ABSTRACT

Title of Document: *METARHIZIUM ANISOPLIAE'S* PERSISTENCE AS A SAPROPHYTE, GENETIC BASIS OF ADAPTATION AND ROLE AS A PLANT SYMBIONT.

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Metarhizium anisopliae is a model for elucidating the basis of entomopathogenicity. However, many aspects of the saprophytic life of *M. anisopliae* remain unclear, including: 1) how the rhizosphere maintains populations of *M. anisopliae*; 2) the potential for microevolution; 3) the genetic factors that allow *M. anisopliae* to adapt to a saprophytic life and 4) the extent to which plant ecology is impacted by the rhizospheric competence of *M. anisopliae*. To extend our knowledge of plant-insect-fungal interactions, a field trial was conducted with *M. anisopliae* applied to turf. The specific objectives were to: 1) investigate the genetic basis of adaptation and strain stability of *M. anisopliae*; 2) monitor long-term survival of *M. anisopliae*, 3) compare the winter survival of *M. anisopliae* Δ*Mcl1* (disrupted in an immune evasion gene and nonpathogenic) with *M. anisopliae* Δ*Mad2* (adhesin-deficient and unable to adhere to plant epidermis) and 4) investigate root colonization and its impact on plant growth of *Triticum aestivum* seeds coated with conidia.

Results showed that *M. anisopliae* Δ*Mad2* had a linear decrease in population and did not colonize roots, while *M. anisopliae* and *M. anisopliae* Δ*Mcl1* cycled with seasons in the soil and colonized roots. Microarray analyses were used to assay for any mutations affecting gene expression during survival in the field. After 3.5 years, field recovered *Metarhizium* isolates differed in gene expression by an average of 0.26 % for the 1,749 expressed sequence tags. Mutations disproportionately affected cell wall and stress responses, while genes coding for pathogenicity determinants such as adhesins and toxins were highly conserved. *Triticum* seeds inoculated with *M. anisopliae* Δ*Mcl1* and *M. anisopliae* Δ*Mad2* prior to planting in the field produced a 14.92%, 4% and 0% increase in seed weight respectively. *M. anisopliae* increased growth rates of winter wheat in microcosms and may act as a biofertilizer by making insoluble phosphate available to plants. This study showed *Metarhizium* benefits plants by protecting them from insect pests and by potentially solubilizing otherwise unavailable nutrients. *Metarhizium*, therefore, may be implemented in an integrated pest management (IPM) approach to reduce the use of chemical insecticides and fertilizers.

METARHIZIUM ANISOPLIAE'S PERSISTENCE AS A SAPROPHYTE, GENETIC BASIS OF ADAPTATION AND ROLE AS A PLANT SYMBIONT.

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Dedication

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Chapter 1: *Metarhizium* and the rhizosphere

Metarhizium anisopliae as a saprophyte

Historically, the preferred natural habitat of common entomopathogens such as *Metarhizium* and *Beauveria* spp. has been uncertain (Prior, 1992). *Metarhizium* spp. is most abundant in undisturbed pasture soils, 2-6 cm deep, where *Metarhizium* populations can reach 10^6 propagules g^{-1} (Milner, 1992). It has not been clear whether the propagules recovered are conidia, mycelia surviving on insect remains, or mycelia living on non-insect substrates as saprophytes. Therefore, it remains unclear if these fungi could genuinely flourish in soil or survive in soil in a dormant state awaiting a susceptible insect host (Prior, 1992; Inglis *et al*., 2001). Presumably, conidia produced on the cadavers of large numbers of insect hosts contribute significantly to *Metarhizium* populations. However, soil populations as large as those characteristic of *M. anisopliae* are normally the result of organic substrates in rhizospheres present in the upper layer of the soil (Dix and Webster, 1995). Given that rooting density is particularly high in grasses and cereal crops with less than 3 mm spaces between roots (Barley, 1970); the *Metarhizium* community may be living in overlapping rhizospheres.

M. anisopliae, therefore, has two distinct lifestyles, one as an insect pathogen and the second as a soil-dwelling saprophyte associated with plant roots at the rhizosphere (Hu and St. Leger, 2002). As an insect pathogen *M. anisopliae* causes green muscardine disease to a wide range of insects, providing protection to an equally wide range of crops (Mikuni *et al*., 1982; Poprawski and Wraight, 2000; Roberts and St. Leger, 2004). As an entomopathogen, *Metarhizium* had been well studied and characteristics that make *Metarhizium* effective in killing target insect pests have been extensively examined (Milner, 1992; Milner *et al.*, 2002). Aside from a report that many soil isolates are non-pathogenic to scarab beetles, there is little information available on the relative virulence of isolates from the soil versus that of isolates from infected insects (Milner, 1992). There are likely different selective pressures for characteristics that would be required for both insect-infection and soil-survival (Prior, 1992). However, genetic groups or strains of *M. anisopliae* seem linked to habitat type (soil type, host plant association, climate, and geographic region) rather than insect host, suggesting that selection for survival in the soil is more important in shaping the population genetics of *M. anisopliae* than is selection for pathogenicity (Bidochka, 2001; Rangel *et al.*, 2005).

Interactions between organisms have an important role in shaping organismal diversity. Such interactions may be observed at the rhizosphere (Sarargchandra *et al.*, 1995). Except for a few limited aspects of host-pathogen and predator-prey interactions, the nature of evolutionary forces acting during these symbiotic processes is poorly understood (Duda and Palumbi, 1999). Even for the mycoparasitic *Trichoderma harzianum* where rhizosphere competence is known to be strongly related to biological control, the genetic and physiological factors controlling rhizospheric competence are poorly understood compared to those controlling pathogenicity (Harman and Dozelli, 2001; Whipps, 2001; Whipps *et al.*, 2001). Likewise, *M. anisopliae* has traditionally been studied exclusively as an insect pathogen with the emphasis on dissecting the mechanistic basis for insect pathogenicity. Little research has been performed on its life outside the insect, even though plant symbiotic strains of *M. anisopliae* could have considerable impact on agroecosystems in temperate regions (Meyling and Eilenberg, 2007).

M. anisopliae and *T. harzianum* are just two of many economically important pathogens for which the most understudied aspect of their biology involves the extended periods of time they survive in soil as saprophytes in the absence of a suitable host (Harman and Bjorkman, 1998; Kerry, 2000). Knowledge of a pathogen's behavior as a free saprophyte is important for predicting and controlling outbreaks of plant or animal diseases. In the particular case of *M. anisopliae*, a better understanding of how this fungus survives as a saprophyte at the rhizosphere may allow for farming techniques and modification of agricultural conditions so as to enhance the survival of *M. anisopliae* in field conditions (Meyling and Eilenberg, 2007). Understanding *Metarhizium's* saprophytic life will facilitate the identification of genes for rhizospheric competence that would be candidates for manipulation to increase the benefits of applying *Metarhizium* not just as an insecticide, but also potentially as a biofertilizer and/or possible competitor against plant pathogens. This could potentially lead to the development of *Metarhizium* as a comprehensive plant symbiont, but to achieve this, would require a much greater understanding of its functional ecology.

The plant rhizosphere as a habitat

The particular emphasis of this study was on survival of *M. anisopliae* at the rhizosphere, as this has been shown to be colonized by at least some strains of *M. anisopliae* (Hu and St. Leger, 2002). The rhizosphere is a narrow zone of soil subject to the influence of living roots, where root exudates may stimulate or inhibit microbial populations and their activities. The amount and composition of root exudates entering the soil depends on plant species, plant age and nutrient status (Brimecombe *et al*., 1991). A model of the submillimeter zone around a generic root estimates that exudates provide ~15 mg organic matter g^{-1} dry soil with 0.78 mg g^{-1} h^{-1} being degraded (Burgmann *et al.*, 2005), but concentrations of dissolved organic matter within millimeters from an individual root surface may be only 0.03 mg/ml (Wenzel *et al.,* 2001). The rhizoplane, also called the root surface, also provides a nutrient base for many species of bacteria and fungi. In return for nutrients provided by the plant, microbes may assist the plant by solubilizing inorganic nutrients or as biological control agents against plant pathogens. The ability of a microorganism to colonize and establish in the rhizosphere of developing roots has been defined as rhizosphere competence. This attribute has been found to be particularly important when considering the potential commercial use of biocontrol agents towards soil-borne plant pathogens (Baker, 1991).

The potential for rhizosphere competence of *M. anisopliae* in turf is particularly significant in an applied sense. *M. anisopliae* is already commercially available for the control of pests on pasture turf and many of its proposed future applications involve turf pests that would otherwise provide a particular challenge to pest control specialists as there are few other microorganisms available for use against them (Milner, 1992; Goettel and Inglis, 1997; Klein *et al.*, 2000). Proposed targets include white grubs, mole crickets, caterpillars, fire ants in pastures and home lawns (Roberts and St. Leger, 2004). Management of sports turf pests alone costs $1.5

billion per annum in the USA, mostly through the use of selective insecticides at the expense of a more holistic management approach (Potter, 2002). *M. anisopliae* is also being considered for control of ticks, including *Ixodes scapularis*-the carrier of Lyme disease (Benjamin *et al.*, 2002) and subterranean termites that in the USA alone cause $1 billion per annum in prevention and control costs (Wright *et al*., 2005). The focus of attention in *M. anisopliae* is shown by strain F52 (registered for use by the EPA in 2003) being targeted against various ticks, beetles and flies in residential and institutional lawns, landscape perimeters and greenhouses. Rhizosphere competence extends to plants growing in potting media in the greenhouse (Bruck and Donahue, 2007).

Although the preferred habitat of many *M. anisopliae* lineages may be grasslands as this is where the highest titers of *M. anisopliae* have been recorded (Milner, 1992; Roberts and St. Leger, 2004), *M. anisopliae* is diverse with respect to the plant roots *Metarhizium* colonizes. It has been demonstrated that *M. anisopliae* will form rhizospheric associations with grasses, cabbage, and legumes. If a strain of *Metarhizium* that is a good root colonizer is chosen, which is capable of being transported by the root through the soil profile, then seed treatment would be an attractive method for introducing a fungus into the soil-plant environment where the fungus may have the opportunity to be the first colonizer of roots (St. Leger, 2001). The seed has already been shown to be an important delivery vehicle for a variety of beneficial microbes for plant growth enhancement and biological disease control (Barea *et al*., 2005). The soluble components in root exudates stimulated germination and hyphal growth of *M. anisopliae* strain 2575 *in vitro* studies (Monica Pava-Ripoll unpublished data). This is likely to result in the germination of spores sometimes observed in the vicinity of the root. However, the initial behavior of the fungus on the roots, the extent to which roots are colonized, the persistence of colonization and the potential insecticidal application of this relationship are not yet known.

Many genotypes of *M. anisopliae* are known to be genetically variable and appear to not only differ in degree of virulence, but also appear to be specialized to different soils, which may need to be considered as a feature for selecting fungal strains for biological control (Bidochka, 2001). However, other environmental factors also influence the distribution of *M. anisopliae*. Fungal populations isolated from different geographic regions have been shown to survive at different optimal temperatures. Populations found closer to the equator survived at higher temperatures than populations isolated from the north and south poles, which showed increased tolerance for cold temperatures (Rangel *et al*., 2004). The maximum temperature *Metarhizium anisopliae* sf. *anisopliae* was shown to germinate was 37º C. Its thermal death point was between 49-60º C (Rangel *et al*., 2004).

Phylogeny of Metarhizium anisopliae

Metarhizium is an imperfect fungus of the subkingdom Dikarya and phylum Ascomycota. *Metarhizium* is a monophyletic group (Curran *et al.*, 1994; Humber, 2008*)*. In phylogenetic studies *M. anisopliae* clusters with family of clavicipitaceous grass endophytes (*Epichloe*) (Sung, *et al.* 2001) which are used commercially by applying them to turf grass. *M. anisopliae* has a clonal population structure (strains persist over time and space); no sexual stage is known in North America (but may exist elsewhere) and heterokaryon incompatibility precludes parasexuality, except between very closely related strains (St. Leger *et al.,* 1992a; Bidochka *et al.,* 1994). Therefore, gene exchange is likely to be a rare event (Prior, 1992), although this has not been properly investigated in field conditions. *M. anisopliae* contains strains with wide host ranges such as the generalist *M. anisopliae* sf. *anisopliae* 2575 as well as some specialists, *Metarhizium anisopliae* var. *majus* 297 (that infects scarab beetles), and *M. anisopliae* sf. *acridum* 324 (the "Green Muscle" strain used for locust control) that show specificity for certain locusts, beetles, crickets, and homopterans but are unable to infect other insects. While some specialized lineages, such as sf. *acridum*, are phylogenetically distant from generalist strains, implying evolutionarily conserved patterns for host infection strategies, closely related strains can also differ greatly in host range and requirements for germination (Fargues, 1984; St. Leger *et al*., 1992a,b; St. Leger *et al*., 1994a,b,c).

Patterns of gene duplication, divergence, and deletion in several generalist and specialist strains were specifically determined by heterologous hybridization of total genomic DNA. DNA from each strain was competitively hybridized to an array of strain 2575 genes. Most genes required for survival had differences in genomic hybridization which averaged less than 5%. One group of genes in the 2575 strain that seems to lack counterparts in specialist strains is mainly composed of mobile genetic elements (transposons). Other poorly conserved genes in specialist strains include some that may function in transporting and catabolizing sugars, non-ribosomal peptide synthases, a P450 cytochrome, a polyketide synthase, and several secreted enzymes including a chymotrypsin (Freimoser *et al*., 2003a,b; Wang *et al*., 2005b). This implies that specialists are losing genes primarily required to live in alternative hosts or as saprophytes.

There are more examples of specialists than generalists where only select members of gene families respond to a component of cuticle or hemolymph (Wang *et al.*, 2005b). Unlike 2575, when 324 is grown in submerged cultures, 324 up-regulates transcripts involved in sporulation. This relates to the unusual ability of 324 to produce spores inside host cadavers as an adaptation to desert living. Examining the role of regulatory variation with respect to adaptive evolution of a pathogen is important to understand the extent to which gene expression is used for a species to evolve and express new phenotypes (Ben-Shahar *et al.*, 2002). Yeast currently provides the model for examining processes in fungi for the heritability of transcription (Brem *et al.,* 2002; Townsend *et al*., 2003), for changes in gene expression levels in response to selection (Ferea *et al*., 1999), and regulatory variation in natural isolates. However, this variation has not been related to adaptation to different environments. The host-adapted subtypes of *M. anisopliae* provide a model where genetic variation can be related to adaptation to particular hosts and environment.

Results from Previous Field Trials

A field trial performed in 2000 on a patch of cabbage plants demonstrated that recombinant, transgenic *M. anisopliae* fungi were genetically stable over one year and their deployment had only ephemeral effects on the dynamics of the indigenous fungal populations (Hu and St. Leger, 2002). The field trial used transformants of *M. anisopliae gpd*-Pr1-4 (EGFP1), overexpressing a toxic protease and a jellyfish-derived green fluorescent protein (GFP) from *Aequorea victoria* (Hu and St. Leger, 2002). The recombinant strain *gpd*-Pr1-4 contained four copies of the Pr1a subtilisin gene under the control of a constitutive *gpd* promoter from *Aspergillus nidulans* and was hypervirulent to insects (St. Leger *et al.*, 1996a,b,c; Screen and St. Leger, 2001; Hu and St. Leger, 2002). The *gfp* gene is driven by a constitutive promoter and produces a cytoplasmically-located protein that strongly labels the whole fungus. Expressing GFP has no detectable effects on fungal growth or pathogenicity (St. Leger and Screen, 2001). GFP was utilized to monitor survival and distribution because there were no precedents for the release of such fungal products, and there is an inherent paucity of knowledge concerning the fate of fungal genotypes at the population and ecosystem level. GFP expressed by introduced strain of *M. anisopliae* will allow the introduced labeled strains to be differentiated form the native strains of *Metarhizium*.

The field test confirmed that GFP is an effective way to monitor pathogen strains in field populations and demonstrated the short term effects of insect transmission to non-target insects. This trial also revealed for the first time that *M. anisopliae* colonizes the rhizosphere, which places focus on the soil/root interface as a site where plants, insects and pathogens will interact to determine fungal efficacy, cycling and survival. In addition the field test showed that deployment of the transgenic strains had only ephemeral effects on the dynamics of the indigenous fungal population structure. However, an important caveat is that common agricultural practices such as tilling had already greatly reduced the abundance of *M. anisopliae* and other fungi in the test field (Hu and St. Leger, 2002). Opportunities for gene flow may therefore be greater in a stable grassy environment where fungal propagules are at least 1000 times more numerous and much more diverse. Samples were taken at 4-5 cm from the cabbage tap root as well as alongside the tap root (0-1 cm) to check for uneven distribution and persistence of spores (Hu and St. Leger, 2002). One day after application, the titer of 2575-GFP was 10^5 propagules/g at depths of 0-3 cm and after 6 months, this had declined in non-rhizosphere soils to 10^3 propagules/g. However, fungal densities remained at 10^5 propagules/g in rhizospheric soils so the R:S ratio at 6 months (the ratio of fungi in the rhizosphere to fungi in the non-rhizosphere soil) was about 100:1. The value remained high after the cabbage plants were killed by frost indicating that the GFP fungus was persisting on the decaying organic matter. The maximum rhizosphere effect was shown at depths of 0-3 cm (where roots were most numerous) and within 1 cm of the root surface, with no measurable increase in the numbers of fungal propagules at 2 cm. In spite of this proximity, most but not all of the fungus could be removed from roots with serial washings. It is important to note that plants are responsive even to the presence of apparently free living microorganisms (Andrews and Harris, 2000).

Although these studies demonstrated differences in pathogen persistence in rhizospheric and non-rhizospheric soils, they did not show that growth occurred. While most studies using other fungal species of proven root colonizers show a decline in population size, perhaps because the initial population added is too large for the carrying capacity of the root (Parke, 1991), population levels of *M. anisopliae* remained constant in the rhizosphere. The apparent carrying capacity for GFP *M. anisopliae* 2575 at the cabbage root rhizosphere was approximately 10^5 conidia (spores) per gram of soil, which is greater than the LC_{50} values for this strain against many insects (Goettel and Inglis, 1997). It may be that entomopathogenic fungi require insect hosts to build up and maintain their populations above 10^5 conidia per gram of soil (Meyling and Eilenberg, 2007).

A field trial in Tasmania used *M. anisopliae* strain DAT F-001 applied to perennial ryegrass and demonstrated that levels of colony forming units (CFUs) per gram of soil increased over four years and appeared to plateau at appropriately 10^5 CFUs per gram of soil supporting the findings that this is the carrying capacity of *M. anisopliae* at the rhizosphere (Rath *et al.*, 1995). There were also reduced numbers of subterranean scarab beetles, *Adoryphorus couloni*, but in the range of invertebrates present was not altered. Populations of *M. anisopliae* DAT F-001 were shown to increase as sporulation occurred on pupal cadavers (Rath *et al*, 1995).

Consequences of rhizospheric competence

Most studies employing the facultative saprophyte *M. anisopliae* have selected strains for optimum virulence against pest insects and have ignored habitat preferences and survival outside the host. The search for highly virulent isolates of this fungus may be inherently flawed given that factors associated with soil-dwelling may be even more critical in the selection of an isolate than virulence (Bidochka *et al.*, 2001). In the case of strains engineered for improved virulence, such strategies may fail if genes are engineered into a strain which survives poorly in the field. The ability to perform reproducible and safe applications of *M. anisopliae* is very important from the risk management point of view. Effective biological control lies in detailed knowledge of the properties of pathogens in their native environments. If rhizospheric competence is a general phenomenon among insect pathogens, this competence may need to be considered as a feature for selecting fungal strains for biological control, which raises the possibility of managing the rhizosphere microflora to achieve insect control. This would complement attempts in integrated pest management systems to manipulate the environment of the plant and insect to enhance insect biological control (Roberts and Hajek, 1992). However, such attempts are more likely to fail if the process of root colonization is poorly understood. Examining the nature and function of microbial gene products expressed during growth in soil and on root exudates is an essential step in attaining an explanation for the relationship between *M. anisopliae* and its environment.

Except for yeasts and some filamentous fungi, which are established research organisms, the majority of fungi remain poorly understood (Birren, 1996). Studying the saprophytic life style and adaptations of *M. anisopliae* is important, as this will provide new insight into fungal strategies. The more than 1 million different species in the fungal kingdom are distinguished by different life histories, developmental processes and ecological niches (Drummond *et al.*, 1987; Bidochka, 2001; Inglis *et al.*, 2001; Rangel *et al.*, 2005). The ecological diversity and range of this kingdom may be better understood from the results of the field study described here.

Rhizosphere competence also raises concerns, particularly if this competence resulted in introduced or hypervirulent transgenic strains persisting in the environment. Research in the St. Leger laboratory has emphasized engineering hypervirulent transgenic fungi (Wang and St. Leger, 2007). Clearly, biological safety issues must be considered prior to the release of these transgenics into the environment (Barea *et al.,* 2005). Mycorrhizial interactions and interactions between bacteria and plants have received the most attention. However, there is an extra dimension in the quality of the interactions with fungi, since unlike bacteria, they can potentially grow and spread through the soil and rhizosphere as hyphal growth. There are many environmental and economic reasons why researchers and industry would not seek to permanently establish an engineered microbial agent in the environment (St. Leger and Screen, 2001), and rhizosphere competence might increase the difficulty of eliminating the pathogen following unanticipated and deleterious environmental effects. Many crop plants are grasses where rhizosphere competence might be expected and, in any event, the relationship *Metarhizium* has with a plant appears to be nonspecific as rhizosphere competence was established with cabbages (Hu and St. Leger, 2002). It is also likely that an entomopathogen applied to fields could contaminate neighboring pastures and woodlands. Nevertheless, a key advantage of microbial control agents is their potential to replicate and persist in the environment providing long term control. A strain to persist in the environment long enough to kill pest insects, but short enough not to survive more than one season may be preferred. If a transgenic pathogen were to persist in the field, then another major biological safety issue to consider is genetic stability. This could be more closely examined when studying the saprophytic life of a pathogen (Barea *et al.*, 2005).

