



SEQUENCED STRAINS OF ASPERGILLUS SPECIES

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ABSTRACT

Aspergillus is a large genus of common contaminants having more than 180 species, a fifth of which is frequently recovered from humans and other animals. The clinically important among these are *Aspergillus fumigatus* causing aspergillosis followed by *A.flavus*, *A.niger*, *A.terreus* and *A.nidulans*. These all species are either of clinical or industrial importance. In addition, *A.nidulans* has been used as an excellent genetical test-system for the in-depth study of genetical damages of various kinds ranging from point mutation to large chromosomal aberrations (Jha et al. 2009). There has been substantial investment in sequencing and initial annotation of the first filamentous fungal genomes. Genomics give new tools for fungal biology that should allow more rapid transfer of ideas, knowledge and expertise between specialist areas. The *aspergilli* provide a good example of what can be gained from genome sequences and highlights some of the problems. The interest in this group comes from their importance as pathogens and allergens of humans (*Aspergillus fumigatus*; *Neosartorya fischeri*), plant pathogens (*Aspergillus flavus*; *Aspergillus parasiticus*), industrial organisms (*Aspergillus oryzae*; *Aspergillus niger*; *Aspergillus terreus*) and as a model organism (*Aspergillus nidulans*). Here we are trying to gather information studied by the various researchers.

KEYWORDS : *Aspergillus* Spp., Genomics

Comparative Genomics

Traditional methods for the identification of these species suffers from many drawbacks. As a result, the traditional methods have been replaced by molecular approaches. The molecular methods such as comparative sequence based methods offer format for a rapid identification of species with the ability to detect even single nucleotide difference and consequently differentiate fungal isolates down to the species /strain levels and has been instrumental in resolving relationships among species within the genus *Aspergillus*. Whole or partial genome sequences are now available for most *Aspergillus* species (<http://www.broad.mit.edu/>) enabling a design of useful DNA sequence based methods for identification. In addition, the Gene Bank Sequence database (<http://www.ncbi.nih.gov/BLAST>) maintained by NLM and EMBL are also accessible (Hinrickson et al. 2005). There are also a good number of sites and software programmes for the study of bioinformatics of various species of *Aspergillus*.

Aspergillus genomics was reviewed by Jones (2007) and review provides URLs for major *Aspergillus* genome projects listing genes, availability of other resources, links to relevant data bases and literature citations. Genome sizes for sequenced species of *Aspergillus* range from approximately 29.3 Mb for *A. fumigatus* to 37.1 Mb for *A. oryzae* while the numbers of predicted genes vary from approximately 9926 for *A. fumigatus* to approximately 12,071 for *A. oryzae* (Machida et al., 2005; Nierman et al., 2005). The genome size of an enzyme producing strain of *A. niger* is of intermediate size at 33.9 Mb (Pel et al., 2007). Up-to-date listings of *Aspergillus* genome projects are available at the Genomes on Line Database (GOLD) at <http://www.genomesonline.org/>. *Aspergillus* species are only one group among a large number of eukaryotes now catalogued in databanks. The recent availability of *Aspergillus* genome sequences (*A.nidulans*, *A.fumigatus*, *A.oryzae*, *A.clavatus*, *A.terreus*, *A.flavus*, *A.niger* and *Neosartorya fischeria*) make this genus a treasure trove for comparative genomic studies (Table 1).

Table.1. Websites And Status Of The *Aspergillus* Genomes That Have Been Sequenced (november 2006).

