search and limit options of the constructed database provided comparable information. The user friendly access to PHP front end facilitates would facilitate addition of sequences of reported entries.

Availability: http://www.actinobase.in.

Keywords: Alkaliphilic actinomycetes, Database, Actinobase

#### **Background:**

The actinomycetes are a group of morphologically and phylogenetically diverse gram-positive bacteria with high G+C contents (>55 mol %) in their DNA. This indicates marked chemical and morphological diversity, with a distinct evolutionary line. They are widely distributed in nature occupying varied habitats including antarctic soil [1], desert regions [2, 3], hot springs [4], highly saline areas [5] and moderately saline habitats [6-9].

Actinomycetes grow well in neutral to slightly alkaline media. The optimal growth of actinomycetes in alkaline media was described by Baldacci, 1944 and alkaliphilic actinomycetes were first isolated from soil [10]. Study of such actinomycetes has lead to the description of new taxa [11] and the discovery of alkaline proteases and new antibiotics [12]. Taxonomically, alkaliphilic mycelial prokaryotes belong to the genera *Streptomyces* and *Nocardiopsis* [13-16]. Halophilic microorganisms can be grouped based on their requirements of NaCl for growth [17]. Marginally halophilic organisms in marine environments can grow in the presence of 2-3% NaCl,

### BIOINFORMATION

while moderate halophiles are able to grow over a much wider NaCl concentration range (5 to 20%, w/v). The extreme halophiles, including the well-known halobacteria and halococci, are able to grow in saturated NaCl and fail to grow in NaCl lower than 12%. The occurrence of actinomycetes in high saline environments and the tolerance of these organisms to high concentrations of salts was earlier described by Tresener **[18]**.

Bioinformatics based analysis and prediction has played pivotal role in understanding and in-depth knowledge of biological molecules with reference to proteomics and genomics. Although, despite significant advancement, there have been limited efforts on the compilation of relevant information for a specific field of interest. With this realization, in the present report, we focused on the wide spread data and information related to the phylogeny and biocatalytic potential of salttolerant alkaliphilic actinomycetes. The information and detailed account on these specific actinomycetes are quite limited and scattered in scientific journals and other reports. Detailed information from the literature were retrieved, systematically compiled and analyzed. This followed the creation of a data base in the form of 'Actinobase' that reflect the diversity and functional analysis of salt tolerant alkaliphilic actinomycetes.

#### Methodology:

Actinobase contains information regarding salt tolerant alkaliphilic actinomycetes with respect to habitat, taxonomy, genus, Gram reaction, biochemical tests, sugar utilization, antibiotic resistance, phylogenetic relatedness, salt, pH, temperature profile and 16S r RNA gene sequence analysis. Back hand database of Actinobase was created in MS Excel and front end in PHP. Hypertext Preprocessor provided easy web access to database for data entry, retrieval and analysis. Data was collected and extracted from original research publications and public databases, i.e. NCBI, DDBJ and EMBL.

#### Data Input:

In Actinobase, we selected 40 salt-tolerant alkaliphilic actinomycetes and integrated detailed information comprising multiple fields: taxonomy, genus, site of isolation, habitat, sample detail, Gram reaction, media characteristics, biochemical properties, sugar utilization, antibiotic resistance, phylogeny, salt, pH, temperature profile and 16S r RNA gene sequence.

#### Data Retrieval

Actinobase is a freely accessed web database constructed using PHP on windows platform. "Actinobase" is the database which provides user friendly search criteria and easy access and retrieval with secure administrator and users. PHP is an HTMLembedded scripting language to dynamically generate pages. PHP is a server side scripting language used on the internet to create dynamic web pages. It's freely available, easy to learn and can run on Windows and UNIX operating systems. It offers effective security mechanisms and easy connectivity, which makes it a popular choice for modular programming.

#### **Discussion:**

#### Actinobase Creation

The Actinobase database **(Figure 1A)** was constructed primarily in Excel 2007 as back hand and exported to PHP as front hand with My SQL for the easy access and portability. The efforts have added to the field of environmental biotechnology with reference to salt tolerant alkaliphilic actinomycetes **[14, 19-21]**. Over the years, our group at the Saurashtra University, Rajkot (India) has focused on the diversity, distribution, molecular diversity, phylogeny and biocatalytic potential of salt tolerant actinomycetes **[22-25]**. In view of the scientific literature and our own contributions, we constructed a web driven database system for salt tolerant alkaliphilic actinomycetes to highlight their various aspects. This data base would be of helpful to scientific community on these alkaliphilic actinomycetes.