To achieve safe and reproducible biological control lies in detailed knowledge of the properties of pathogens in their environments. Such knowledge would fulfill the USDA Stakeholder Workshop’s (2003) which proposed reducing the ability of the organism to spread through a lack of saprophytic competence. The current field trial is designed to show whether *M. anisopliae* principally survives in field conditions through recycling in insects or as colonists of roots. This will generate knowledge and test strategies that could reduce persistence of transgenic pathogens

M. anisopliae as a model entomopathogen

M. anisopliae is a tractable model system offering ready culturability, synchronous germination and growth, EST collections, microarray analysis (Freimoser *et al.*, 2003a,b; Freimoser *et al.*, 2005; Wang *et al.*, 2005a,b; Wang and St. Leger, 2005), promoters that allow expression of foreign genes (St. Leger *et al.*, 1996a,b; Screen *et al.*, 2001) and gene disruption technology (Wang and St. Leger, 2006). *M. anisopliae* also produces many different cell types which may be employed for developmental studies. These cell types include conidia (asexual spores), hyphae, appressoria (pre-penetration swellings produced by many plant and insect pathogens used to attach to and penetrate their host) and unicellular blastospores (spores produced by budding). Another advantage of working with *M. anisopliae* is that there is a significant ecological and genetic difference between strains, which facilitates comparative studies on life strategies (St. Leger *et al.,* 1992a, b; Bidochka *et al.*,1994).

The importance of field trials and researching M. anisopliae as a possible plant symbiont

There is much more to learn about the biology of *M. anisopliae.* Fortunately, the *gfp* gene has made analyzing population dynamics in soil much less arduous than in the past. Keller and Zimmerman (1989) concluded a review on the mycopathogens of soil insects with some questions that this research may now be able to answer. How long is a saprophytic life possible? Does the saprophytic ability vary between strains? How does the fungus spread? Does manner of application and/or time of application alter survival of the fungus? Answers to these and other questions are fundamentally and practically valuable to understanding soil-insect-mycopathogen relationships.

The experiments discussed here used recombinant mycoinsecticide *M. anisopliae-gfp* to: 1) monitor long-term survivorship in field conditions of introduced *Metarhizium* using *gfp*-marked fungal strains, 2) monitor and compare indigenous *M. anisopliae* population structure to introduced populations, 3) determine possible mechanisms for how populations of *M. anisopliae* are maintained by examining the role of rhizospheric competence and insect pathogenicity using knock-out mutants, 4) analyze the genetic (mitotic) stability of an introduced strain over time in field conditions and 5) examine the nature of the relationship between *M. anisopliae* the rhizospheric plant (tall fescue grass and winter wheat). A better understanding of *Metarhizium's* life as a saprophyte may be elucidated by researching *Metarhizium's* population patterns, permanent changes in gene expression and *Metarhizium's* relationship with the rhizospheric host plant.

Chapter 2: Testing the survival of *Metarhizium anisopliae* sf *anisopliae 2575* and its knockout mutants Δ*Mcl1* and Δ*Mad2* in field conditions

Introduction

A *gfp* labeled lab strain of *M. anisopliae* sf. *anisopliae* 2575 and two transgenic knockout strains (also labeled with *gfp*) were used in this study to provide insight as to how *M. anisopliae* survives in field conditions. The knockouts, Δ*Mcl1* and Δ*Mad2,* are disrupted in critical genes required for insect pathogenicity and rhizospheric competence, respectively. Measuring populations of *M. anisopliae* Δ*Mcl1* in field conditions will determine the importance of cycling through insects for fungal survivorship. *Metarhizium* collagen-like protein, MCL1, is the most highly expressed gene when *Metarhizium* 2575 is grown in hemolymph (5.6% of all ESTs). Gene knockout confirmed that MCL1 is required for immune evasion and pathogenicity (Wang and St. Leger, 2006). MCL1 is transcriptionally-regulated during growth in the hemolymph of a very diverse array of insect species, consistent with the broad host range of 2575. However, MCL1 is not expressed in root exudates, starvation conditions, or nutrient rich artificial media indicating that this gene is involved exclusively in pathogenesis (Wang and St. Leger, 2006). With this gene's function disrupted, *Metarhizium* 2575 is no longer a viable pathogen to insects and would have to survive solely as a saprophyte in the field. The ability of Δ*Mcl1* to grow in root exudates in a Petri dish on nutrient agar was not altered by disrupting

MCL1, neither was its ability to grow on nutrient agar. The plasmid used to produce Δ*Mcl1* contains the *gfp* genes as well as the *bar* gene from *Streptomyces* that encodes phosphinothricin acetyltransferase, PAT. The *bar* gene confers resistance to Pestanal™, the commercial name for the fungicide glufosinate-ammonium. The addition of *bar* and the *gfp* genes provide a means of selectively isolating this strain from native strains collected from the field (Wang and St. Leger, 2006). Use of Δ*Mcl1* in a field trial will demonstrate, for the first time, whether cycling though insect populations is required for maintaining the large populations of *M. anisopliae* found in grasslands.

Metarhizium adhesin-like protein 2, MAD2, codes for a protein that allows *M. anisopliae* 2575 to adhere to the epidermis of plants. Disruption of this gene resulted in conidia that were unable to adhere to plant surfaces, but did not alter the pathogenicity of *M. anisopliae* to insects (Wang and St. Leger, 2007). It was hypothesized that *M. anisopliae* Δ*Mad*2 may have reduced competence at the rhizosphere surface because of its inability to adhere to the plant. *M. anisopliae* Δ*Mad*2 has been labeled with the *gfp* and *bar* marker genes to monitor population levels and distribution in field conditions. By measuring survival of *M. anisopliae* Δ*Mad*2 in field conditions, the importance of fungal associations with plants in maintaining population size will be determined.

Protocols will include examining persistence of the colonies, measuring fungal migration, and examining any genotypic and phenotypic changes to the fungal colonies over time. The experiments were conducted at the University of Maryland, Paint Branch Turf Grass Research Facility at 395 Greenmeade Drive, and at Dr. Raymond St.

Leger’s laboratory in the Plant Science Building, College Park MD 20742. The turf grass was predominantly tall fescue and plots were irrigated according to rainfall. Irrigation continued from early summer until mid-November.

Materials and Methods

Measuring the 4 year survival of M. anisopliae 2575-GFP *on turf and bare plots*

All experiments employed *M. anisopliae* ARSEF 2575 (isolated from a pecan weevil in South Carolina) or its disruption mutants Δ*Mcl1* or ΔM*ad2*. Plots of turf land and bare soil were each inoculated with one liter of a 10^6 conidial suspension l^{-1} per square meter plot. Conidia were prepared for inoculation by growing *M. anisopliae* 2575 on PDA plates until heavy sporulation was visible. Conidia were scrapped off plates and mixed into a 0.01% Tween™ solution forming a 10^6 conidia l^{-1} suspension (estimated using a hemocytometer). Ten liters of a final concentration of 10^6 conidia l^{-1} were prepared and one liter of this suspension was applied to each plot. A block design was used to set up five one meter^2 turf plots and five one meter^2 freshly tilled bare soil plots. There was a one meter turf buffer between each plot. Plots were marked using spray paint. Population levels of *Metarhizium* per gram weight of soil were determined on a rose bengal, cycloheximide-based selective medium (Hu and St. Leger, 2002). At the end of each month, three one gram soil samples were taken using a cork borer from a depth of five centimeters from each of the five turf and bare plots. Samples were prepared for plating by vortexing 9 ml of a 0.01% Tween solution with each of the thirty 1 gram soil samples. Then 100µl of this suspension was plated on a *M. anisopliae* selective media consisting of 31.55g Rose Bengal Agar (RBA), 1 l distilled water, 10g l^{-1} oxgall powder (bile salts), 80mg l^{-1} oxytetracycline, 80mg l^{-1} streptomycin, 80mg l^{-1} penicillin, 500mg l^{-1} chloramphenicol , and 250mg l^{-1} cycloheximide to screen for colonies. Total colony forming units (CFUs) of *Metarhizium* and the subset of these expressing GFP were counted approximately twelve days after plating the samples. GFP recombinants were distinguished from indigenous strains of *Metarhizium* spp. by utilizing an Olympus SZX12 dissecting scope with Scientific, Inc. mercury 100W light source and GFP 470/500 filter. Plates were examined and compared to the original lab sample of *M. anisopliae* to identify any noticeable changes in colony phenotypes. The numbers of CFUs were monitored at monthly intervals over a period of 4 years from August 2004 (the time of the initial application) until July 2008. The effects of temperature were plotted against CFUs to measure any influence temperature may have on fungal survival (Figure 2.2). All weather data were obtained from the National Oceanic and Atmospheric Administration (NOAA). Samples were collected at the beginning and end of the experiment from uninoculated control turf plots to titer the native *Metarhizium* CFUs.

It is possible that transgenic fungi will spread on or in the plant; therefore leaves, shoots and roots were sectioned into 5 cm segments from randomly selected plants that were removed from the turf plots. The plants were washed, air-dried and placed on selective medium to look for fluorescent growth (Hu and St. Leger, 2002). Since GFP decays quickly in dead fungi (St. Leger, unpublished data), these measurements quantify living populations. Direct observations by the above methods were conducted several times over four years and indicated the distribution of *M. anisopliae* in the soil and root surface.

For the 4 year field study, an analysis of variance (ANOVA) analysis was performed using SAS to compare gfp-labeled *Metarhizium* on bare and grass plots to native *Metarhizium* recovered from bare and grass plots. Survivorship in field conditions of these recovered strains were analyzed using proc mixed protocol after data were transformed using log10+1 to satisfy assumptions of normality and equal variance. Samples were analyzed by calendar year. Monthly population means of the four groups: native *Metarhizium* on turf, *Metarhizium*-GFP on turf, native *Metarhizium* on bare soil and *Metarhizium*-GFP on bare soil were compared to each other for each year from 2004-2008. ANOVA analysis was also performed to examine the population dynamics of each individual treatment for each of these years ($p<0.05$).

Measuring the winter survival of M. anisopliae sf *anisopliae* 2575, *M. anisopliae* sf *anisopliae* 2575 Δ*Mcl*1, and *M. anisopliae* sf *anisopliae* 2575Δ*Mad2 on turf plots*

Fifteen turf plots were inoculated with a 10^6 conidia l^{-1} suspension per square meter plot. Conidia were prepared for inoculation by growing *M. anisopliae* sf *anisopliae* 2575, *M. anisopliae* sf *anisopliae* 2575 Δ*Mcl*1, and *M. anisopliae* sf *anisopliae* 2575Δ*Mad2* on PDA plates. A random block design was used to set up five turf plots for each variant. Each plot was 1 meter by 1 meter, with a minimum buffer of 1 meter between each plot (Figure 2.1). Protocols for collecting and examining samples each month were the same as that for measuring the 4 year survival of *M. anisopliae* 2575 on turf and bare plots described above.

ANOVA analysis was performed using SAS to compare the knockout mutants Δ*Mcl1* and Δ*Mad2* survivorship in field conditions to recovered native strains. Survivorship in field conditions of these recovered strains were analyzed using proc mixed protocol after data were transformed using log10+1 to satisfy assumptions of normality and equal variance outliers were identified and removed. In addition, ANOVA analysis was performed using SAS to compare population dynamics between months within each of the three classes (native, Δ*Mcl1* and Δ*Mad2*) examined.

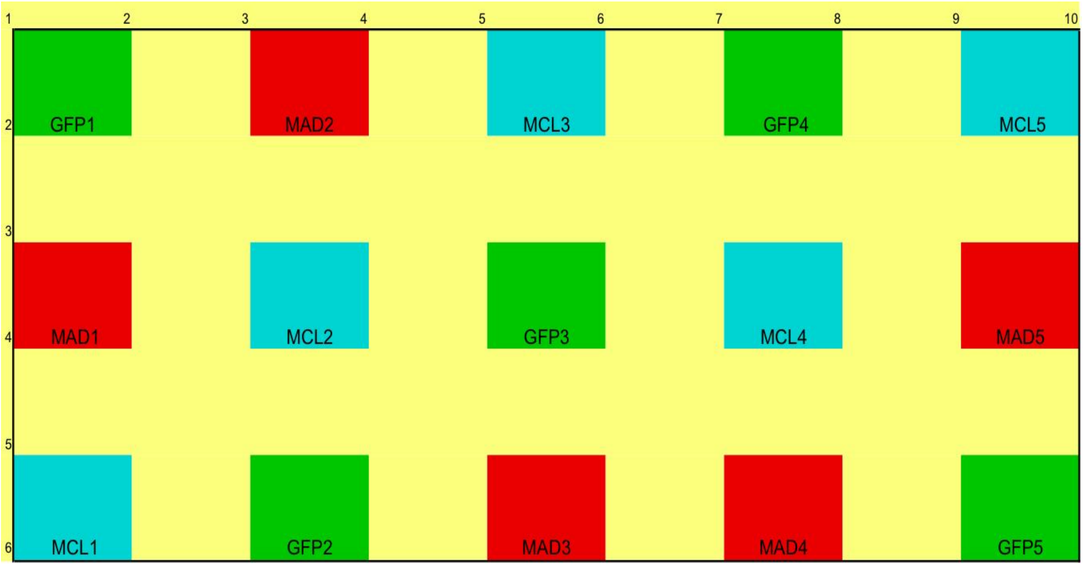


Figure 2.1 **The random block design used to set up turf plots**. Each plot measured 1 meter by 1 meter. There was a minimum one 1 meter of soil buffer between each plot. A suspension of one liter of 10^6 conidia was applied to each plot. Three soil samples from each of the 15 plots were collected at the end of each month.

Results

Overall Trends

The morphology of CFUs isolated from the plots as judged by their appearance and growth rate on selective media agar plates did not change over 4 years. It took an average of 12 days for colonies recovered from the field to form measurable CFUs when plated on rose bengal based selective media. Native *Metarhizium* strains had to be differentiated from the introduced strain 2575 under an ultraviolet (UV) florescence dissecting scope, since native colonies and introduced colonies had no other distinguishable morphological differences.

The distribution of *M. anisopliae* populations was patchy within plots accounting for high standard errors (Tables 2.2-2.10). Insect cadavers were never observed, but other researchers have also described localized high densities of CFUs, suggesting that the cadavers had disintegrated or blown away in the wind (Rath *et al.*, 1995). At 6 month intervals, soil samples were taken to measure drift at 0.5, 1.5, and 2 meters from the plots. No CFUs of the introduced *Metarhizium* were recovered from regions outside of the inoculated plots.

When entire grass plants were examined for the presence of *Metarhizium*, fungus was only observed on the roots. Samples from grass blades showed no presence of *Metarhizium*.

Measuring the 4-year survival of M. anisopliae 2575 *on turf and bare plots*

Over the past four years since the initial application of conidia, populations of transgenic *Metarhizium anisopliae* have progressively decreased but several intermittent recoveries in population size were observed, so the decrease was non-linear (Figure 2.2) (Tables 2.1, 2.2, 2.3). Native strains cycled in similar patterns as the introduced strain.

Decreases in CFUs in winter months

Metarhizium populations of the introduced strains on turf and bare soil as well as native populations on bare soil decreased in response to the winter months of December 2004, January 2005, February 2005 and March 2005 (Figures 2.3, 2.4, 2.8, 2.9, 2.10, 2.11, 2.12, 2.13) (Tables 2.1, 2.2, 2.3). For these three months, there was an average of 21 days per each month with temperatures that fell below 0º C. *Metarhizium* populations also decreased in response to the winter months of December 2005 which had 26 days that had temperatures fall below 0º C (Figures 2.4, 2.10, 2.11, 2.12, 2.13) (Tables 2.1, 2.3) of January 2007 and December 2007 which had 21 and 20 days that had temperatures fall below 0º C respectively (Figures 2.6, 2.18, 2.19, 2.20, 2.21) (Tables 2.1, 2.5).

Decreases in CFUs in summer months

Some major summer declines in both the native and introduced CFUS occurred June 2005, July 2005 and August 2005 which had 9, 10 and 10 days that had temperatures above 32º C respectively (Figures 2.4, 2.10, 2.11, 2.12, 2.13) (Tables 2.1, 2.3). The same decline of populations was observed for June 2006, July 2006, and August 2006 which had 4, 14 and 14 days that had temperatures above 32º C respectively (Figures 2.5, 2.14, 2.15, 2.16, 2.17) (Tables 2.1, 2.4). During the summer months of 2007, CFUs of the introduced strain showed significant decline, but the populations of the native strains were not repressed to the same extent (Figures 2.6, 2.18, 2.19, 2.20, 2.21) (Tables 2.1, 2.5). And once again, in the summer of 2006, 2007 and 2008, the same pattern was observed (Figures 2.7, 2.22, 2.23, 2.24, 2.25) (Tables 2.1, 2.6).

Populations of M. anisopliae recovered as temperature became more moderate in the spring and autumn months

Major rebounds in *M. anisopliae*-GFP populations were recorded for the spring and autumn seasons when temperatures became more moderate. During April 2005 and November 2005 (Figures 2.4, 2.10, 2.11) (Table 2.1, 2.3), CFUs of the introduced strain increased significantly. In September 2006 (Figures 2.5, 2.17) (Table 2.1, 2.4) a significant increase in population was observed for the native colonies on the grass plots. In March 2007, May 2007 and October 2007 all four treatments showed various degrees of recovery in population size (Figures 2.6, 2.18, 2.19. 2.20, 2.21) (Table 2.1, 2.5). And finally in May 2008 the native strains on turf and bare plots showed recovery (Figures 2.7, 2.23, 2.24) (Table 2.1, 2.6).

Overall, the more days in a particular month with temperatures above 32° C (summer months), the lower the CFU counts for that month. Population counts increased as the temperature became milder in the spring and autumn months. Populations also dropped when temperatures fell below 0° C during the winter months. Intolerance to temperatures below 0° C and above 32° C impacted field populations each month, but high temperatures repressed population size more than extreme cold temperatures.

The decline of *M. anisopliae* CFUs may actually be the effect of repression of *Metarhizium* populations in response to how turf roots are adapting to temperature changes. Photosynthetic rates in bentgrass decreased as temperature increases (Huang *et al.*,1998). In addition, as temperatures increase to >32º C, grasses such as tall fescue become unproductive and go semi-dormant for several weeks in the summer (Burns and Chamblee, 2000). This may have an adverse affect on *M. anisopliae* populations, because this semi-dormant state (observed in high temperatures) causes the production of new roots to decrease and the number of dead roots to increase (Huang and Liu, 2003). Plants also provide a carbohydrate source for many microorganisms at the rhizosphere. This nutrition source may be required to maintain colonies of *M. anisopliae.* Loss of habitat and a food source may be contributing factors to *M. anisopliae* population dynamics.

While CFUs have shown an overall decline over four years, it is noteworthy that the transgenic fungal populations expressing GFP have persisted. The population trends observed for the introduced lab strain followed those for the native *Metarhizium* in the plots (Figure 2.2). While introduced strains initially suppressed native strains, suggesting a maximum carrying capacity for the soil, the native populations had recovered 3 months post application and subsequently native and introduced strains cycled in tandem. Both native and introduced strains were observed in all soil samples recovered from the field.

Populations of *Metarhizium* in bare soil followed the same trends as those in the turf plots. However, it was determined that within two months of the initial tilling to clear the plots of roots, roots had regrown from the buffer zone. The measurements taken from the bare plots, therefore, do not derive from a rootless environment.

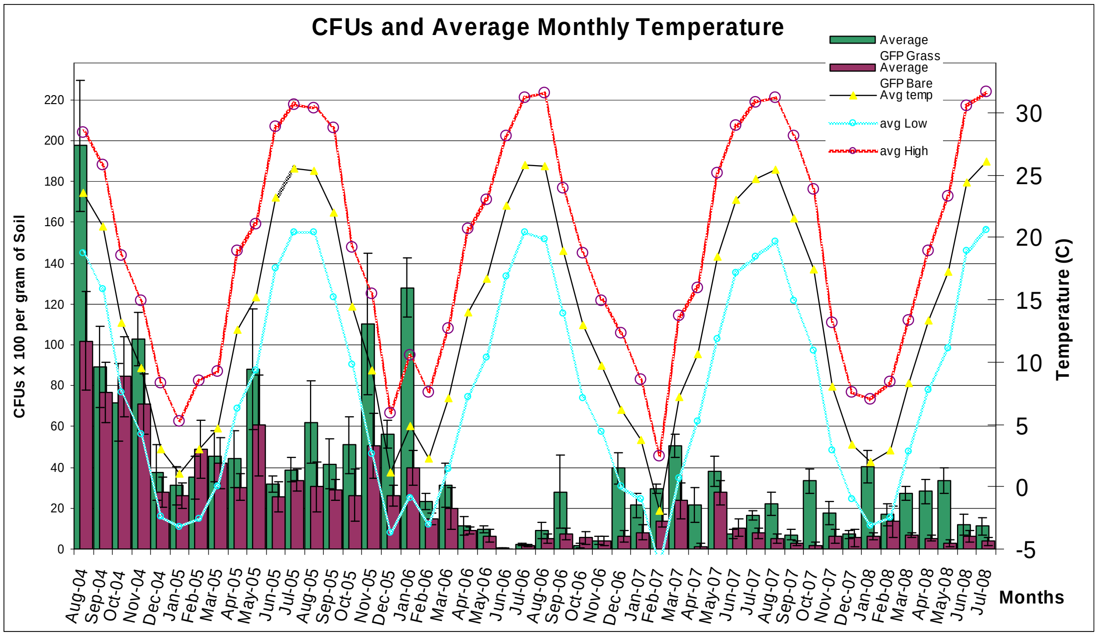


Figure 2.2 ***M. anisopliae* CFUs cycled with seasonal changes.** Counts of CFUs increased as temperatures moderated. Populations declined in the heat of summer, CFUs were the lowest each year in summer months when temperatures were >32º C and recovered in fall as temperatures became more mild. Summer decline in *Metarhizium* populations may have been in response to plant physiology, tall fescue grass reduced photosynthetic rates and becomes semi-dormant, the number of new root formation decreases, the number of dead roots increase and there are also fewer herbivorous insects present at temperatures >32º C. Similar declines in populations were observed in colder winter months, but not to the same extent as the summer decline, and may have been attributed to decreased photosynthetic rates in plants and that *Metarhizium* is a mesophylic fungus that is most physiological active at temperatures of 25-30º C. As the temperatures became milder in the spring, populations recovered. For the 4 year field study, an analysis of variance (ANOVA) analysis was performed using SAS to compare gfp-labeled *Metarhizium* on bare and grass plots to native *Metarhizium* recovered from bare and grass plots ($p<0.05$). Survivorship in field conditions of these recovered strains were analyzed using proc mixed protocol after data were transformed using log10+1 to satisfy assumptions of normality and equal variance. Samples were analyzed by calendar year.