Species	Strain	URL	Referene
<i>A.clavatus</i>	NRRL1	http://msc.tigr.org/aspergillus/aspergillus_clavatus_nrrl_1/index.shtml	Wortman et al. (2006)

<i>A.flavus</i>	NRRL3357	http://www.aspergillusflavus.org/	Payne et al. (2006)
<i>A.fumigatus</i>	Af293	http://www.tigr.org/tdb/e2k1/afu1/	Nierman et al.(2005)
<i>A.nidulans</i>	FGSCA4	http://www.cadre.man.ac.uk/Aspergillus_nidulans/	Wortman et al.(2006)
<i>A.niger</i>	ATCC1015	http://genome.jgipst.org/Aspni1/Aspni1.home.html	Galagan et al.(2005a)
<i>A.oryze</i>	RIB40	http://www.bio.nite.go.jp/dogan/MicroTop?GENOME_ID=ao	Payne et al (2006)
<i>A.terres</i>	NIH2624	http://www.ncbi.nlm.nih.gov/entrez/viewer.fcgi?val=AA00000000	Wortman et al.(2006)
<i>A.parasitics</i>	N.A	http://www.genome.ou.edu/fungal.html	Not Available
<i>N.fischerii</i>	NRRL181	http://msc.tigr.org/aspergillus/neosartorya_fischeri_nrrl_181/index.shtml	Wortman et al.(2006)

Sequenced *Aspergillus* Species

Aspergillus Fumigatus Af293 And A1163

A.fumigatus is a ubiquitous inhabitant of vegetable matter composts and human habitats and is considered by many to be the world's most harmful mould. It is the leading cause of IA (Invasive Aspergillosis) in the people with compromised immune function. IA is the most common manifestation of *A.fumigatus* infection in immunocompromised patients, having incidence rates of 10-15% in allogenic bone marrow recipients, 7% in acute leukaemia and 40% in inherited chronic granulomatous disease(Marr et al.,2000).The therapeutic management options for IA are limited and even with antifungal therapy the mortality rate is approximately 50%.In addition, *A.fumigatus* causes allergic disease in the form of allergic bronchopulmonary aspergillosis and fungal sinusitis and may be of major significance in many adults with severe asthma(Boyer et al.,2006). Most *A.fumigatus* isolates belong to one phylogenetic subspecies with a global distribution and is characterized by surprisingly low levels of

genetic diversity (Pringle 2005). This is in contrast with the high levels observed in some of its closest sexual relatives, such as *Neosartorya fischeri* (Rydholm 2006).

Neosartorya Fischeri (aspergillus Fischerianus) Nr1181

N.fischeri is a very close homothallic sexual relative of *A.fumigatus*. Despite the phylogenetic proximity of these two species, *N.fischeri* is very rarely identified as an invasive pathogen. Like most *Aspergilli* *N.fischeri* is saprophytic and plays a role in food spoilage. Its thermo resistant ascospores allow it to survive heat processing and cause spoilage of processed fruits and juices. The *N. fischeri* genomes contain a large number of transposable elements, which are thought to contribute to its genome size expansion.

Aspergillus Clavatus Nr11

A.clavatus is a very rare human pathogen with only one invasive medical case reported, that of post-operative endocarditis and occasional external otitis. It does appear to be potentially allergenic in humans and can cause neurotoxicosis in sheep and cattle fed infected grain. *A.clavatus* may be an important allergenic fungus and has been shown to be the cause of extrinsic allergic alveolitis known as malt workers lung. It grows more slowly at 37°C than *A.fumigatus* and has a bigger spore size, which is thought to prevent lung penetration. It produces a number of mycotoxins including patulin, kojic acid, cytochalasins and tremorgenic mycotoxins. The *A.clavatus* genome (27.9Mb), which was sequenced by the WGS method (Fedorova *et al.*, 2008), is the smallest seen to date among the *Aspergilla*. The reduced genome size was probably accompanied by massive gene loss. There are currently only 9125 predicted protein coding genes. Comparisons to the genomes of *A.fumigatus Af293* and *N.fischeri* revealed approx.120 genes unique to *A.clavatus*, including patulin biosynthesis and other mycotoxin clusters as well as paralogous gene families.