'Actinobase' provides platform for the easy access and retrieval of data from the database (Figure 1 B & C). While, Centraalbureau voor Schimmelcultures (CBS) Actinomycetes database (Utrecht, The Netherlands) already exists, the one described in the present report represents first attempt on the creation of a specialized database for salt tolerant alkaliphilic actinomycetes. The functionality to add and edit data to the database through a user friendly web based portal facilitates updating and maintenance of the database system. Information required by the researchers for salt tolerant alkaliphilic actinomycetes can be accessed and analyzed under a single system.

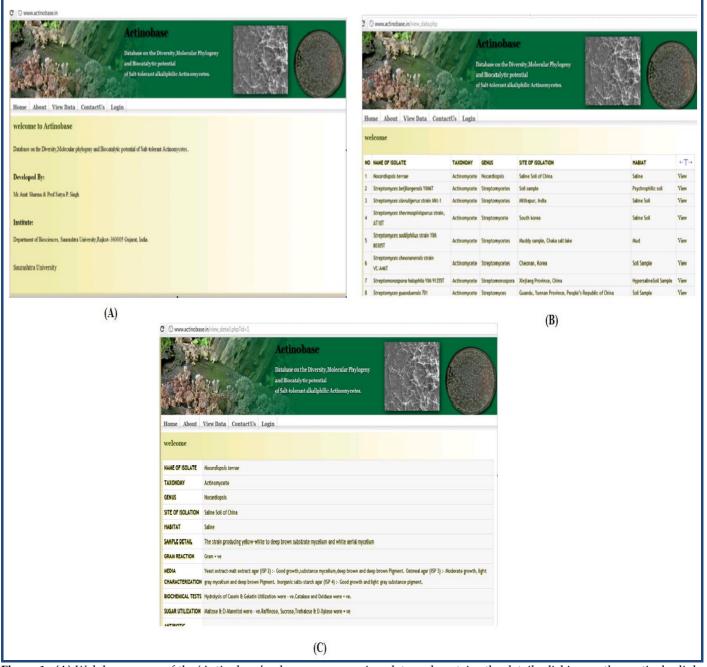
The analysis of the genus wise status of the salt tolerant alkaliphilic actinomycetes indicated the presence of 6 different genuses among the 40 classified entries of salt tolerant alkaliphilic actinomycetes. The results represented wide spread occurrence of salt tolerant alkaliphilic actinomycetes from the diverse taxonomic positions. The database contains the collection of 16S rRNA and protein sequences of the reported salt tolerant alkaliphilic actinomycetes. Multiple sequence alignment analysis was carried our and phylograms created for the available sequences of actinomycetes.

With reference to some actinomycetes, there was direct correlation between the genus and the cluster, which related to their geographical occurrence, protein sequences and physiological requirement under hyper saline or saline conditions. On the other hand, some members were not organized in clusters. Few strains of different genera have closer phylogenetic distances and the clustering reflected on their geographical location, similarity in sequences as well as requirement of salt and pH for growth. Thus, the patterns suggested that halophilic and alkaliphilic nature was not confined to a specific group of the actinomycetes. Adaptation to hyper saline and hyper pH conditions can be acquired by wide range of the actinomycetes from different taxonomic groups **[26].** 

Overall, the work highlighted on the biocatalytic potential and diversity of the salt tolerant actinomycetes with respect to colony morphology, Gram reaction, antibiotic profile, sugar utilization, media characterization and biochemical properties. The outcome highlighted on the versatility of the organisms with respect to these features. Additional properties, such as extra cellular enzyme secretion, growth at different salt and pH have added wider perspective to the study. The molecular phylogeny based on 16S rRNA and protease gene sequences is revealed in the data base. The information obtained on the halo

## BIOINFORMATION

tolerant, alkali tolerant and alkaliphilic actinomycetes would provide valuable platforms to research in this field.



**Figure 1**: **(A)** Web home page of the 'Actinobase', where users can view data and contains the details clicking on the particular link available on the home page; **(B)** the Snap Shot for the Data browsing of the 'Actinobase', which contains information related to the names of the isolates, taxonomy, genus, sites of isolation and the habitats; **(C)** reflects Data browsing of the 'Actinobase' for the individual organism which contains the detail information of the particular organism.

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