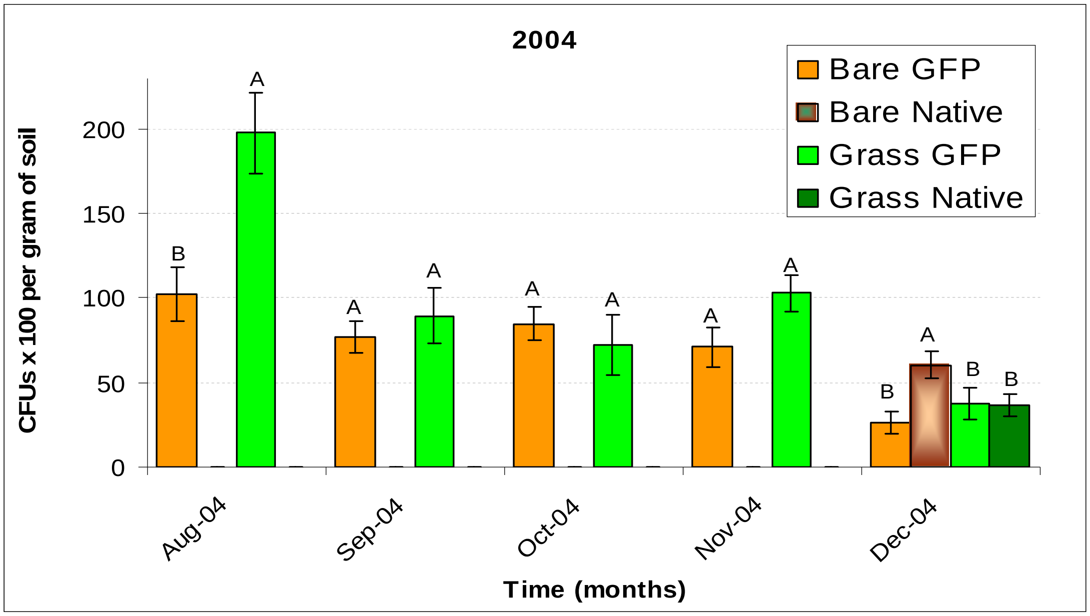


Figure 2.3 **Comparisons of Population Patterns Between Treatments for 2004**. After initial application of fungal conidia in August 2004, *Metarhizium*-GFP retrieved from the bare plots (Bare GFP) has significantly fewer CFUs than samples retrieved from turf plots (Grass GFP). After the initial application, population counts were not significantly different for the rest of 2004. This effect may have been caused by grass roots filling in the areas of the bare plots (even though the grass blades continued to be trimmed above soil level), providing a habitat for the fungus. Populations of native fungus on turf plots (Grass Native) were observed titers in equivalent to the introduced strains December 2004. Native *Metarhizium* recovered for the bare plots (Bare Native) were significantly higher that those recovered from the other three plots in December 2004. Summer decline in *Metarhizium* populations may have been in response to plant physiology, tall fescue goes semi-dormant, the number of new root formation decreases, the number of dead roots increase, there are also fewer herbivorous insects present at temperatures >32º C. Similar declines in populations were observed in colder winter months, but not to the same extent as the summer decline, and may have been attributed to decreased photosynthetic rates in plants and that *Metarhizium* is a mesophylic fungus that is most physiological active at temperatures of 25-30º C. For the 4 year field study, an analysis of variance (ANOVA) analysis was performed using SAS to compare gfp-labeled *Metarhizium* on bare and grass plots to native *Metarhizium* recovered from bare and grass plots. Survivorship in field conditions of these recovered strains were analyzed using proc mixed protocol after data were transformed using log10+1 to satisfy assumptions of normality and equal variance. Population of all four groups, native *Metarhizium* on turf, *Metarhizium*-GFP on turf, native *Metarhizium* on bare soil and *Metarhizium*-GFP on bare soil were compared to each other for each year from 2004-2008. Different letter within each month indicate significant differences for that month amongst the four groups ($p<0.05$)

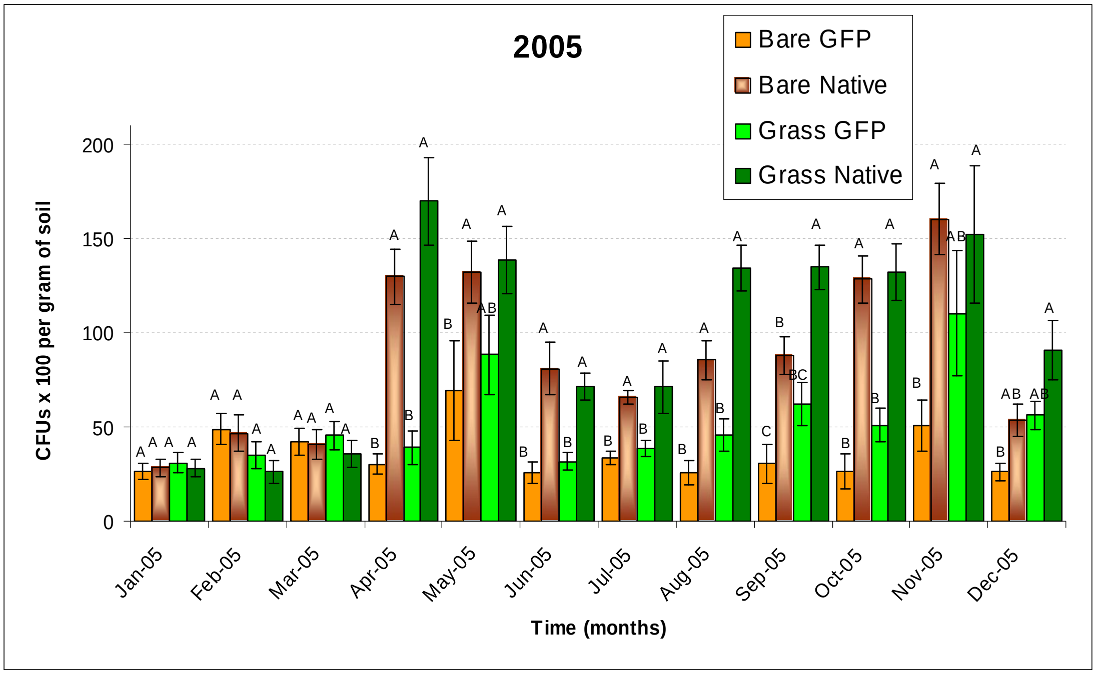


Figure 2.4 **Comparisons of Population Patterns Between Treatments for 2005**. CFUs for native *Metarhizium* strains recovered from the bare (Bare Native) and turf plots (Grass Native) and were not significantly different that the CFUs recovered for the introduced *Metarhizium*-GFP from the turf (Grass GFP) and bare (Bare GFP) plots until April 2005. CFUs for native *Metarhizium* continued to remain higher than the introduced strain for the rest of the year. The introduced strains showed significantly lowers CFUs than the native strains during the summer months beginning in April 2005 until the end of the year. For the 4 year field study, an analysis of variance (ANOVA) analysis was performed using SAS to compare gfp-labeled *Metarhizium* on bare and grass plots to native *Metarhizium* recovered from bare and grass plots. Survivorship in field conditions of these recovered strains were analyzed using proc mixed protocol after data were transformed using log10+1 to satisfy assumptions of normality and equal variance. Samples were analyzed by calendar year. Populations of all four groups, native *Metarhizium* on turf, *Metarhizium*-GFP on turf, native *Metarhizium* on bare soil and *Metarhizium*-GFP on bare soil were compared to each other for each year from 2004-2008. Different letter within each month indicate significant differences for that month amongst the four groups ($p<0.05$)

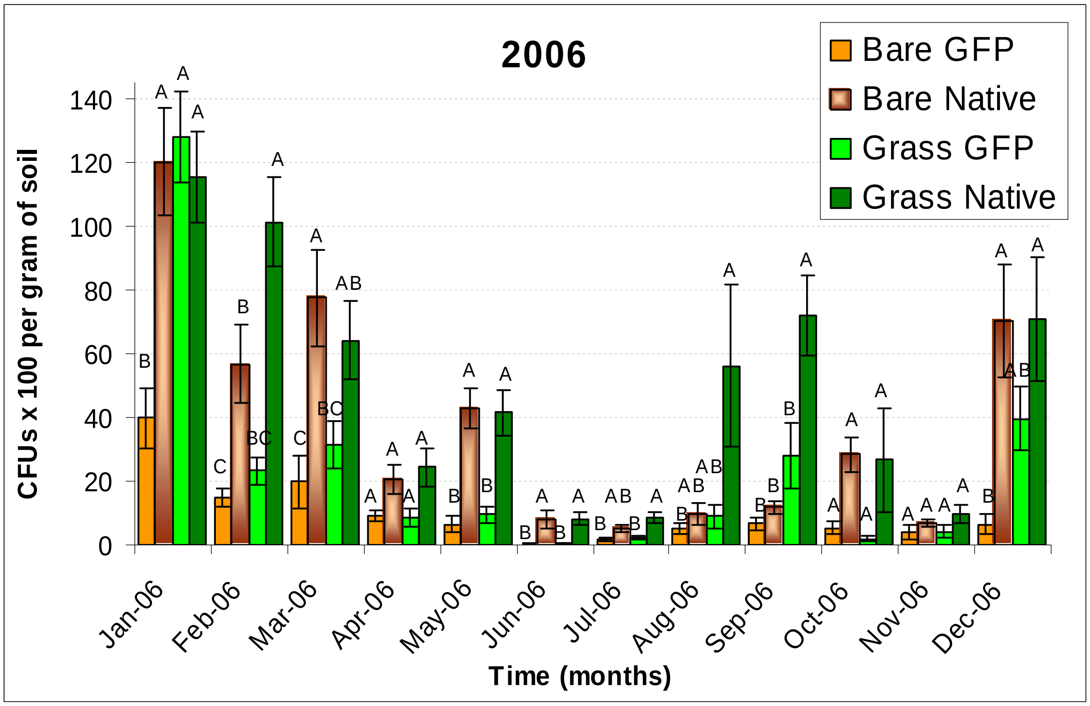


Figure 2.5 **Comparisons of Population Patterns Between Treatments for 2006**. CFUs for native *Metarhizium* populations and the introduced strain cycled in similar patterns. Populations of native strains were significantly higher than introduced strains in all months but January 2006, April 2006 and November 2006. All populations were repressed during the summer. For most of the treatments titers were up to 10 times lower in June and July 2006, than for January. For the 4 year field study, an analysis of variance (ANOVA) analysis was performed using SAS to compare gfp-labeled *Metarhizium* on bare and grass plots to native *Metarhizium* recovered from bare and grass plots. Survivorship in field conditions of these recovered strains were analyzed using proc mixed protocol after data were transformed using log10+1 to satisfy assumptions of normality and equal variance. Samples were analyzed by calendar year. Populations of all four groups, native *Metarhizium* on turf, *Metarhizium*-GFP on turf, native *Metarhizium* on bare soil and *Metarhizium*-GFP on bare soil were compared to each other for each year from 2004-2008. Different letter within each month indicate significant differences for that month amongst the four groups ($p<0.05$)

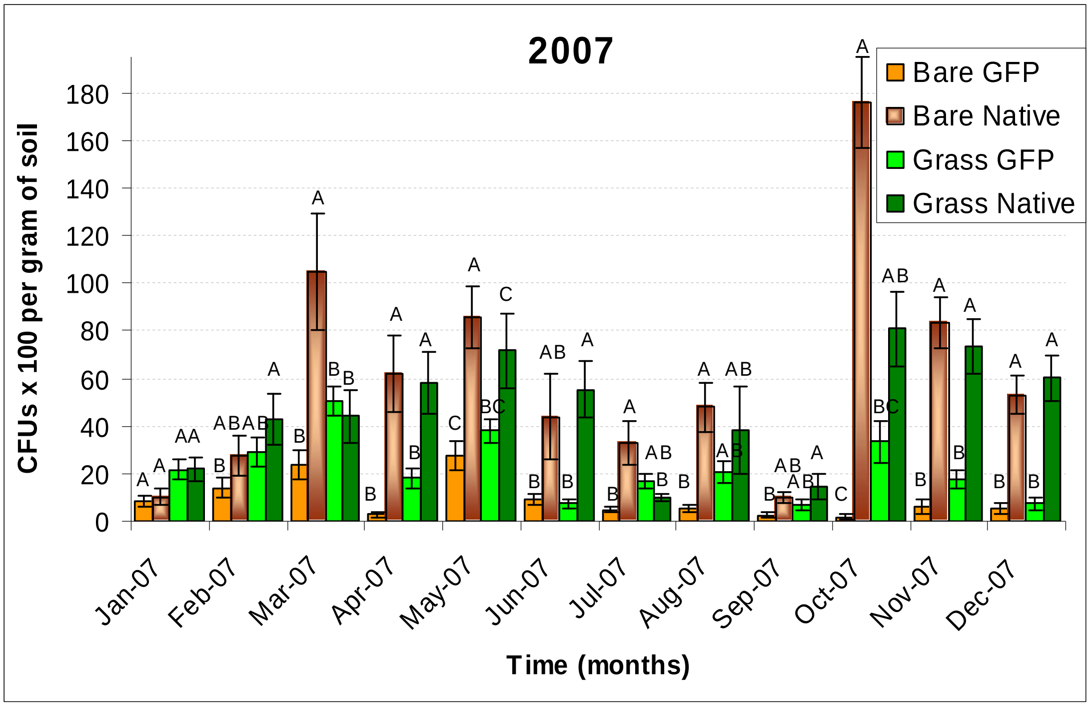


Figure 2.6 **Comparisons of Population Patterns Between Treatments for 2007**. CFUs for *Metarhizium* populations of the introduced strains showed summer repression of CFUs. Native strains did not demonstrate the same extent of decline in population that was observed in the introduced strains. CFUs of the introduced strains were significantly lower than the native strains in all months except January 2007. For the 4 year field study, an analysis of variance (ANOVA) analysis was performed using SAS to compare gfp-labeled *Metarhizium* on bare and grass plots to native *Metarhizium* recovered from bare and grass plots. Survivorship in field conditions of these recovered strains were analyzed using proc mixed protocol after data were transformed using log10+1 to satisfy assumptions of normality and equal variance. Samples were analyzed by calendar year. Populations of all four groups, native *Metarhizium* on turf, *Metarhizium*-GFP on turf, native *Metarhizium* on bare soil and *Metarhizium*-GFP on bare soil were compared to each other for each year from 2004-2008. Different letter within each month indicate significant differences for that month amongst the four groups ($p<0.05$)

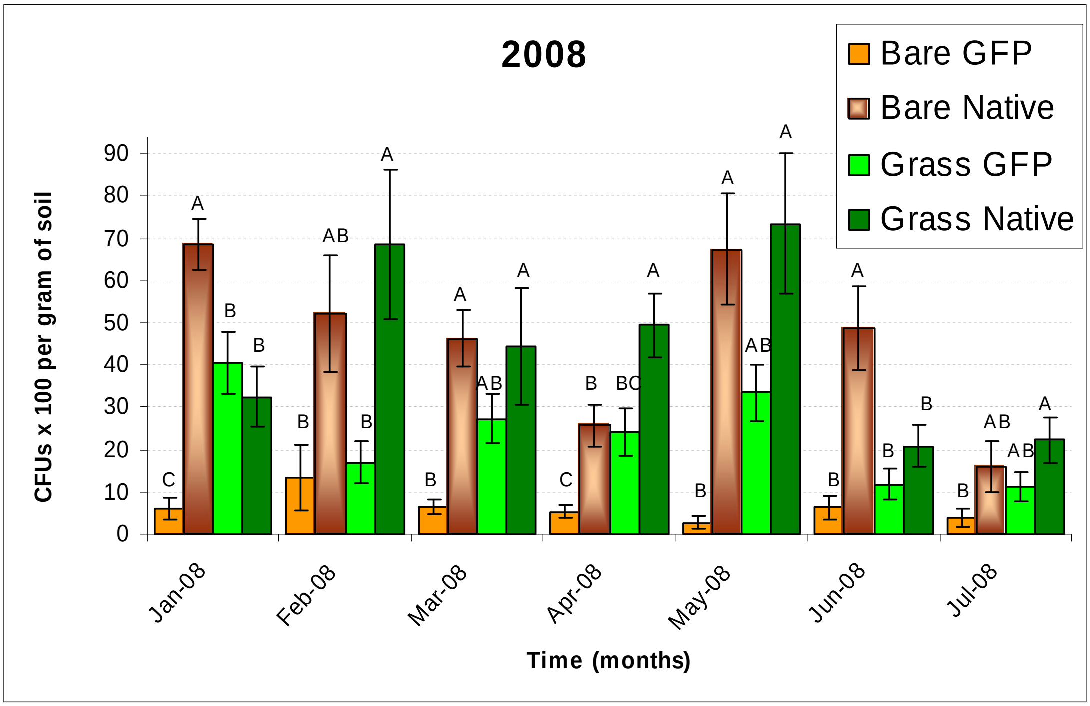


Figure 2.7 **Comparisons of Population Patterns Between Treatments for 2008**. CFUs for introduced *Metarhizium* populations on bare plots were significantly lower than the native strains from January until July 2008. CFUs for introduced *Metarhizium* populations on turf plots were significantly lower than the native strains in February 2008, April 2008 and June 2008. For the 4 year field study, an analysis of variance (ANOVA) analysis was performed using SAS to compare gfp-labeled *Metarhizium* on bare and grass plots to native *Metarhizium* recovered from bare and grass plots. Survivorship in field conditions of these recovered strains were analyzed using proc mixed protocol after data were transformed using log10+1 to satisfy assumptions of normality and equal variance. Samples were analyzed by calendar year. Populations of all four groups, native *Metarhizium* on turf, *Metarhizium*-GFP on turf, native *Metarhizium* on bare soil and *Metarhizium*-GFP on bare soil were compared to each other for each year from 2004-2008. Different letter within each month indicate significant differences for that month amongst the four groups ($p<0.05$)

Month	**Group Mean**	**f, df, p**
Aug 2004	149.80	11.31,29, 0.0022
Sept 2004	83.10	0.42,29,0.5210
Oct 2004	78.23	0.39,29,0.5368
Nov 2004	86.90	3.94, 29, 0.057
Dec 2004	40.30	3.33, 59,0.0260
Jan 2005	28.45	0.17, 59, 0.9164
Feb 2005	39.17	1.80, 59, 0.1569
Mar 2005	40.97	0.32,59, 0.8073
Apr 2005	92.18	21.87, 59, <.0001
May 2005	107.07	2.62, 59, 0.0597
June 2005	52.52	10.38, 59,<.0001
July 2005	52.48	11.13, 59,<.0001
Aug 2005	77.00	14.29, 59, <.0001
Sept 2005	78.77	15.80, 59, <.0001
Oct 2005	84.48	20.93, 59,<.0001
Nov 2005	118.27	3.41, 59, 0.0236
Dec 2005	56.73	7.03, 59, 0.0004
Jan 2006	100.77	8.75, 59, <.0001
Feb 2006	48.95	16.34, 59, <.0001
Mar 2006	48.23	5.84, 59, 0.0015
Apr 2006	15.63	3.58, 59, 0.0193
May 2006	25.10	14.96, 59, <.0001
June 2006	4.22	6.89, 59, 0.0005
July 2006	4.38	9.34, 59, <.0001
Aug 2006	19.9	3.51, 59, 0.020
Sept 2006	29.63	13.00, 59, <.000
Oct 2006	15.57	2.59, 59, 0.0616
Nov 2006	6.15	1.52, 59, 0.219
Dec 2006	46.82	4.67, 59, 0.0056
Jan 2007	15.50	3.70, 59, 0.0169
Feb 2007	28.42	2.37, 59, 0.080
Mar 2007	55.70	6.02, 59, 0.0013
Apr 2007	35.10	7.61, 59, 0.0002
May 2007	55.65	6.05, 59, 0.0012
June 2007	28.97	4.98, 59, 0.0039
July 2007	16.02	5.99, 59, 0.0013
Aug 2007	28.08	3.07, 59, 0.035
Sept 2007	8.58	2.60, 59, 0.0608
Oct 2007	72.93	34.21, 59, <.0001
Nov 2007	45.02	22.12, 59, <.0001
Dec 2007	31.47	20.31, 59, <.0001
Jan 2008	36.93	17.82, 59, <.000
Feb 2008	37.75	4.97, 59, 0.0040
Mar 2008	31.18	5.12, 59, 0.0034
Apr 2008	26.17	11.45, 59, <.0001
May 2008	44.27	8.71, 59, <.0001
June 2008	21.90	10.05, 59, <.0001
July 2008	13.25	3.03, 59, 0.0366

Table 2.1 **Tukey's Studentized Range (HSD) Test for Group Means for Each Month**. A group mean was calculated for each month for the four conditions, Native *Metarhizium* on bare plots, native *Metarhizium* on turf plots, *Metarhizium*-GFP on bare plots and *Metarhizium*-GFP on turf plots. F, degrees of freedom (df) and p values were calculated for the group means for each month. Values that were significantly different ($p<0.05$) than the calculated monthly group means are marked in Figures 2.3-2.7.

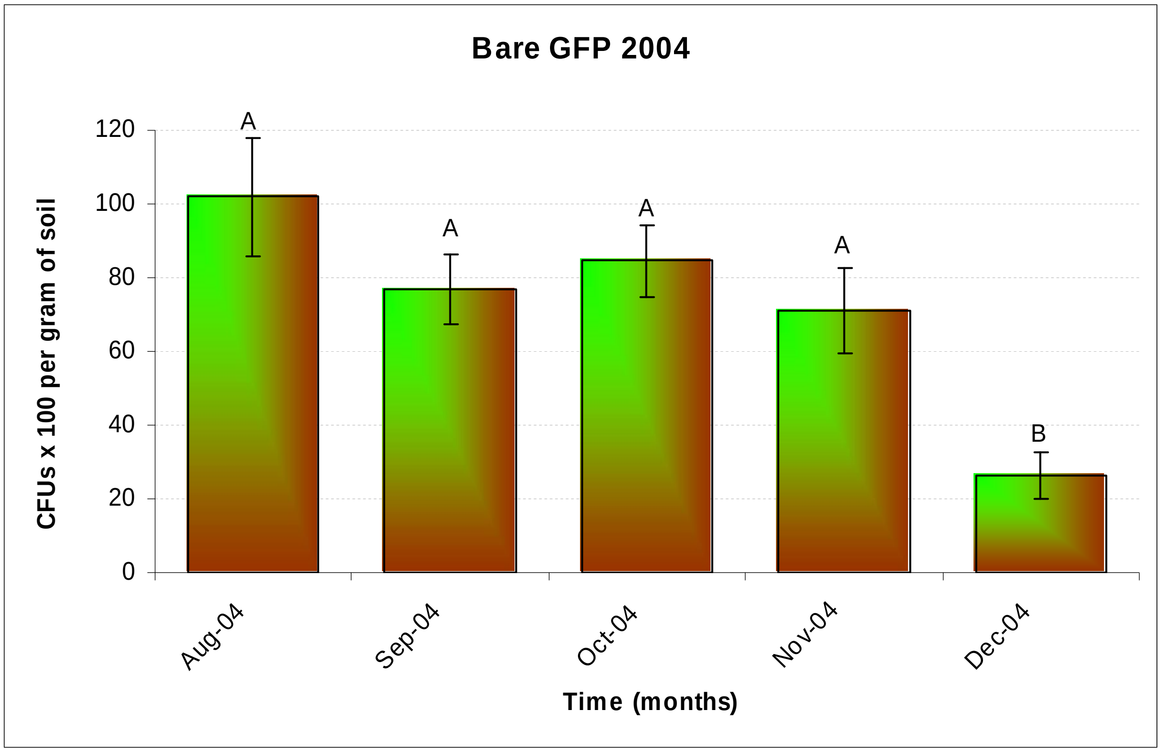


Figure 2.8 **Population Patterns for *Metarhizium*-GFP Isolated from Bare Plots in 2004**. An ANOVA analysis was preformed for *Metarhizium*-GFP recovered from bare plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. Populations of the introduced strain on bare soil were significantly lower in December 2004.