Aspergillus Terreus Nih2624

A.terreus produces numerous other secondary metabolites (e.g., Patulin, Citrinin, Isoterrin, and Asterriquinone) and commercially important enzymes (e.g., Xylanase). Understanding the *A.terreus* genome and differences between it and the genome of carefully selected relatives will vastly improve our knowledge of pathogenesis, antibiotic resistance and the biology of this infectious agent. *Aspergillus terreus* is an emerging fungal pathogen, which has been recognised as a cause of frequently lethal infections that are often refractory to amphotericinB (Walsh *et al.*, 2003). After *A.fumigatus* and *A.flavus*, it is the most frequently isolated *Aspergillus* species isolated from patients with IA. The genome sequence of *A.terreus* strain NRRL12624 is available at the NCBI web site and some information on sequencing project is available at <http://www.broad.mit.edu>.

Aspergillus Nidulans Fgsc4

Similar to *N.fischeri*, *A.nidulans* possesses a known sexual cycle, which made it genetically malleable organism with a well-developed genetic system. It can produce both asexual (conidia) and sexual spores (ascospores). It has been used extensively as a model organism for studies of cell biology including such intracellular functions as mitosis, the assembly of microtubules and the mitotic motors kinesin and cytoplasmic dynein. Carbon and nitrogen regulation are well studied. It was rarely has been isolated from IA patients and is considered as extremely weak pathogen. *A.nidulans* is a distinct pathogen in chronic granulomatous disease (CGD) and is often associated with more severe complications than that of *A.fumigatus* (Segal 1998). The *A.nidulans* genome sequence (30.0Mb) was reported as a comparative genomic analysis with *A.fumigatus* and *A.oryzae* (Galagan *et al.*, 2007).

Aspergillus Niger Cbs513.55 And Atcc1015

A.niger is used extensively as an industrial fermentation

organism for the production of citric and gluconic acid and enzymes used in food production and a wide range of industrial applications. It is used for production of heterologous proteins. It is a member of black *Aspergilli*. In their natural habitat, they secrete large amounts of a wide variety of enzymes needed to release nutrients from environmental biopolymers. It is the high secretory capacity that is exploited in both solid state and submerged culture fermentations.

CONCLUSION

The genomic mining may uncover new natural products (Archer *et al.*, 2006, Bok *et al.*, 2006). The comparative genomics helped in the preparation of dendrogram, phylogenetic tree and reveals many new features of evolutionary relationship, in addition to molecular identification and characterization of species. DNA data has allowed the reconstruction of the possible scenarios for the evolution of mating types, secondary metabolite cluster, enzymes involved in biomass degradation and other important pathways (Payne *et al.*, 2006). Comparative genomic data also characterize biosynthetic processes. The use of microarrays and proteomics technology reveals the levels of expression. The advanced bioinformatics and data analysis tools, throw light on many of the functional properties and activities of *Aspergillus* fungal genome (Bok *et al.*, 2006). Interpretation of the database has revealed many new facts of metabolism and also enlightened fundamental insights into cell biology and the nature and design of drugs.

The *Aspergillus* Genome Database (AspGD) is an online genomics resource for researchers studying the genetics and molecular biology of the *Aspergilli*. AspGD combines high-quality manual curation of the experimental scientific literature examining the genetics and molecular biology of *Aspergilli*, cutting-edge comparative genomics approaches to iteratively refine and improve structural gene annotations across multiple *Aspergillus* species, and web-based research tools for accessing and exploring the data. AspGD provides bulk search and download functionality through a Batch Download tool, and a browsable download site. The Batch Download tool allows retrieval of sequence and other information for a list of chromosomal features (e.g. protein-coding genes or tRNA genes). The AspGD download site includes files of basic gene information, sequence information, GO annotations, phenotype curation, interspecies homology mappings and other types of data. The primary mission of the *Aspergillus* Genome Database (AspGD) is to serve the needs of the scientific community in order to facilitate and accelerate *Aspergillus* research in the laboratory.

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