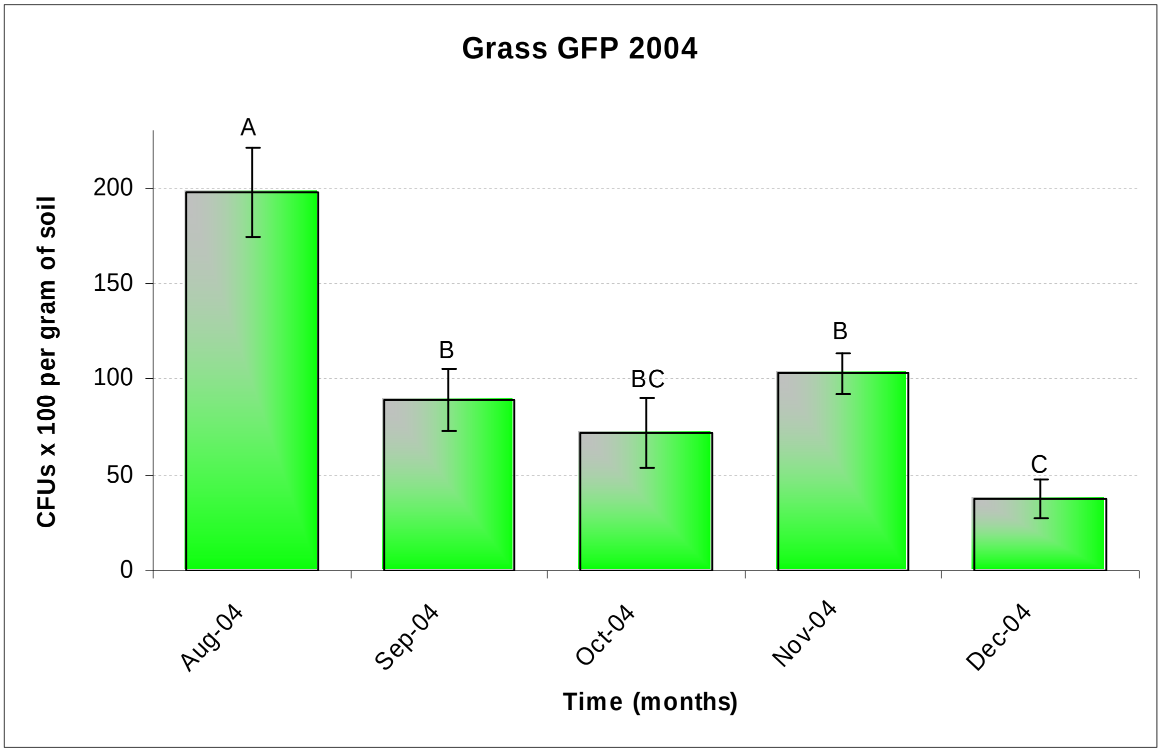


Figure 2.9 **Population Patterns for *Metarhizium*-GFP Isolated from Turf Plots in 2004**. An ANOVA analysis was preformed for *Metarhizium*-GFP recovered from turf plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. Populations began to decrease after the initial inoculation in August 2007. They were significantly lower in September 2004, and another significant decrease in CFUs was observed in December 2004.

	Bare GFP Mean	**Grass GFP Mean**	**Bare Native**	**Grass Native**
Yearly mean by treatment (f,df,p)	72.17 (6.40, 4, 0.0002)	99.83 (13.18, 4, <0.0001)		
8/04	101.93 ± 15.92 A	197.67 ± 23.61 A		
9/04	76.93 ± 9.49 A	89.27 ± 16.43 B		
10/04	84.60 ± 9.83 A	71.87 ± 17.83 BC		
11/04	70.93 ± 11.65 A	102.87 ± 11.09 B		
12/04	26.47 ± 6.35 B	37.47 ± 9.71 C	60.53 ± 8.17	36.73 ± 6.92

Table 2.2 **Tukey's Studentized Range (HSD) Test for Yearly Means for Each Treatment for 2004.** Population dynamics were examined for 2004 for each treatment listed in each column. Significant differences ($p<0.05$)in CFUs for each month based on treatment are designated with a different letter.

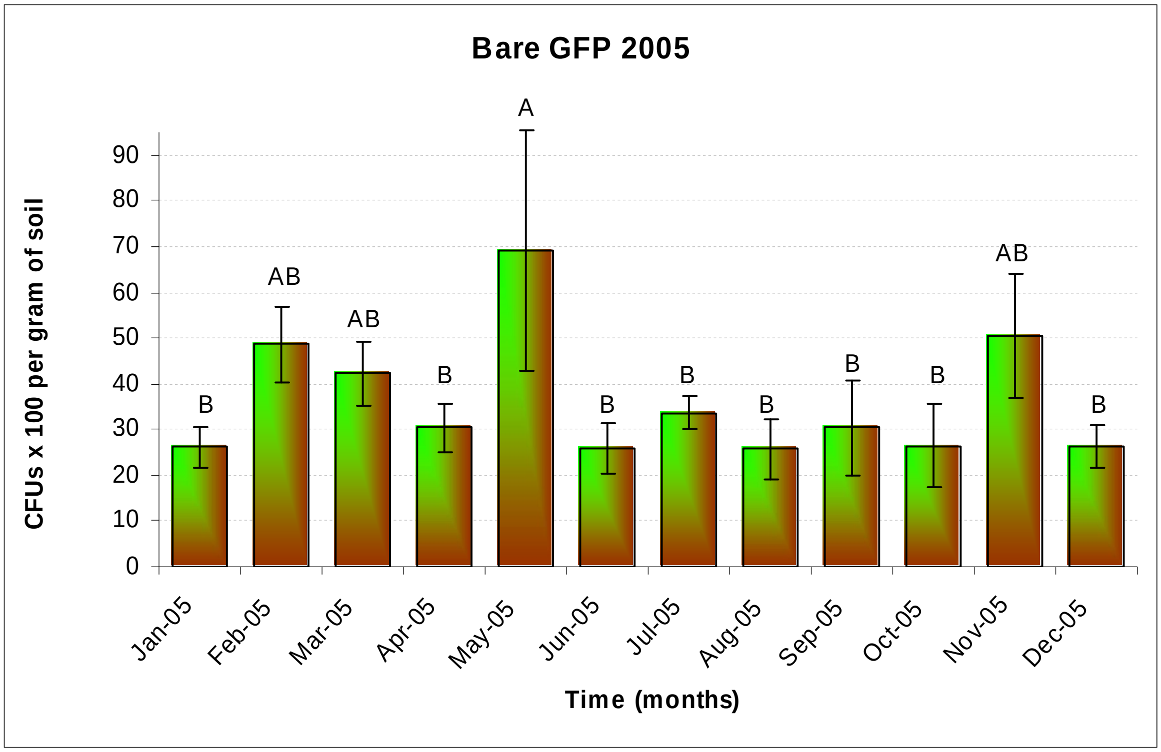


Figure 2.10 **Population Patterns for *Metarhizium*-GFP Isolated from Bare Plots in 2005**. An ANOVA analysis was preformed for *Metarhizium*-GFP recovered from bare plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. There was a significant increase in the population in May 2005. After that, populations were significantly lower for the rest of the year.

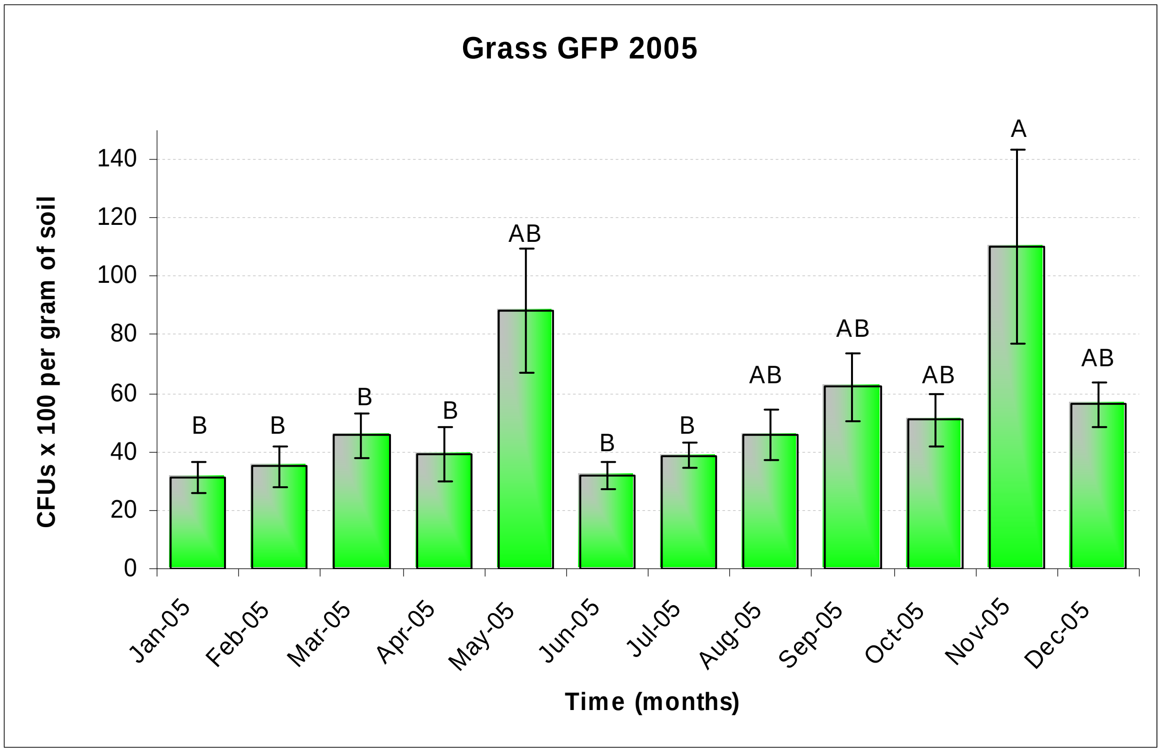


Figure 2.11 **Population Patterns for *Metarhizium*-GFP Isolated from Turf Plots in 2005**. An ANOVA analysis was preformed for *Metarhizium*-GFP recovered from turf plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. A slight increase in the population was observed in May 2005, and a significant increase in CFUs was observed in November 2005.

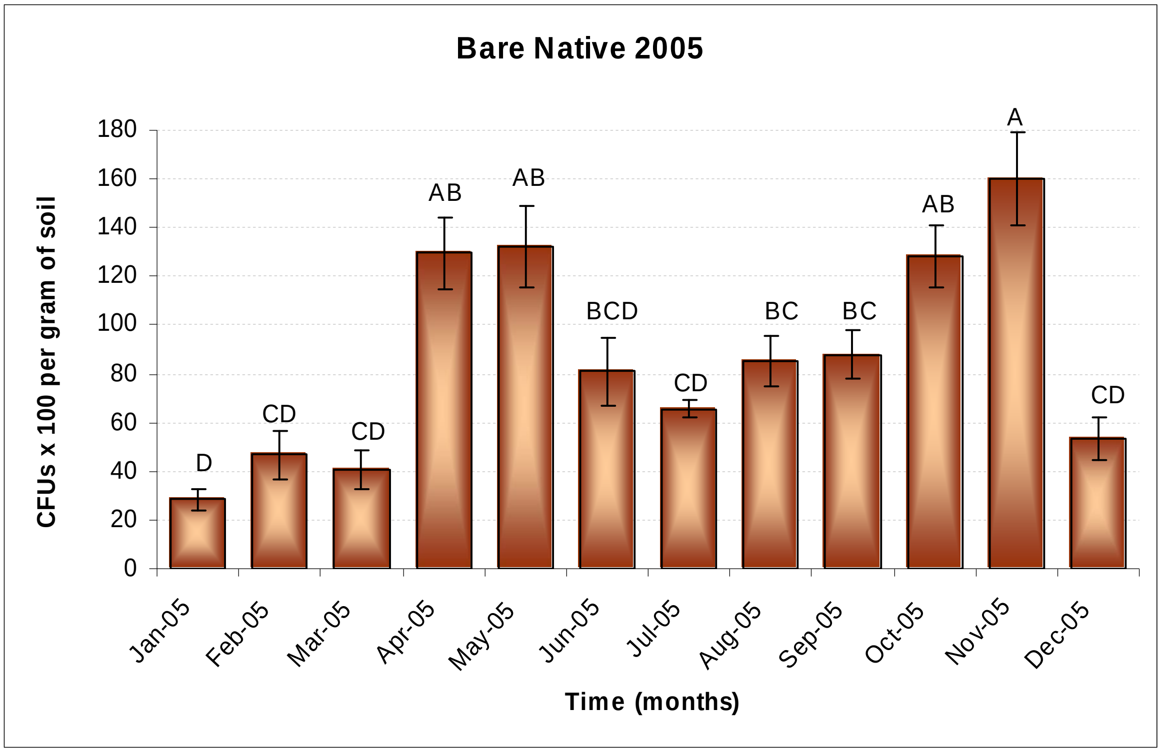


Figure 2.12 **Population Patterns for Native *Metarhizium* Isolated from Bare Plots in 2005**. An ANOVA analysis was preformed for native *Metarhizium* recovered from bare plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. CFUs in the spring and autumn months of April 2005, May 2005, October 2005 and November 2005 were significantly higher than the other months of 2005.

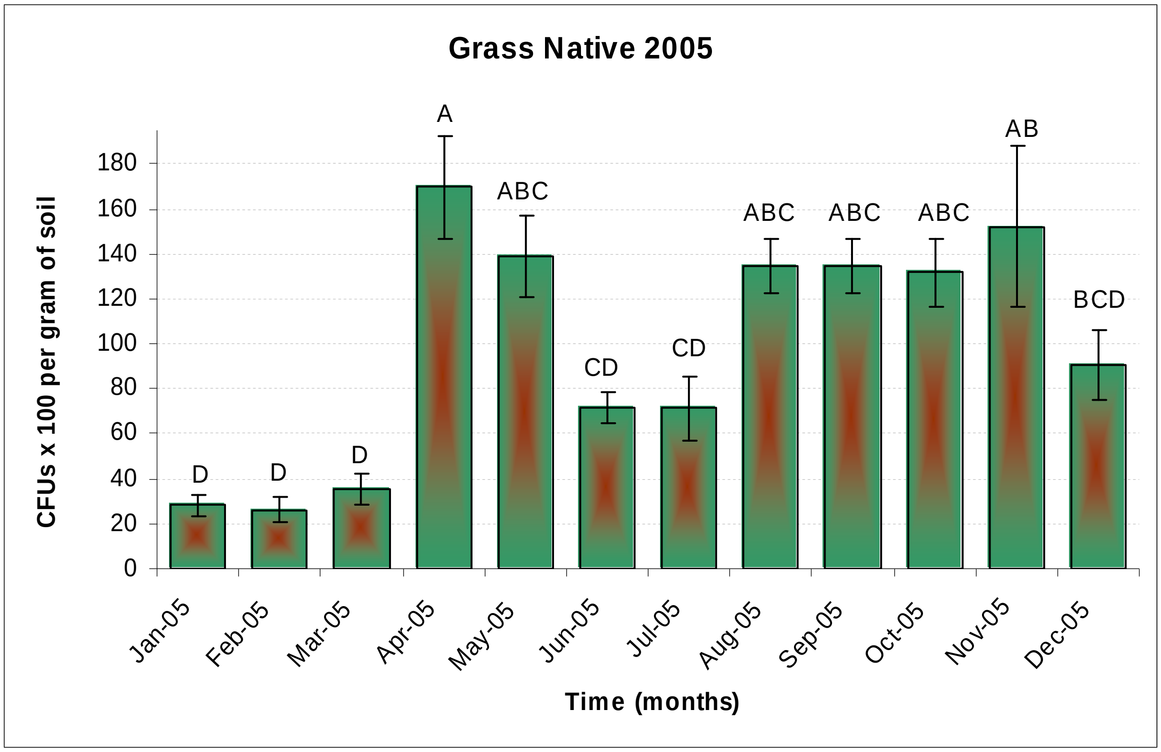


Figure 2.13 **Population Patterns for Native *Metarhizium* Isolated from Turf Plots in 2005**. An ANOVA analysis was preformed for native *Metarhizium* recovered from turf plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. Populations were significantly higher in spring and autumn months of April 2005, May 2005 and November 2005.

	Bare GFP Mean	**Grass GFP Mean**	**Bare Native Mean**	**Grass Native Mean**
Yearly mean by treatment (f,df,p)	36.69 (1.56, 11, 0.1153)	54.26 (3.14, 11, 0.0007)	86.82 (12.92, 11, <0.0001)	98.85 (9.94, 11, <0.0001)
1/05	26.27 ±4.45 B	31.00 ± 5.25 B	28.33 ± 4.43 D	28.20 ± 4.70 D
2/05	48.73 ± 8.24 AB	35.00 ± 7.01 B	46.67 ± 9.64 CD	26.27 ± 5.92 D
3/05	42.27 ± 6.95 AB	45.53 ± 7.58 B	40.53 ± 7.73 CD	35.53 ± 7.00 D
4/05	30.33 ± 5.31 B	39.07 ± 9.12 B	129.67 ± 14.79 AB	169.67 ± 22.95 A
5/05	69.33 ± 26.29 B	88.27 ± 20.94 AB	132.13 ± 16.44 AB	138.53 ± 18.13 ABC
6/05	25.67 ± 5.51 B	31.67 ± 4.71 B	81.07 ± 14.06 BCD	71.67 ± 7.07 CD
7/05	33.67 ± 3.55 B	38.67 ± 4.40 B	65.47 ± 3.55 CD	72.13 ± 9.34 CD
8/05	32.07 ± 6.64 B	45.73 ± 8.72 AB	85.40 ± 10.30 BC	71.20 ± 13.84 ABC
9/05	30.40 ± 10.36 B	62.13 ± 11.75 AB	87.87 ± 10.10 BC	134.67 ± 12.01 ABC
10/05	26.47 ± 9.19 B	51.00 ± 9.05 AB	128.47 ± 12.60 AB	132.00 ±15.09 ABC
11/05	50.47 ± 13.61 AB	110.13 ± 33.33 A	160.20 ± 18.89 A	152.27 ± 36.20 AB
12/05	26.27 ± 4.49 B	56.13 ± 7.60 AB	53.60 ± 8.89 CD	90.93 ± 15.58 BCD

Table 2.3 **Tukey's Studentized Range (HSD) Test for Means for Each Treatment for 2005.** Population dynamics were examined for each treatment. Each column lists the monthly means for each treatment. Significant differences ($p<0.05$)in CFUs for each month based on treatment are designated with a different letter.

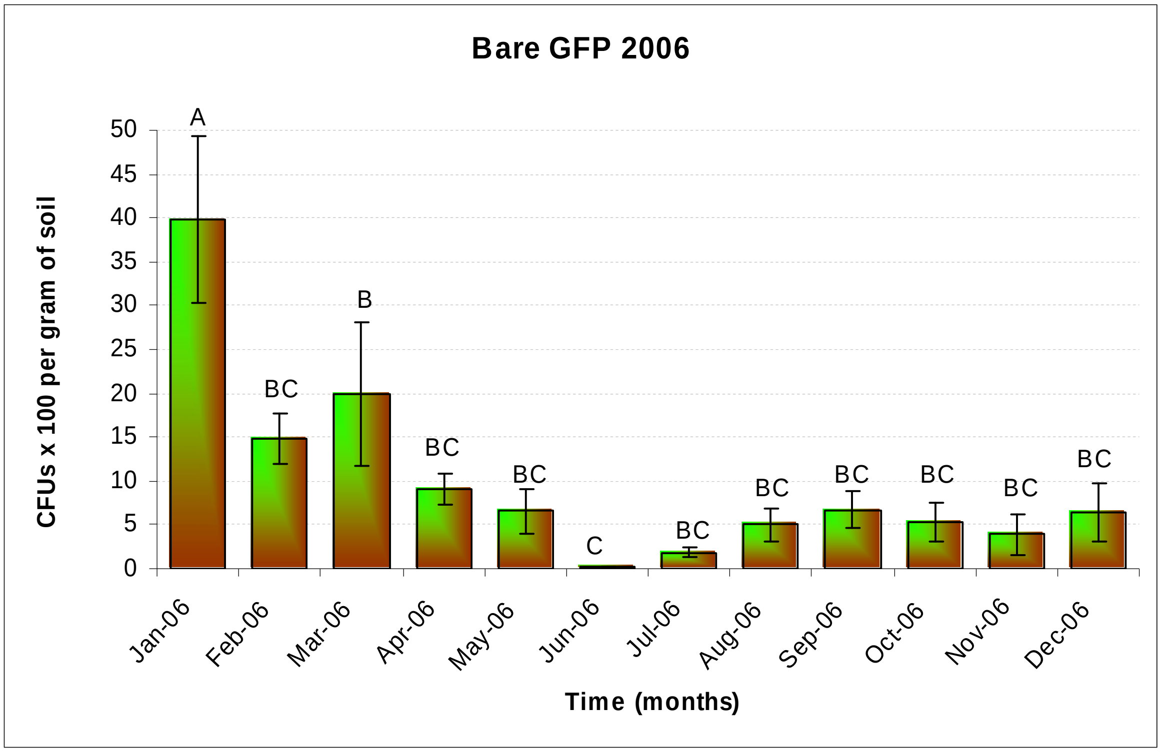


Figure 2.14 **Population Patterns for *Metarhizium*-GFP Isolated from Bare Plots in 2006**. An ANOVA analysis was preformed for *Metarhizium*-GFP recovered from bare plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. Populations were the highest in January 2006 and began to decrease until June 2006 when the fewest CFUS were observed. Population began to slowly recover after June 2006.

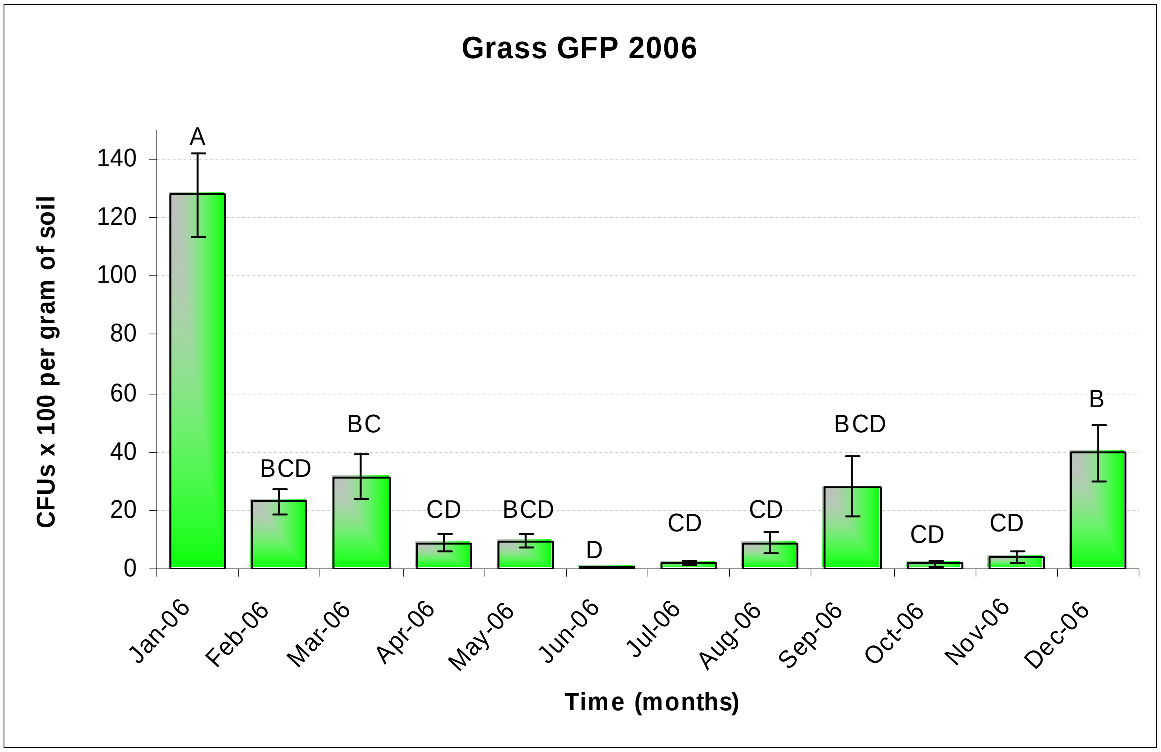


Figure 2.15 **Population Patterns for *Metarhizium*-GFP Isolated from Turf Plots in 2006**. An ANOVA analysis was preformed for *Metarhizium*-GFP recovered from turf plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. Populations were the highest in January 2006 and began to decrease until June 2006 when the fewest CFUS were observed. Population began to slowly recover after June 2006.

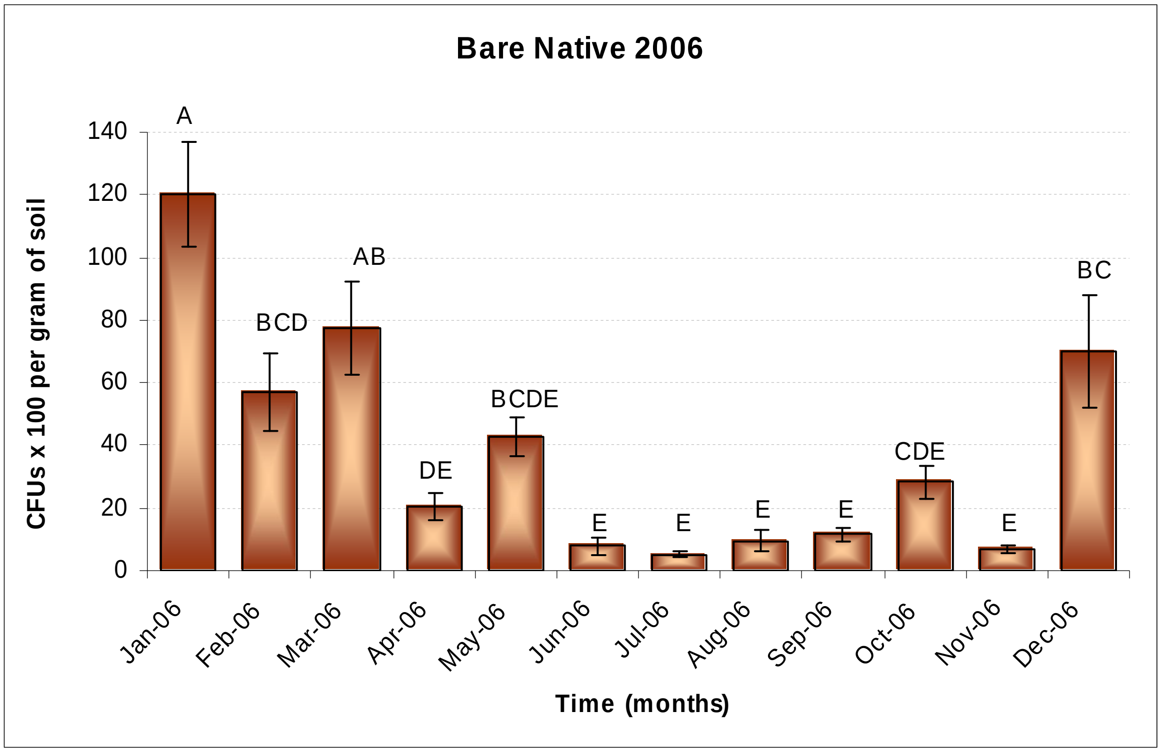


Figure 2.16 **Population Patterns for Native *Metarhizium* Isolated from Bare Plots in 2006**. An ANOVA analysis was preformed for native *Metarhizium* recovered from bare plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. Populations were the highest in January 2006 and began to decrease until June 2006. Populations remained repressed until December 2006.

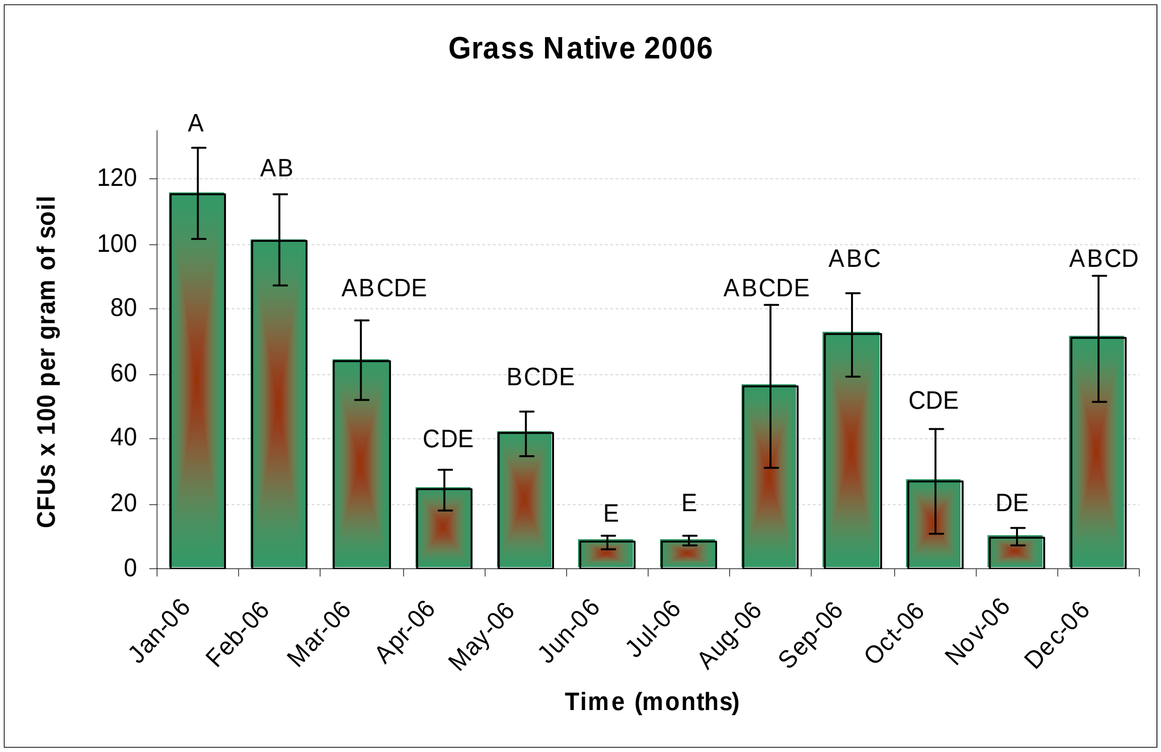


Figure 2.17 **Population Patterns for Native *Metarhizium* Isolated from Bare Plots in 2006**. An ANOVA analysis was preformed for native *Metarhizium* recovered from turf plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. Populations were the highest in January 2006 and began to decrease until June 2006 when the fewest CFUS were observed. Population began to recover after July 2006.

	Bare GFP Mean	**Grass GFP Mean**	**Bare Native Mean**	**Grass Native Mean**
Yearly mean by treatment (f,df,p)	9.9 (6.81, 11, <0.0001)	23.81 (29.31, 11, <0.0001)	38.10 (14.49, 11, <0.0001)	49.90 (7.54, 11, <0.0001)
1/06	39.80 ± 9.55 A	127.87 ±14.10 A	120.07 ± 16.70 A	115.33 ± 14.08 A
2/06	14.80 ± 2.87 AB	23.20 ± 4.33 BCD	56.73 ± 12.40 BCD	101.07 ± 13.97 AB
3/06	19.93 ± 8.27 B	31.33 ± 7.62 BC	77.53 ± 15.04 AB	64.13 ± 12.19 ABCDE
4/06	9.07 ± 1.74 BC	8.67 ± 3.00 CD	20.47 ± 4.54 DE	24.33 ± 6.19 CDE
5/06	6.53 ± 2.45 BC	9.60 ± 2.53 BCD	42.73 ± 6.44 BCDE	41.53 ± 7.08 BCDE
6/06	0.20 ± 0.11 C	0.47 ± 0.19 D	8.00 ± 2.77 E	8.20 ± 1.98 E
7/06	1.87 ± 0.62 C	2.07 ± 0.59 CD	5.07 ± 0.95 E	8.53 ± 1.60 E
8/06	5.07 ± 1.90 BC	8.87 ± 3.80 CD	9.60 ± 3.27 E	56.13 ± 25.29 ABCDE
9/06	6.67 ± 2.08 BC	28.07 ± 10.15 BCD	11.73 ± 2.14 E	72.07 ± 12.65 ABC
10/06	5.40 ± 2.22 BC	1.80 ± 0.83 CD	28.33 ± 5.35 CDE	26.73 ± 16.26 CDE
11/06	3.93 ± 2.37 BC	4.20 ± 2.01 CD	6.73 ± 1.29 E	9.73 ± 2.81 DE
12/06	6.47 ± 3.31 BC	39.60 ± 9.80 B	70.20 ± 17.92 BC	71.00 ± 19.36 ABCD

Table 2.4 **Tukey's Studentized Range (HSD) Test for Means for Each Treatment for 2006.** Population dynamics were examined for each treatment. Each column lists the monthly means for each treatment. Significant differences ($p<0.05$)in CFUs for each month based on treatment are designated with a different letter.

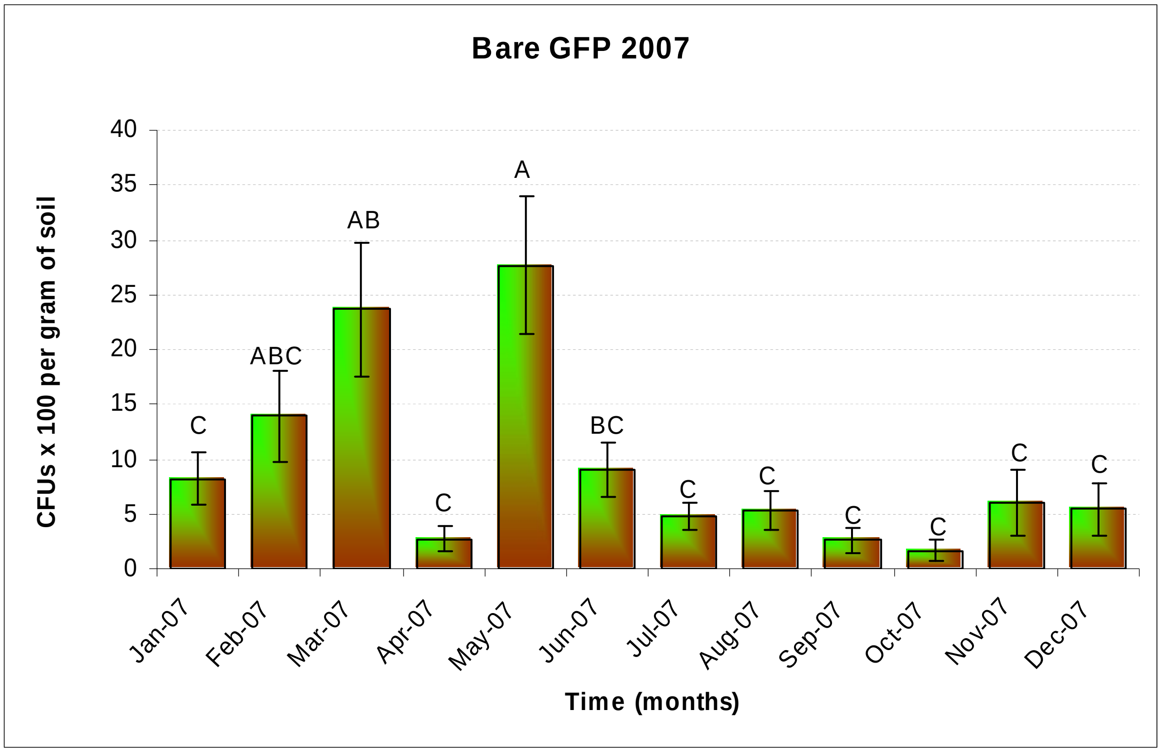


Figure 2.18 **Population Patterns for *Metarhizium*-GFP Isolated from Bare Plots in 2007**. An ANOVA analysis was preformed for *Metarhizium*-GFP recovered from bare plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. Peaks in population were observed in February 2007, March 2007 and May 2007. Populations for the other times of the year were significantly lower.

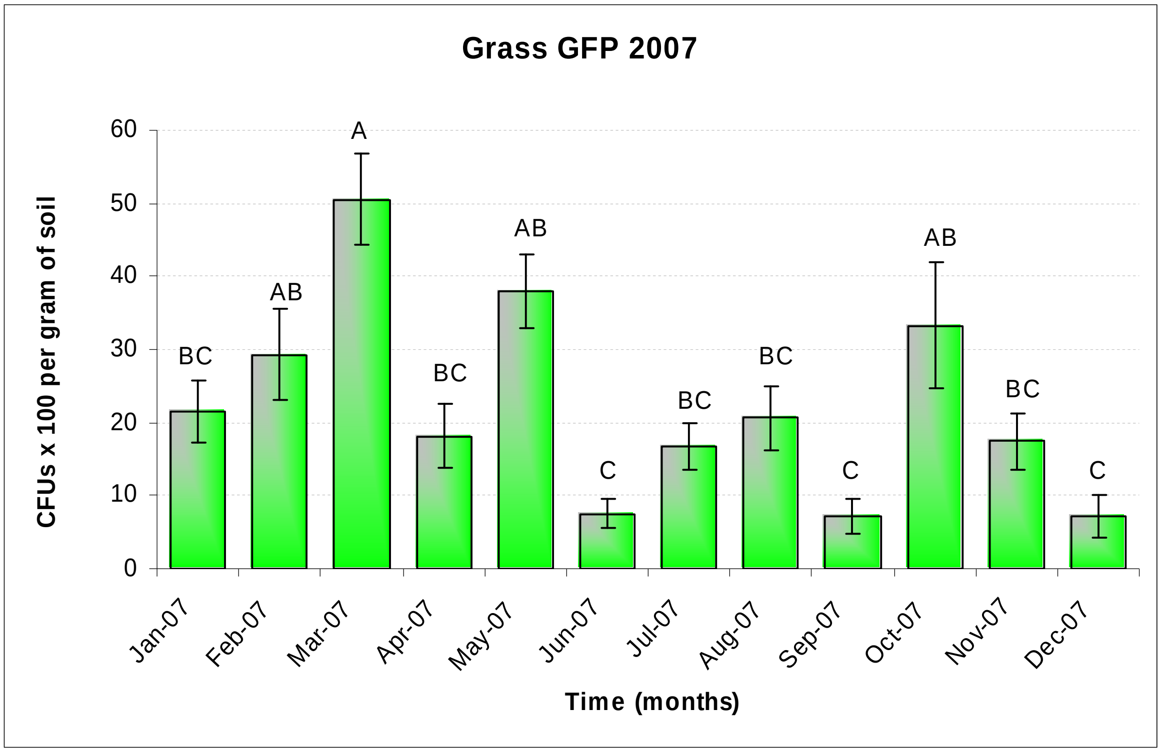


Figure 2.19 **Population Patterns for *Metarhizium*-GFP Isolated from Turf Plots in 2006**. An ANOVA analysis was preformed for *Metarhizium*-GFP recovered from turf plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. Populations were the highest in March 2007 and began to decrease until June 2006 when the fewest CFUS were observed. Populations began to slowly recover after June 2006 and were significantly higher by October 2007, but began to fall again and were significantly lower by December 2007.

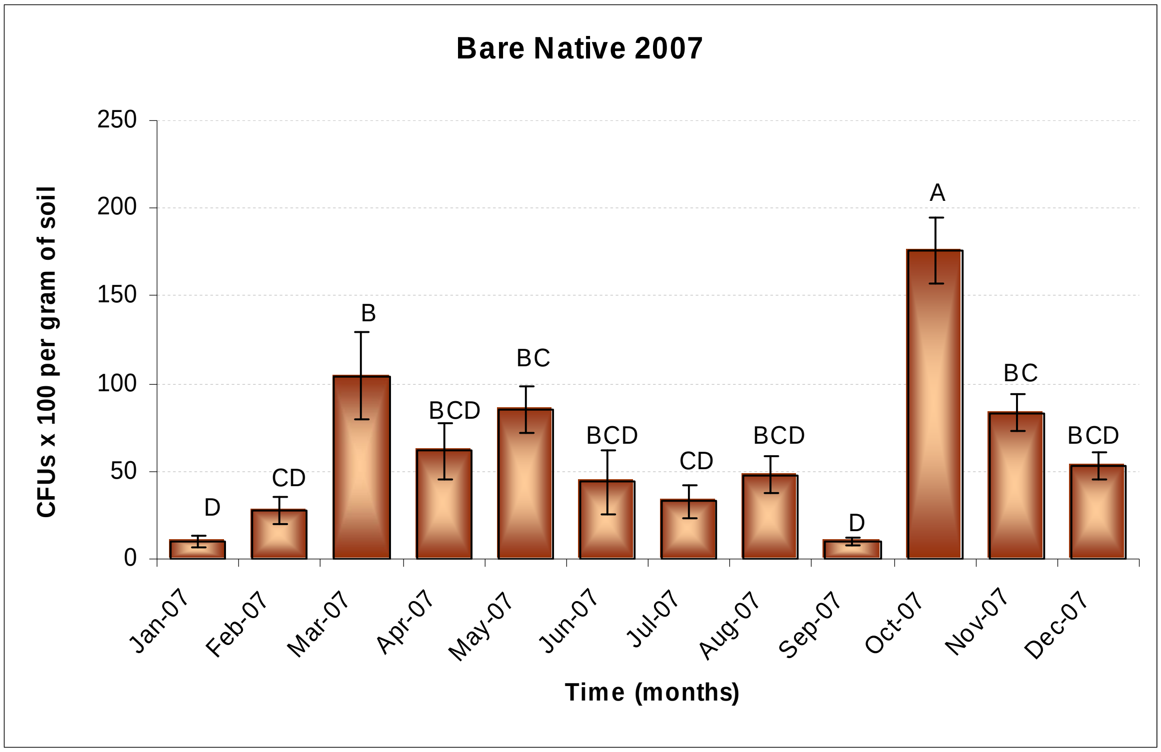


Figure 2.20 **Population Patterns for Native *Metarhizium* Isolated from Bare Plots in 2007**. An ANOVA analysis was preformed for native *Metarhizium* recovered from bare plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. Populations were their highest in October 2007. There was also a second peak observed in March 2007.

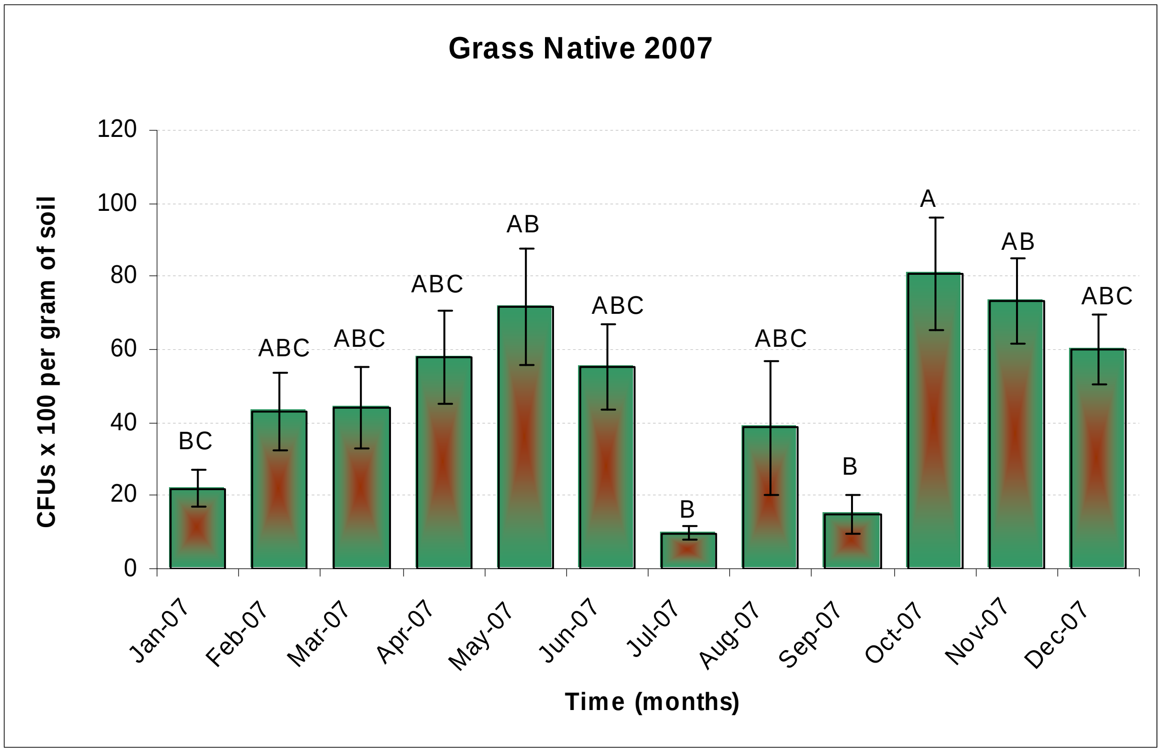


Figure 2.21 **Population Patterns for Native *Metarhizium* Isolated from Turf Plots in 2007**. An ANOVA analysis was preformed for native *Metarhizium* recovered from turf plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. CFUs for January 2007, July 2007 and September 2008 were significantly ($p<0.05$) lower than the other months examined.

	Bare GFP Mean	**Grass GFP Mean**	**Bare Native Mean**	**Grass Native Mean**
Yearly mean by treatment (f,df,p)	9.25 (6.62,11 <0.0001)	22.28 (7.79, 11, <1.0001)	61.34 (11.98, 11 <0.0001)	47.61 (3.92, 11,<0.0001)
1/07	8.20 ± 2.35 C	21.53 ± 4.25 BC	10.27 ± 3.24 D	22.00 ± 4.83 BC
2/07	13.93 ± 4.13 ABC	29.27 ± 6.24 AB	27.53 ± 8.07 CD	42.93 ± 10.78 ABC
3/07	23.67 ± 6.06 AB	50.53 ± 6.20 A	104.53 ± 24.40 B	44.07 ± 11.01 ABC
4/07	2.73 ± 1.12 C	18.07 ± 4.37 BC	61.73 ± 16.22 BCD	57.87 ± 12.88 ABC
5/07	27.67 ± 6.24 A	37.93 ± 5.05 AB	85.47 ± 13.14 BC	71.53 ± 16.00 AB
6/07	9.07 ± 2.46 BC	7.53 ± 1.90 C	43.87 ± 18.15 BCD	55.40 ± 11.70 ABC
7/07	4.73 ± 1.24 C	16.67 ± 3.14 BC	32.87 ± 9.27 CD	9.80 ± 1.71 B
8/07	5.27 ± 1.81 C	20.67 ± 4.39 BC	47.87 ± 10.46 BCD	38.53 ± 18.30 ABC
9/07	2.60 ± 1.14 C	7.13 ± 2.31 C	9.73 ± 2.19 D	14.87 ± 5.37 B
10/07	1.67 ± 1.03 C	33.27 ± 8.55 AB	176.00 ± 19.06 A	80.80 ± 15.43 A
11/07	6.07 ± 3.00 C	17.47 ± 3.86 BC	83.20 ± 10.53 BC	73.33 ± 11.77 AB
12/07	5.40 ± 2.46 C	7.27 ± 2.92 C	53.00 ± 7.89 BCD	60.20 ± 9.53 ABC

Table 2.5 **Tukey's Studentized Range (HSD) Test for Means for Each Treatment for 2007.** Population dynamics were examined for each treatment. Each column lists the monthly means for each treatment. Significant differences (p<0.05) in CFUs for each month based on treatment are designated with a different letter.

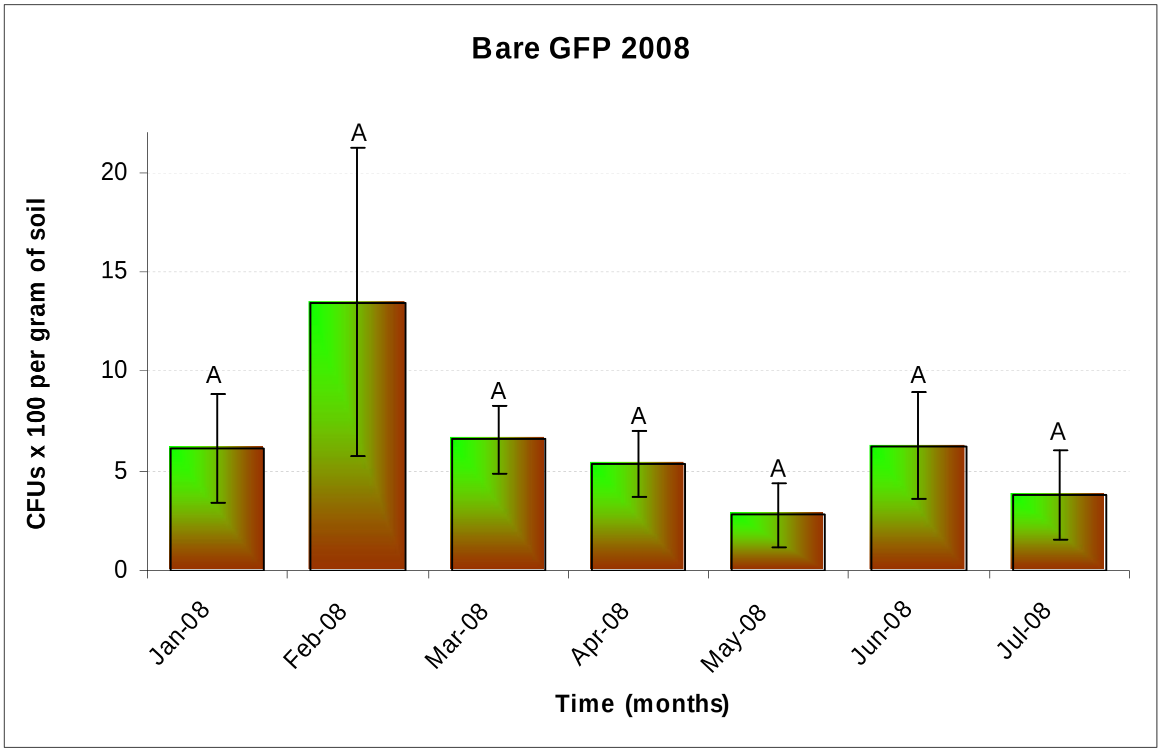


Figure 2.22 **Population Patterns for *Metarhizium*-GFP Isolated from Bare Plots in 2008**. An ANOVA analysis was preformed for *Metarhizium*-GFP recovered from bare plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. There were no significant differences in the population of *Metarhizium*-GFP recovered in 2008.

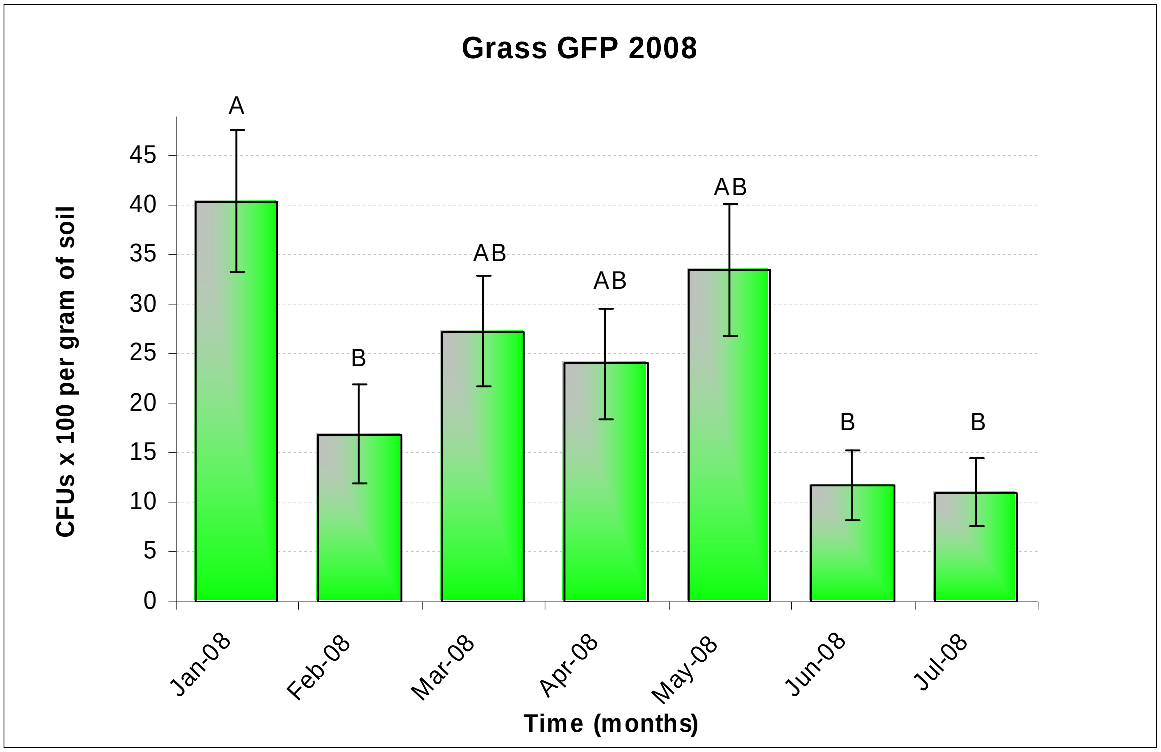


Figure 2.23 **Population Patterns for *Metarhizium*-GFP Isolated from Turf Plots in 2008**. An ANOVA analysis was preformed for *Metarhizium*-GFP recovered from turf plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. There were significantly fewer CFUs recovered for February 2008, June 2008 and July 2008.

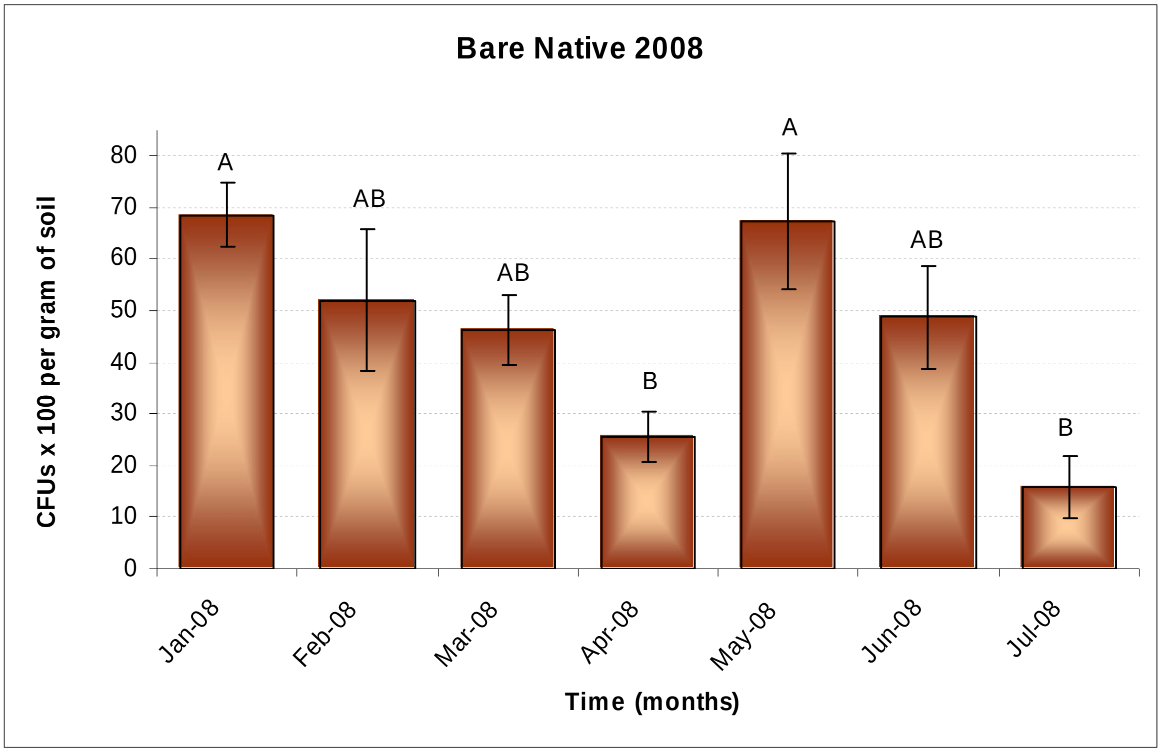


Figure 2.24 **Population Patterns for Native *Metarhizium* Isolated from Bare Plots in 2006**. An ANOVA analysis was preformed for native *Metarhizium* recovered from bare plots for the year. Months with different letters indicate significant (p<0.05) differences in CFUs recovered between those months. There were significantly fewer CFUs recovered for April 2008 and July 2008.

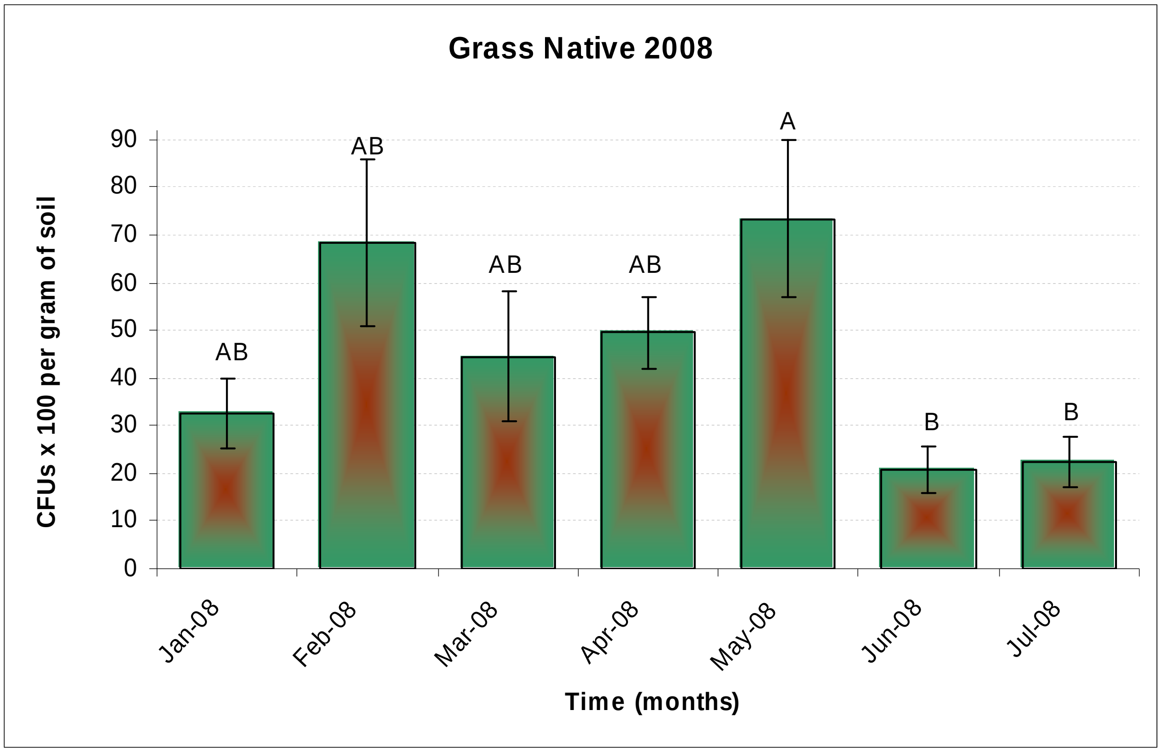


Figure 2.25 **Population Patterns for Native *Metarhizium* Isolated from Turf Plots in 2006**. An ANOVA analysis was preformed for native *Metarhizium* recovered from turf plots for the year. Months with different letters indicate significant ($p<0.05$) differences in CFUs recovered between those months. There were significantly fewer CFUs recovered for June 2008 and July 2008.

	Bare GFP Mean	**Grass GFP Mean**	**Bare Native Mean**	**Grass Native Mean**
Yearly mean by treatment (f,df,p)	6.34 (0.95, 6, 0.4607)	23.59 (4.12, 6, 0.0010)	46.39 (4.50, 6, 0.0005)	44.50 (3.31, 6, 0.0052)
1/08	6.13 ± 2.68 A	40.47 ± 7.24 A	68.60 ± 6.12 A	32.53 ± 7.16 AB
2/08	13.47 ± 7.71 A	16.93 ± 5.05 B	52.07 ± 13.85 AB	68.53 ± 17.53 AB
3/08	6.60 ± 1.70 A	27.33 ± 5.67 AB	46.27 ± 6.64 AB	44.53 ± 13.71 AB
4/08	5.33 ± 1.64 A	24.07 ± 5.56 AB	25.73 ± 4.90 B	49.53 ± 7.55 AB
5/08	2.80 ± 1.62 A	33.47 ± 6.64 AB	67.40 ± 13.15 A	73.40 ± 16.53 A
6/08	6.27 ± 2.66 A	11.80 ± 3.51 B	48.80 ± 9.91 AB	20.73 ± 4.94 B
7/08	3.80 ± 2.20 A	11.07 ± 3.38 B	15.87 ± 5.95 B	22.27 ± 5.33 B

Table 2.6 **Tukey's Studentized Range (HSD) Test for Means for Each Treatment for 2008.** Population dynamics were examined for each treatment. Each column lists the monthly means for each treatment. Significant differences ($p<0.05$)in CFUs for each month based on treatment are designated with a different letter.

Measuring the winter survival of M. anisopliae sf *anisopliae* 2575, *M. anisopliae* sf *anisopliae* 2575 Δ*Mcl*1, and *M. anisopliae* sf *anisopliae* 2575Δ*Mad2 on turf plots*

CFUs of *gfp*-labeled *M. anisopliae* were compared to the winter survival of the two knockout mutants Δ*Mcl1* and Δ*Mad2* (Figure 2.26)(Table 2.7). Δ*Mcl1* was chosen for this field trial because the mutation has no apparent pleiotropic effects. The gene is only expressed in response to different hemolymphs and apparently functions exclusively in immune evasion. The mutant performs as well as the wild type on plant roots so it could be inferred that any reduction in persistence in the field was due to failure to cycle in insect populations. To date, not a single insect infected with Δ*Mcl* has been isolated, showing that the strategy is working (Sibao Wang, Unpubl. data). CFU counts for Δ*Mcl* followed similar trends as the native strain (Figures 2.26, 2.27, 2.28)(Tables 2.7, 2.8, 2.9).

Δ*Mad2* was applied to fields to test the hypothesis that populations are maintained on roots. Counts of CFUs for Δ*Mad2*, which lacks a critical protein required for adhesion to a plant's epidermis, did not follow the same trends as Δ*Mcl1* or the *gfp*-labeled lab strain. CFUs of Δ*Mad2* declined each month after the initial application (Figure 2.29)(Table 2.10). There were no months that showed a significant increase in CFUs for the Δ*Mad2* plots. This indicates that the population trends shown by *M. anisopliae* are dependant on conidia or conidial germlings being attached to roots.

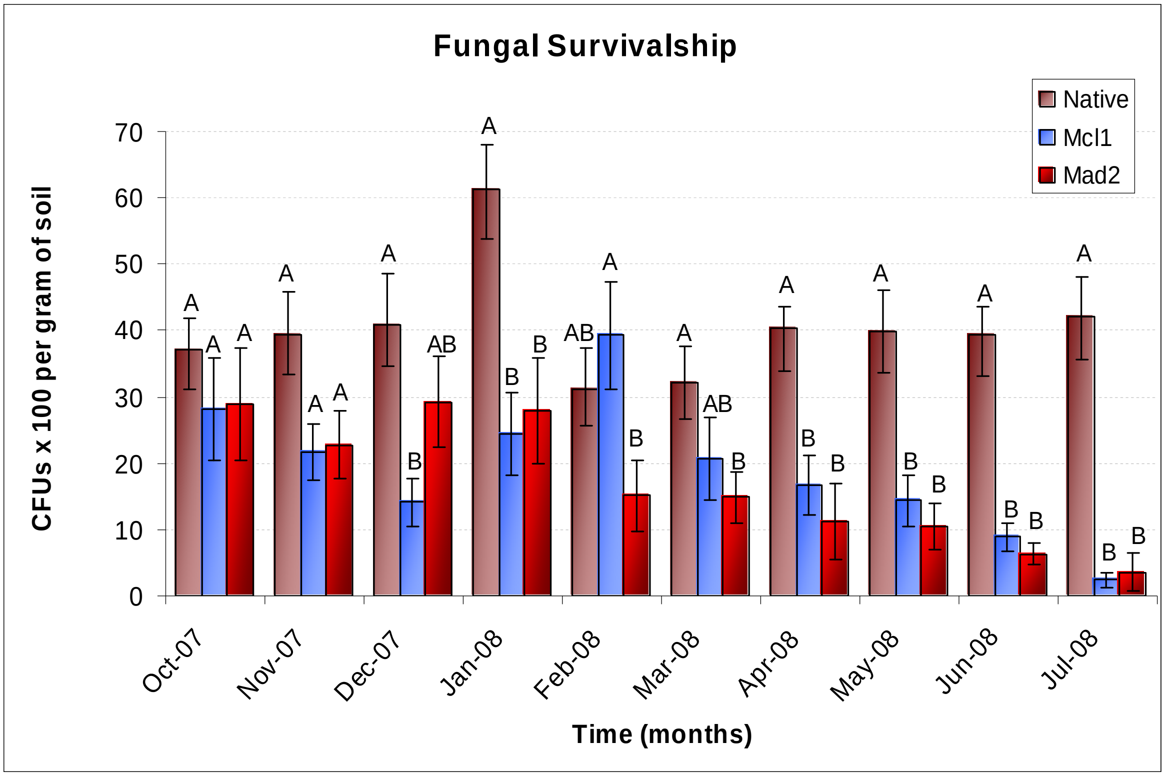


Figure 2.26 **CFUs recovered for *Metarhizium* Δ*Mcl1*, *Metarhizium* Δ*Mad2*, and Native *Metarhizium* strains.** Populations for the three treatments were compared to each other for significant differences in CFU counts each month. Populations of *Metarhizium* Δ*Mcl1* decreased in the November and December 2007, but showed recovery in February and March 2008. Populations of *Metarhizium* Δ*Mad2* continued to decline since the initial application and never recover. When CFUs for Δ*Mcl1* from November 2007 were compared to *Metarhizium* Δ*Mcl1* CFUs for February 2008, there was a significant increase in population. No such cycling was observed for population of *Metarhizium* Δ*Mad2*. After April 2008 populations of *Metarhizium* Δ*Mcl1I* and *Metarhizium* Δ*Mad2* had significantly lower CFUs than the native strains. This significant ($p<0.05$) difference was observed until the end of the experiment.

Treatment	**Native *Metarhizium***	***Metarhizium* Δ*Mcl1***	***Metarhizium* Δ*Mad2***	**Group Mean**	**F, df, P**
Month					
Oct 2007	37.20 ± 4.66 A	28.067 ± 7.73 A	28.86 ± 8.46 A	31.43	02.68, 2, 0.0808
Nov 2007	39.47 ± 6.41 A	21.60 ± 4.28 A	22.69 ± 5.13 A	28.16	3.56, 2, 0.0376
Dec 2007	40.73 ± 7.81 A	14.14 ± .62 B	29.20 ± 6.82 AB	28.34	4.22, 2, 0.0215
Jan 2008	61.40 ± 6.63 A	24.33 ± 6.21 B	27.93 ± 7.94 B	37.8	8.61, 2, 0.0007
Feb 2008	31.13 ± 6.16 AB	39.27 ± 8.11 A	15.13 ± 5.32 B	28.51	3.43, 2, 0.0419
Mar 2008	32.13 ± 5.50 A	20.71 ± 6.16 AB	14.87 ± 3.80 B	22.61	2.91, 2, 0.0660
Apr 2008	40.27 ± 3.27 A	16.67 ± 4.49 B	11.27 ± 5.67 B	22.73	11.32, 2, 0.0001
May 2008	39.93 ± 6.07 A	14.33 ± 3.85 B	10.40 ± 3.45 B	21.56	12.15, 2, <.0001
June 2008	39.47 ± 4.22 A	8.86 ± 2.17 B	6.33 ± 1.58 B	18.43	40.58, 2, <.0001
July 2008	42.07 ± 5.95 A	2.40 ± 1.19 B	3.60 ± 2.84 B	16.02	34.03, 2, <.0001

Table 2.7 **Tukey's Studentized Range (HSD) Test for Comparing Monthly Population Dynamics Between Three Groups**. An ANOVA analysis comparing the three treatments to each other for each month. By April 2008, population of Δ*Mcl1* and Δ*Mad2* were significantly ($p<0.05$) lower than that of the native strains.

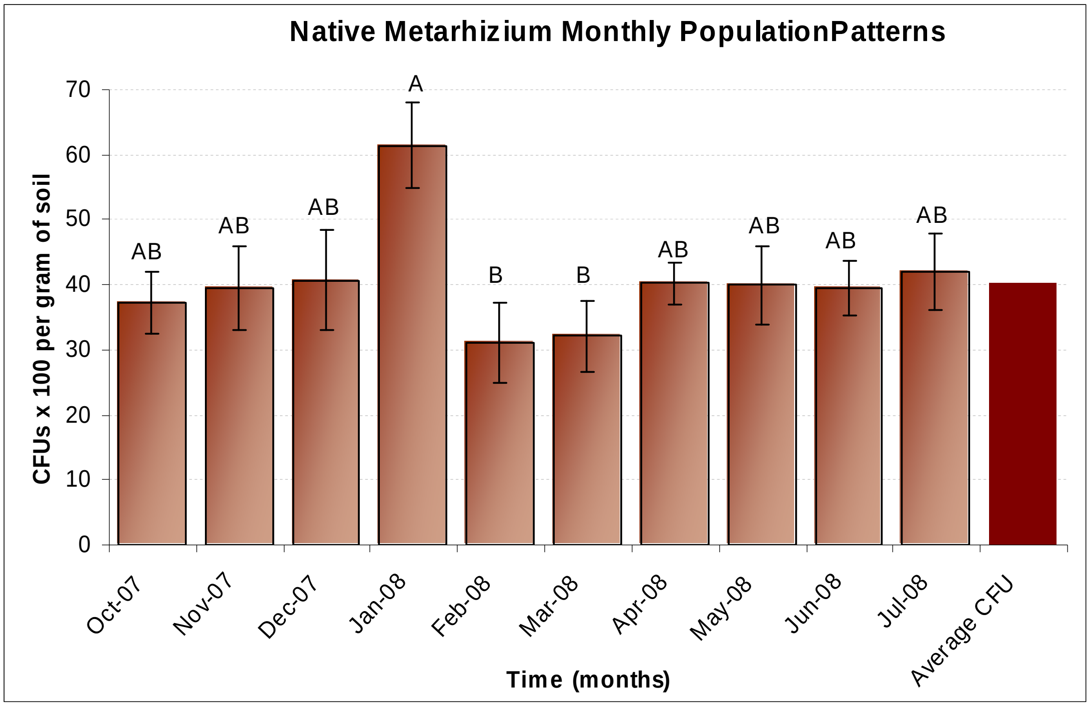


Figure 2.27. **Population Patterns for Native *Metarhizium* Isolated from Turf Plots.** Populations of the native strains showed no significant decrease in population counts in February 2008 and March 2008 (B). There was a significant ($p<0.05$) increase in CFUs for January 2008 (A). The solid bar to the right is the average group mean for the native *Metarhizium* population for the duration of the experiment that each monthly average was statistically compared to.

Treatment	**Native**
Month	
Group Mean for Native	40.38
Oct 2007	37.20 ± 4.66 AB
Nov 2007	39.47 ± 6.41 AB
Dec 2007	40.73 ± 7.81 AB
Jan 2008	61.40 ± 6.63 A
Feb 2008	31.13 ± 6.16 B
Mar 2008	32.13 ± 5.50 B
Apr 2008	40.27 ± 3.27 AB
May 2008	39.93 ± 6.07 AB
June 2008	39.47 ± 4.22 AB
July 2008	42.07 ± 5.95 AB

Table 2.8 **Tukey's Studentized Range (HSD) Test for Population Dynamics of Native *Metarhizium***. Comparison for how each population acted each month. F = 2.01, df= 9,and p = 0.0419. Significant differences ($p<0.05$)in CFUs for each month based on treatment are designated with a different letter.

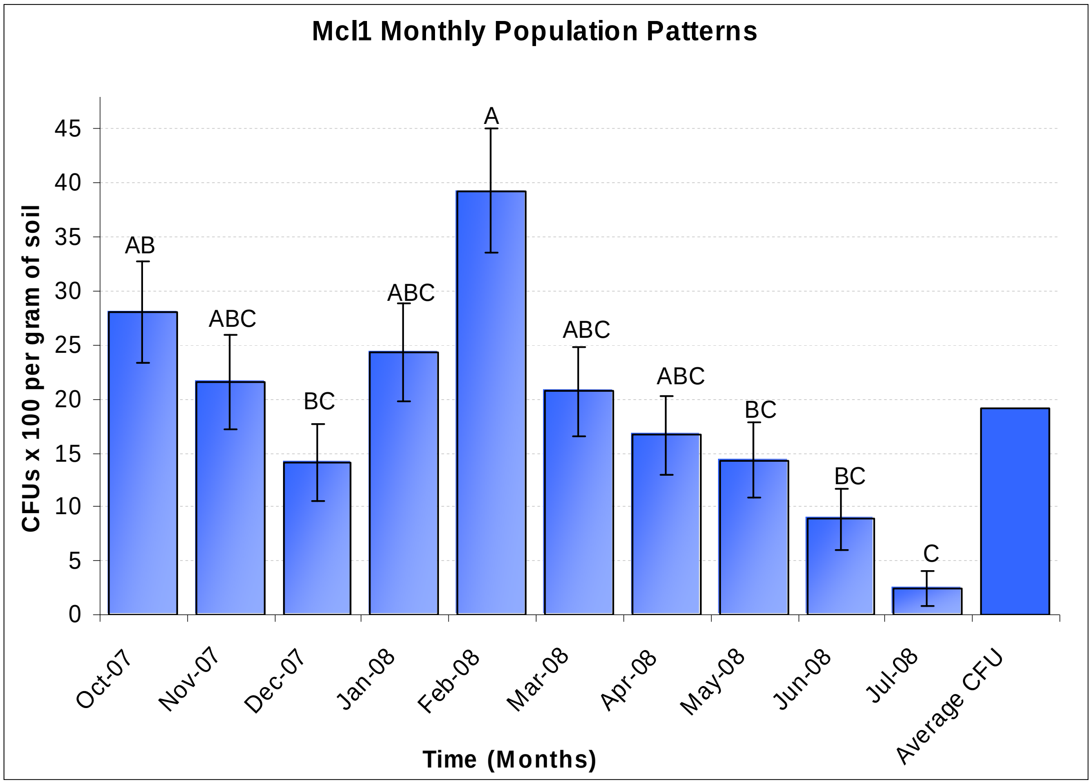


Figure 2.28 **Population Patterns for *Metarhizium* Δ*Mcl1* Isolated from Turf Plots.** Populations of Metarhizium Δ*Mcl1* showed a significant ($p<0.05$) decrease in December 2007 (BC) when compared to October 2008 (AB), populations recovered by February 2008 (A). After February 2008, CFUs began to decrease and were their lowest in July 2008 (C). The solid bar to the right is the average group mean for the *Metarhizium* Δ*Mcl1* population for the duration of the experiment that each monthly average was statistically compared to.

Treatment	**Mcl1**
Month	
Group Mean for Mcl1	19.13
Oct 2007	28.067 ± 7.73 AB
Nov 2007	21.60 ± 4.28 ABC
Dec 2007	14.14 ± .62 BC
Jan 2008	24.33 ± 6.21 ABC
Feb 2008	39.27 ± 8.11 A
Mar 2008	20.71 ± 6.16 ABC
Apr 2008	16.67 ± 4.49 ABC
May 2008	14.33 ± 3.85 BC
June 2008	8.86 ± 2.17 BC
July 2008	2.40 ± 1.19 C

Table 2.9 **Tukey's Studentized Range (HSD) Test for Population Dynamics of *Metarhizium* Δ*Mcl1***. Comparison for how each population behaved each month. F = 3.87, df= 9,and p = 0.0002. Significant differences ($p<0.05$)in CFUs for each month based on treatment are designated with a different letter.

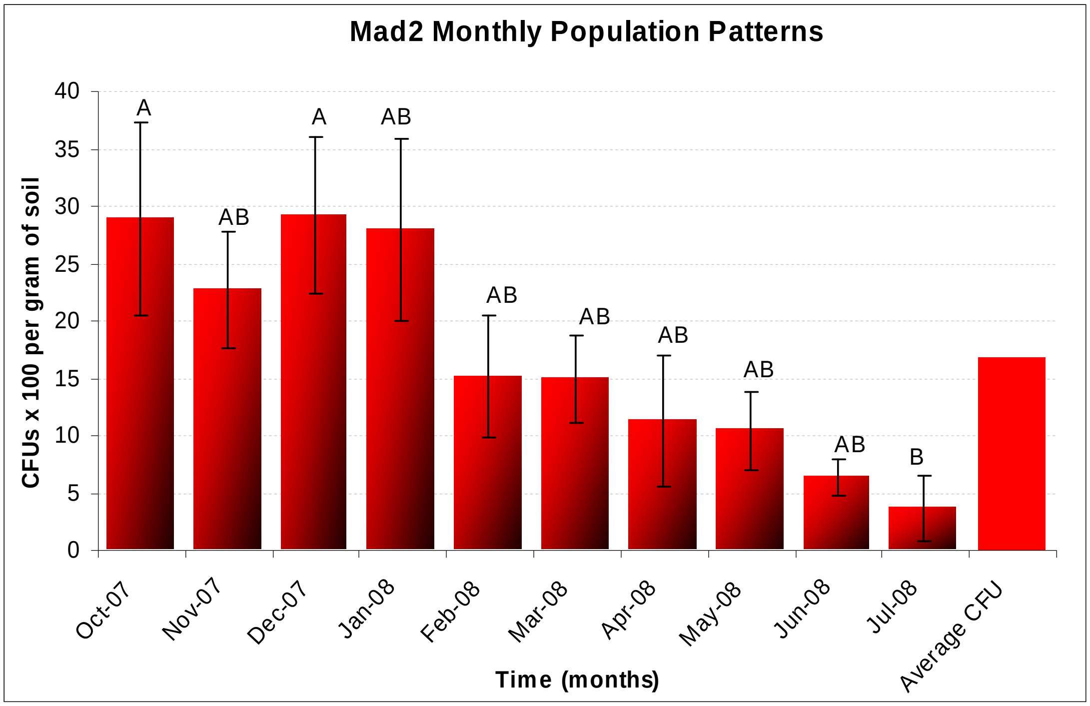


Figure 2.29 **Population Patterns for *Metarhizium* Δ*Mad2* Isolated from Turf Plots.** Populations of *Metarhizium* Δ*Mad2* did not cycle and showed a significant (p<0.05) decrease in CFUs by the end of the month experiment. By July 2008 populations of were significantly lower (B) than October 2007 (A) when plots initially inoculated. The solid bar to the right is the average group mean for the *Metarhizium* Δ*Mad2* population for the duration of the experiment that each monthly average was statistically compared to.

Treatment	**Mad2**
Month	
Group Mean for Mad2	16.87
Oct 2007	28.86 ± 8.46 A
Nov 2007	22.69 ± 5.13 AB
Dec 2007	29.20 ± 6.82 A
Jan 2008	27.93 ± 7.94 AB
Feb 2008	15.13 ± 5.32 AB
Mar 2008	14.87 ± 3.80 AB
Apr 2008	11.27 ± 5.67 AB
May 2008	10.40 ± 3.45 AB
June 2008	6.33 ± 1.58 AB
July 2008	3.60 ± 2.84 B

Table 2.10 **Tukey's Studentized Range (HSD) Test for Population Dynamics of *Metarhizium* Δ*Mad2***. Comparison for how each population acted each month. F = 3.04, df= 9, and p = 0.0024. Significant differences ($p<0.05$)in CFUs for each month based on treatment are designated with a different letter.

Discussion

Two releases of transgenic fungi were conducted with the intent of examining the relationships of plant physiology on fungal population levels. The four year study was performed to establish survivability and population structure over several seasons and multiple generations of insects. The second field trial, which lasted for 10 months, was intended to demonstrate whether survivability and/or maintenance of *M. anisopliae* depended on its saprophytic competence or its ability to cycle through insect populations.

The four year study confirmed that GFP-2575 can survive for years in soils, but with fluctuating population levels. Thus transgenics are potentially competitive with native strains and could potentially provide long-term efficacy against pest insects. While there was an overall decline in CFU counts in soil following the initial application in August 2004, intermittent increases in the CFU counts occurred each year. These population cycles coincided with temperature extremes as temperatures below 0º C and especially temperatures above 32ºC, caused precipitous drops in population levels. The effects of extreme temperature on population counts are consistent with laboratory determinations of permissive temperatures for sporulation, germination, growth and infection processes. At cold temperatures <0º C, *M. anisopliae*, a mesophylic fungus, is not as physiologically as active as it is at temperatures ranging from 25-30º C where *Metarhizium* has optimal growth in lab conditions (Croos and Bodochka, 2001). However, other effects such as reduced productivity of plants and insects at extreme temperatures would also be expected to detrimentally impact population structures of *M. anisopliae*. This may be due to the host plant's response to temperatures. The turf, tall fescue, goes semi-dormant in temperatures >32º C (Burns and Chamblee, 2000). Many grasses in temperate zones reduce root formation and many older roots die (summer root decline) in the summer and decrease photosynthetic rates (Huan and Liu, 2003). It would be predicted that decreased photosynthetic rates would results in fewer carbon sources for *M. anisopliae* and herbivorous insects.

The seasonal changes in population counts of the introduced strain were also shown by the native strains, indicating that they were controlled by the same biotic and abiotic factors. The fact that both the indigenous and introduced populations of *Metarhizium* were in gradual decline suggests that conditions were generally unfavorable to the fungus over the period of this study. This is also suggested by the low total counts which seldom exceeded 10^4/CFUs gm of soil. *M. anisopliae* can reach 10^6/CFUs gm of soil in pastures (Milner, 1992). However, Rath (1995) conducting a field trial in Tasmanian pasture land found populations of *M. anisopliae* plateaud at10^4/CFUs gm of soil.

Population counts of the two knockout strains Δ*Mcl1* and Δ*Mad2* showed different trends. The Δ*Mcl1* mutant is unable to infect insects. Its survival will, therefore, depend on saprophytic associations that presumably include being rhizosphere competent. It is of considerable interest therefore that CFU counts of Δ*Mcl1* cycled in a similar manner to the original *gfp*-labeled strain, although the CFU counts of Δ*Mcl1* were 3-fold lower within 2 months of application. Evidently *Metarhizium* populations are not dependent on the presence of insect hosts for cycling. The cycling of CFUs may be in response to host plant's physiology.

In spite of being infectious to insects, the∆*Mad2* mutant did not show the intermittent increases in population size demonstrated by ∆*Mcl1* or the *gfp* labeled *M. anisopliae.* Populations continually declined indicating that the ability to adhere to root surfaces plays an important part in maintaining population size, irrespective of the presence of insects. The ∆*Mad2* mutant does not show different sensitivity to temperature extremes than the wild type, at least in laboratory conditions, suggesting that the fluctuations in population sizes shown by rhizosphere competent strains of *M. anisopliae* depends on a temperature dependent interaction with plants rather than on differential survival of dormant spores.

The survival strategy of *M. anisopliae* in turf may be more dependant on being able to associate with roots than on *Metarhizium* being an insect pathogen. If an appropriate insect host should encounter the fungus, infection and sporulation on the cadaver can lead to a localized increase in CFUs. These results suggest therefore that populations of *Metarhizium* may be sustained at the rhizosphere, but increases in population size above the carrying capacity of the rhizosphere can arise following sporulation on the cadavers of insect hosts. *Metarhizium* is therefore in the fortuitous position of being able to maintain populations on plant roots while at the same time escaping competition from other fungi and build up population levels by functioning as an insect pathogen.

Chapter 3: Using microarray based mutation accumulation assays to inventory strains recovered from the field site for changes in gene content and altered patterns of gene regulation.

Introduction

Expressed sequence tags (ESTs) are expressed portions of genes obtained by sequencing cDNAs. They have been used for microarray analysis of gene expression to examine plant-fungus-insect interactions in microcosms (Wang *et al.,* 2004). These experiments were the first comprehensive effort to characterize gene expression during *Metarhizium's* facultative saprophyte life cycle to a level comparable to that sought for its virulence genes. Microarrays are currently one of the most efficient, comprehensive, and integrative means of assigning ecological and functional significance to genes of unknown function (Kim *et al.,* 2001). The idea behind using microarrays for examining field isolates is that while integrative transformants are very stable when grown for long periods in the absence of selection pressures in pure culture under lab conditions, stability may be different in a complex field environment (St. Leger *et al*., 1995a, b).

Previous microarray studies on M. anisopliae in the St. Leger laboratory

Microarrays have been used to examine physiological parameters such as the regulation of secreted products such as enzymes and toxins, as well as protein phosphorylation events, transcriptional regulatory factors and physiological cues that will define at the molecular level the life of strain 2575 as a pathogen and as a saprophyte. In particular, microarray surveys have been used to examine differential gene expression of insect-response genes to 1) identify genes that coordinate the production of enzymes/toxins; 2) identify genes involved in determining host range and 3) investigate physiological responses to growth in different insects (Freimoser *et al.*, 2003b; Freimoser *et al.*, 2005; Wang *et al*., 2005a).

The *M. anisopliae* sf *anisopliae* strain 2575 dataset contains about 11,000 partial sequencing of randomly selected cDNA clones and has defined 3,563 EST unigenes (transcript sequences that seem to come from the same gene). Approximately 30% of 2575's total genes are single copy genes (Freimoser *et al.*, 2003b; Freimoser *et al.*, 2005). About 60% of the ESTs expressed by 2575 during growth on insect cuticle encoded secreted enzymes and toxins. It has been speculated that the large number and diversity of these effectors may be the key to explaining the ability of strain 2575 to infect a wide variety of insects (Freimoser *et al*., 2003a).

Four microarray studies have been published showing how sets of functionally related genes are coordinately induced or repressed by *M. anisopliae* in response to host related stimuli (Wang *et al.*, 2005b; Wang and St. Leger, 2006). More than 700 up-regulated genes have been identified in the 2575 strain during adaptation to host cuticle or to hemolymph. Some genes have provided insight into the mechanisms which *M. anisopliae* has adapted to survive in these environments. Various aspects of hyphal growth in cuticle and hemolymph are associated with up-regulation of different genes encoding components of signal transduction. Genes involved in membrane biogenesis, synthesis of cell wall components, storage or mobilization of nutrient reserves and protein folding are also highly expressed, indicative of the manufacturing of cell structures. Other features highlighted by this work include the production of antimicrobial molecules and the very early cuticle-induced production of a variety of transporters and permeases that allow the fungus to analyze the cuticle and then respond with secretion of a plethora of proteins. Mechanisms involved in adaptation to hemolymph include a dramatic remodeling of cell walls and lipid composition, the accumulation of solutes that increase internal osmotic pressure and up-regulation of non-oxidative respiratory pathways. A diverse range of genes that encode virulence factors that help defend against possible host defenses such as oxidative and nitrosative (such as the production of nitric oxide) stress and phenolics are up-regulated on cuticle and/or hemolymph along with an abundance of genes for extracellular enzymes and toxins that contribute to host damage (Wang and St. Leger, 2006).

The array of ESTs from strain 2575 may be used for heterologous hybridization with DNA or RNA from diverse strains of *M. anisopliae* (Wang *et al*., 2005a). This enables the role of regulatory variation to be examined with respect to adaptive evolution of a pathogen. Changes in gene expression may be used by a species to evolve and express new phenotypes (Ben-Shahar *et al*., 2002). Yeast currently provides the model for examining processes in fungi for the heritability of transcription (Brem *et al.,* 2002; Townsend *et al*., 2003), for changes in gene expression levels in response to selection (Ferea *et al*., 1999), and for regulatory variation in natural isolates. However, this variation has not been related to adaptation to different environments.

Patterns of gene duplication, divergence, and deletion in several generalist and specialist strains of *M. anisopliae* were specifically determined by heterologous hybridization of total genomic DNA. DNA from each strain was competitively hybridized to an array of strain 2575 genes (Wang *et al*., 2005a,b). Most genes required for major life processes had differences in genomic hybridization which averaged less than 5%. One group of genes in the 2575 strain which seems to lack counterparts in the other strains is mainly composed of mobile genetic elements (transposons). Exceptionally, there was an expansion in the number of insertion elements in the specialist strain 443 suggesting that evolution could occur in leaps (Freimoser *et al*., 2003b; Freimoser *et al.,* 2005; Wang *et al*., 2005a). This has implications for strain stability, including the possibility of alterations in virulence and host range, which could impact commercial development.

To date, the focus of EST approaches has been on probing the associations between entomopathogenic fungi and their hosts. The genes required for saprophytic survival have not been as extensively studied (Freimoser *et al*., 2003a, b; Freimoser *et al*., 2005; Wang *et al*., 2005a,b). However, the EST data set also includes root exudate induced transcripts (Wang *et al*., 2005a), and within the arrayed EST collections are 592 genes expressed when strain 2575 lives saprophytically (Wang *et al*., 2005a). This research initiated attempts to identify genes implicated in soil survival so as to establish the role of *Metarhizium* in the ecology of grassland soils, which may be the preferred habitat of many strains (Prior, 1992; Hu and St. Leger, 2002).

Strategy behind the current research

Rapid evolution of new phenotypes is usually caused by changes in gene expression (DeRisi, 1997; Ben-Shahar *et al.,* 2002; Enard *et al*., 2002). In this study array based mutation accumulation assays were used to inventory strains recovered from the field site for changes in gene content and altered patterns of gene regulation and expression that can be used to predict any physiological changes (Wang and St. Leger, 2005; Wang *et al.,* 2005b). This will reveal the capacity of *M. anisopliae* to undergo rapid evolution in gene content and expression. Measuring changes in gene expression with microarrays provides a very sensitive assay for mutant accumulation as mutations in many different loci can affect abundance of any one transcript and a single mutation in a regulatory locus can affect dozens of genes (Gibson, 2005).

Thus, recovering a strain of *M. anisopliae* that was applied to the field 3.5 years earlier, allows for characterizing permanent changes in gene expression required for adaptation to the soil-dwelling, saprophytic phase. If similar changes in gene content or gene regulatory profiles were observed in multiple isolates from replicate plots, this would show selection for certain genes to be expressed in the grassland lifestyles. Such mutations in expression of genes necessary for survival may allow for increased fitness and be selected for over time, a process known as microevolution.

Materials and methods

M. anisopliae isolates recovered from the test site were screened for the following parameters: growth rate, colony morphology, level of conidia and number of colony forming units (CFUs) (St. Leger *et. al.*, 1996a,c). Aside from the marker genes (*bar* and *gfp*), a selection from over 1,700 ESTs (Appendix A) were utilized as probes for examining selective loss of genes and chromosomal rearrangements. ESTs encoding viral genes implicated in retrotransposons were used as probes to detect jumping events.

Isolation of RNA samples for microarray analysis

Gene expression levels of each field isolate (recovered 3.5 years after initial application on turf) were directly compared with the input strain by co-hybridizing cDNAs obtained eight hours after transfer from Sabouraud's Dextrose Broth (SDB) to *Manduca sexta's* insect cuticle, as this triggers substantial transcriptional activity (Freimoser *et al*., 2005), with broad inter-strain variation in gene expression (Wang and St. Leger, 2005). Five random field isolates from the rose bengal selective media plates (Chapter 2) were selected. Single spore isolations were transferred to fresh selective media containing PDA, 10μg/ml of dodine, 250 μg/ml cyclohexamide, 80 oxytetracycline dihydrate μg/ml, 80 μg/ml streptomycin sulfate, and 80 μg/ml penicillin-G potassium.

This process was repeated twice to eliminate transitional physical variations due to physiological adaptation to field conditions. Spores harvested from these plates were grown in SDB medium at 27º Celsius at 250 RPM. After 36 hours the mycelium was harvested by filtration and resuspended in a total volume of 25ml with sterilized tap water in a sterile 50ml conical tube. This sample was then poured into a 125ml flask containing 25ml of 2% *Manduca sexta* cuticle, to create a total volume of 50ml of 1% cuticle plus mycelium suspension. This suspension was incubated for 8 hours at 27º Celsius and the cuticle/mycelium suspension was comminuted under liquid nitrogen and freeze dried. The RNA was extracted using the Qiagen RNeasy Plant Minikit™ model 74904. One hundred mg of freeze-dried mycelium was mixed with 600 µl of RLC buffer, and ß-mercaptoethanol was added in the ratio of 10 microliters per 1µl of RLC buffer. The sample was vortexed rigorously to ensure a homogenous suspension. The suspension was incubated for 3 minutes at 56º C, transferred to the QIAshredder spin column and centrifuged for 2 minutes at maximum speed. The total volume of supernatant was measured and half the volume equivalent of 100% ethanol was added to the sample. The sample was then transferred to an RNeasy mini column and centrifuged for 15 seconds at 10,000 RPMs. The supernatant was discarded and eighty microliters of DNase was placed on top of the RNeasy mini column filter to digest any DNA that may be present; this was allowed to incubate for 15 minutes. Then 700 µl of Buffer RW1 was added to the RNeasy column. The sample was centrifuged for 15 seconds. The RNeasy column was then transferred into a new 2ml collection tube and 500 µl of RPE buffer was added. The sample was centrifuged for 15 seconds. An additional 500 µl of RPE buffer was added to the RNeasy column and the sample was centrifuged for an additional 2 minutes. The sample was then re-centrifuged to ensure all ethanol had been removed. To remove RNA from the filter, 50 µl of RNase-free water was added to the column. The column was placed in a new collection tube and centrifuged for two minutes. The supernatant was removed and placed in the column again to collect any residual RNA. The concentration of RNA was measured at 260 nm. A minimum of 2,500 µg/ml was required to obtain satisfactory results for a microarray analysis.

The RNA samples were processed by the University of Maryland Biotechnology Institutes in Shady Grove, MD. The Ovation Aminoallyl RNA Amplification and Labeling system version 1.1 protocol was used to amplify the RNA and label the RNA samples. Corning GapII (gamma amino propyl) microarray slides were used for hybridization. The Axon Scanner 4200 (Molecular Devices) was used to capture images of the microarrays after hybridization. All slides contained 3 technical replicates for each EST tested, along with blanks, and negative controls of water and SSC buffer. Three biological replicates were also performed for each field isolate and the original control strain. A loop design (Figure 3.1) was used to compare differential gene expression between the lab strain (labeled: Control) and 5 field isolates randomly selected after 3.5 years in turf (labeled: Field 1, Field 2, Field 3, Field 4, and Field 5).

The TM4 microarray suite (http://www.tm4.org) was used to process the raw cDNA files which were in the .tiff image format. All microarrays underwent Lowess normalization and flip-dye analysis for the Cy3 (green) and Cy5 (red) dyes. Flip-dye is required as Cy5 breaks down quickly. Flip-dye also ensures neither dye is preferentially binding to the samples.

In-slide-replicate analysis was implemented on all normalized arrays, and the MeV (MultiExperiment Viewer utility in TM4) was used to perform t-tests.

Resultant volcano plots were obtained, as well as significance values of differential gene expression, using a p-value of 0.05 as the cut-off. All genes with significant differential gene expression were categorized by fold change. Fold changes are the measure of the ratio of fluorescent signal intensities, and provide insight into differential gene expression between samples (Cui and Churchill, 2003). Genes with a fold change of > 1.5 were analyzed and sorted by function. Genes with significant (p<0.05) differential gene expression of 2> were also sorted by function, in addition, RT-PCR (reverse transcriptase-polymerase chain reaction) was performed for these EST to validate the microarray findings (Freimoser *et al.*, 2005). RT PCR data were analyzed by using the densitometer program Un-Scan-It. Intensities for the two constitutive bands, *Tef* and *Gdp* were averaged. The average calculated average value was then used to determine a ratio for expression for the *Tef* and *Gpd* gene intensity. The range between the *Tef* and *Gdp* intensities was set as the base-line. The intensities for the genes examined by RT PCR were divided by the average *Tef-Gdp* intensity value. If the calculated ratio values higher than the *Tef* or *Gdp* ratio values were up-regulated, values in the same range as the *Tef-Gdp* ratios showed no change, and values less than the *Tef-Gdp* ratio were down-regulated. Intensity ratios for the original input samples (2575) were compared to the other 10 recovered isolated (Re1-Re10) to determine the changes in gene expression between samples.

MIAME (http://www.ebi.ac.uk/microarray-as/ae/) is the minimum information about microarray database. All protocol with regards to array design, experimental design and sample preparation, as well as results, must be submitted to MIAME before publication of any microarray data is permitted in most journals.

THE LOOP DESIGN

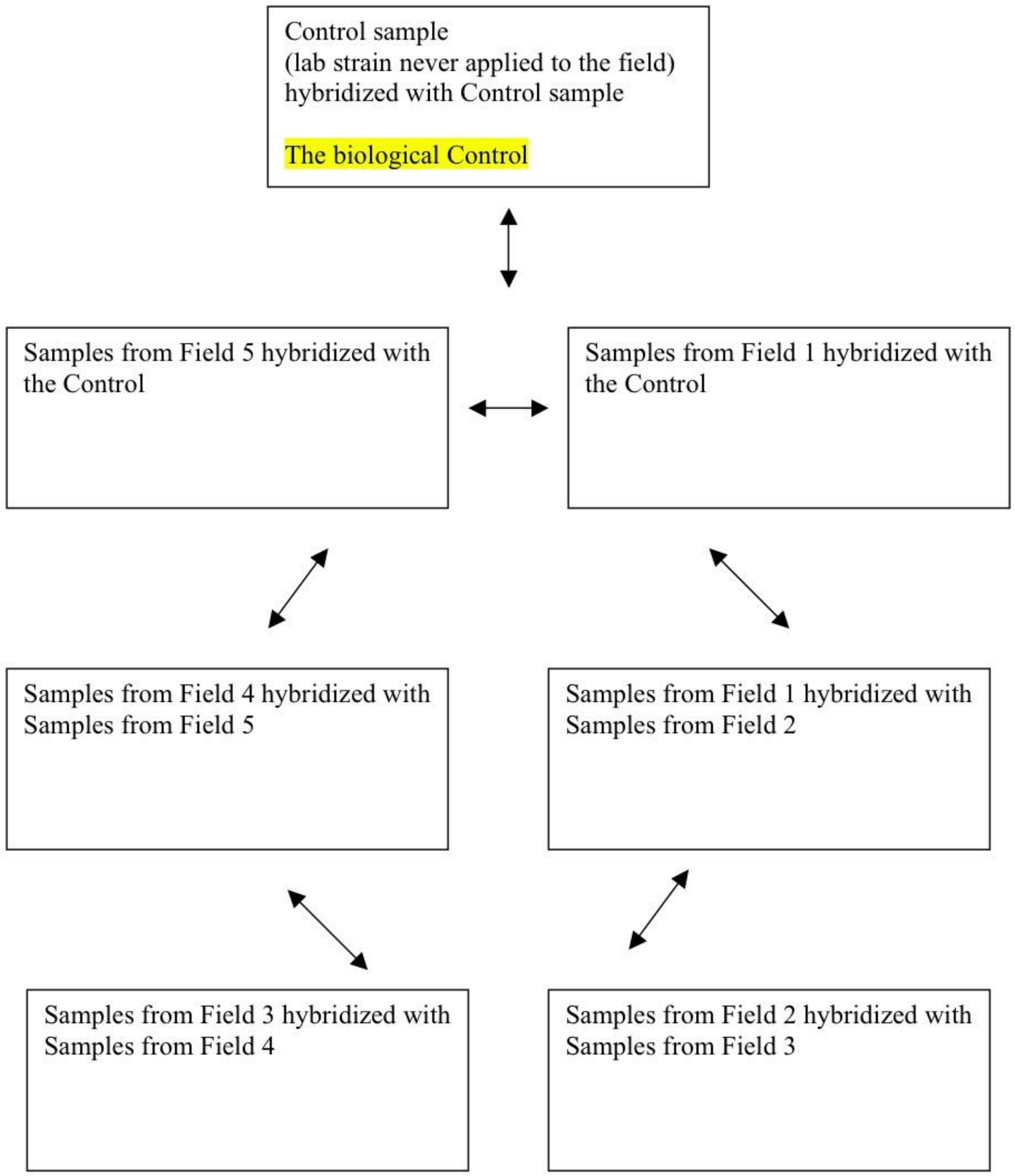


Figure 3.1 **The Loop Design.** A schematic illustrating how the five recovered field isolates were compared to each other and to the control. Seven combinations of hybridizations were examined. Field 1 and Field 5 isolates were compared to the control. The control samples contained samples of were compared to themselves to establish a base-line for natural variation in gene expression.

Results

Differential gene expression was calculated for the 7 conditions tested. Slides that were probed with two samples of control RNA were used to standardize the data and to confirm there were no technical issues with the design or protocol. As expected, there were no significant differences in gene expression at >1.5 fold change in the control hybridized with control slides. Mutation rates were determined by calculating differential expression between two field isolates and the control, and between the five field isolates that had significant levels of differential gene expression with fold changes greater than two (Freimoser *et al.*, 2005). Because of variation and noise between the 9 replicates for each RNA sample, not all differences in gene expression greater than 2 were significantly different. For the most part, this natural variation between replicates accounted for genes with >2-fold differences in expression between isolates but with $p > 0.05$. These genes were housekeeping genes involved in general metabolism. Genes with fold changes > 2 but fell short of the significance level ($p<0.05$) are not discussed, but are listed in Appendix B.

Comparing the control slides to that of the 5 field isolates identified only 23 genes that had significant ($p<0.05$) differential gene expression with greater than a 2 fold change in regulation (Figure 3.2a-d). There were an additional 40 genes that had significant ($p<0.05$) differential gene expression with fold changes between 1.5 and 2. Primers were created for reverse transcriptase PCR validation of differential expression patterns (Figure 3.14-3.15).

Volcano plots (Figures 3.6 -3.12) were used to visualize gene expression patterns and trends for the five recovered field isolates, as well as the control input strain. As expected, none of the genes showed significantly altered regulation when RNA from the input strain applied to the field was compared with itself (i.e. reference hybridized with the reference) so experimental variation would not contribute false positives. Likewise, no significant differences in gene expression were observed for the slide with the Control (input strain) hybridized to Field 1 isolate, Field 1 isolate hybridized to Field 2 isolate, Field 2 isolate hybridized to Field 3 isolate. The mutation rate when the samples from Field 3 isolates were compared to Field 4 isolates was 3/1749 ESTs, or 0.1715% of the genes showing significantly altered expression after 3.5 years for genes with a fold change over 2. When Field 4 was compared to Field 5, six genes with significantly altered expression were observed (6/1749 or 0.3431 % mutated/3.5 years). The highest rate of mutation was observed for the Control hybridized to Field 5 slides. There were 16 events of differential expression with a fold change of 2 or greater. The calculated rate of change was 14/1749 or 0.972% for 3.5 years. The total proportion of the genome subject to mutations in the 5 recovered field isolates was 23/1749 or 1.31 % for 3.5 years. Using the less stringent 1.5>fold change, the mutation rate was 63/1749 or 3.6% of the genome over 3.5 years.

Genes were categorized by functional group and given group numbers as follows: 1a amino acid metabolism (66 genes assayed), 1b carbohydrate metabolism (156 genes assayed), 1c lipid metabolism (52 genes assayed), 1d nucleotide metabolism (24 genes assayed), 1e nitrogen, phosphorus or sulfur metabolism (19 genes assayed), 1f cofactor and vitamins (17 genes assayed), 2 energy metabolism (64 assayed), 3a ribosomal proteins (45 genes assayed), 3b translation (28 genes assayed), 3c proteolysis (57 genes assayed), 3d protein modification and targeting (42 genes assayed), 4a RNA metabolism (13 genes assayed), 4b RNA modification (26 genes assayed), 4c transcription (58 genes assayed), 5a cell wall (36 genes assayed), 5b cytoskeleton (28 genes assayed), 5c extracellular matrix/cell wall protein (17 genes assayed), 5d transport proteins (64 genes assayed), 5e cellular transportation (39 genes assayed), 5f signaling (47 genes assayed), 6a cell cycle (37 genes assayed), 6b chromosome structure (11 genes assayed), 6c DNA synthesis (8 genes assayed), 6d cell polarity (8 genes assayed), 6e sexual cycle (10l genes assayed), 6f clock control/light response (6 genes assayed), 6g cell death (13 genes assayed), 7a stress response (37 genes assayed), 7b detoxification (27 genes assayed), 7c secondary metabolites (30 genes assayed), 8 transposable elements (13 genes assayed), 9a hypothetical proteins (234 genes assayed) and 9b unknown proteins (415 genes assayed). The differentially expressed genes were expressed as a fraction of the total number of genes within that functional group (Figure 3.3). The majority of genes (Figure 3.4) (Figure 3.5) with significant differential gene expression at >1.5 and >2 fold change coded for cell wall and hypothetical proteins. Thus no genes coding for known pathogenicity determinants such as adhesins, toxins, and immune evasion genes (including 45 genes for proteases) showed altered regulation. Approximately 60% of the arrayed genome encodes for secreted products, and expression of these genes was highly conserved.

In addition to examining changes in expression patterns, the possibility of gross chromosomal structural changes was investigated. Southern blots were performed using transposable elements as probes to examine the genomic DNA of 10 recovered field isolates compared to the original input strain (Figure 3.15). The five field isolates used for microarray analysis were included in the Southern blot analysis. All isolates were chromosomally identical to the control input strain.

Control hybridized with Control								
Fold Change	GenBank#		best match	Func	Function	source	description	Gene std dev
gnificant genes with a 1.5 > fold change								

Control hybridized with Field 1								
Fold Change	GenBank#	ID Number on Volcano Plot	best match	Func	Function	source	description	Gene std dev
-1.99	CN808375	1	EAA68393	9a	Hypothetical Protein	Gibberella zeae PH-1	hypothetical protein FG00663	0.01

Control hybridized with Field 5								
Fold Change	GenBank#	ID Number on Volcano Plot	best match	Func	Function	source	description	Gene std dev
-5.12	AJ272832	1	EAA54957	9a	Hypothetical Protein	Magnaporthe grisea	hypothetical protein MG05748.4	0.80
-4.87	AJ272765	2	AAN74815	7c	Secondary metabolites	Gibberella moniliformis	Fum12p, required for fumonisin biosynthesis	0.91
-3.20	AJ272811	3	CAB96004	5d	Transport proteins	Schizosaccharomyces pombe	mitochondrial carrier protein; yeast yor222w homolog	0.51
-3.19	AJ273870	4	BAB33421	6g	Cell death	Pisum sativum	putative senescence-associated protein	0.54
-2.72	AJ272799	5	AAC35942	5a	Cell wall	Aspergillus fumigatus	GEL1 protein	0.36
-2.63	AJ272761	6	EAA69714	9a	Hypothetical Protein	Gibberella zeae PH-1	hypothetical protein FG02083	0.22
-2.63	AJ272778	7	EAA64687	9a	Hypothetical Protein	Aspergillus nidulans	hypothetical protein AN2582.2	0.30
-2.59	CN809178	8	AAA20128	5a	Cell wall	Magnaporthe grisea	hydrophobin-like protein	0.17
-2.32	AJ272786	9	NP_593423	3c	Proteolysis	Schizosaccharomyces pombe	Highly conserved ring finger; similar to yeast apc11 SPC(ubiquitin-protein ligase activity)	0.11
-2.10	CN809258	10	XP_372959	1d	Nucleotide metabolism	Homo sapiens	similar to rRNA intron-encoded homing endonuclease	0.42
-1.92	AJ272961	11		9b	Unknown Protein		unknown	0.33
-1.88	AJ274064	12	CAE76477	6a	Cell cycle	Neurospora crassa	related to cell cycle regulation and aging protein	0.26
-1.83	AJ273711	13	NP_593899	6d	Cell polarity	Schizosaccharomyces pombe	possibly involved in cell polarity; by similarity to yeast rax2	0.07
-1.81	CN808382	14	O94284	7b	Detoxification	Schizosaccharomyces pombe	Sulfide: quinone oxidoreductase, mitochondrial precursor (Heavy metal tolerance protein 2) (Cadmium resistance protein)	0.33
-1.77	CN808435	15	AAO34674	1b	Carbohydrate metabolism	Gibberella zeae	glucosidase	0.30
-1.72	CN809442	16	EAA16547	9a	Hypothetical Protein	Plasmodium yoelii yoelii	hypothetical protein	0.22
-1.67	AJ272997	17	NP_955904	7a	Stress response	Danio rerio	DnaJ (Hsp40) homolog, subfamily C, member 3	0.18

Figure 3.2a **Significant Genes with a fold change of 1.5 or greater (part 1 of 4).** All significant genes with differential expression of 1.5 fold change or greater are listed. Genes that are down-regulated are in red and up-regulated genes are in blue. All yellow-highlighted cells represent genes with a 2 fold or greater change in gene expression.

Control hybridized with Field 5								
Fold Change	GenBank#	ID Number on Volcano Plot	best match	Func	Function	source	description	Gene std dev
-1.63	AJ273380	18	P78695	3d	Prot modification/targeting	Neurospora crassa	78 kDa glucose-regulated protein homolog precursor (grp 78) (immunoglobulin heavy chain binding protein homolog) (BIP)	0.17
-1.62	AJ272972	19	BAA19073	2	Energy Metabolism	Aspergillus niger	NADP-dependent isocitrate dehydrogenase precursor	0.17
-1.60	CN809572	20	AAP68979	5b	Cytoskeleton	Gibberella zeae PH-1	beta-tubulin	0.06
-1.58	CN809192	21	EAA70750	9a	Hypothetical Protein	Gibberella zeae PH-1	hypothetical protein FG00804	0.11
-1.52	AJ273975	22	CAB88598	1c	Lipid metabilism	Neurospora crassa	lanosterol synthase related protein	0.23
-1.51	AJ272697	23	AAC49319	7c	Secondary metabolites	Aspergillus parasiticus	versicolorin B synthase	0.21
1.52	CN809570	24	NP_822771	1c	Lipid metabilism	Streptomyces avermitilis	putative 3-ketosteroid-delta-1-dehydrogenase	0.24
1.55	CN809045	25	AAM48104	3d	Prot modification/targeting	Emericella nidulans	theta class glutathione S-transferase	0.15
1.57	AJ273303	26	T45525	5a	Cell wall	Kluyveromyces marxianus var. lactis	WSC4 homolog, WSC4p is involved in cell wall integritz and stress response	0.08
1.58	AJ274374	27	S74210	1b	Carbohydrate metabolism	Aspergillus niger	glucokinase (Glk)	0.25
1.59	CN808158	28	AAN27998	1c	Lipid metabolism	Leptosphaeria maculans	sterol delta 5,6-desaturase ERG3	0.14
1.59	AJ273314	29		9b	Unknown Protein		unknown	0.23
1.60	CN809024	30	EAA70431	7a	Stress response	Gibberella zeae PH-1	Heat shock 70 kDa protein (HSP70)	0.12
1.64	CN809078	31	NP_011461	5e	Cellular transportation	Saccharomyces cerevisiae	Protein localized to COPII-coated vesicles, involved in vesicle formation and incorporation of specific secretory cargo; required for the delivery of bud-site selection protein Axl2p to cell surface	0.22
1.65	AJ273266	32	AAB18274	1e	N-, P-, S-metabolism	Emericella nidulans	sconCp (sulphur metabolite repression regulation)	0.19
1.73	AJ273019	33	AAD00154	3c	Proteolysis	Metarhizium anisopliae	ubiquitin conjugating enzyme	0.27
1.93	CN807971	34	CAD21046	2	Energy Metabolism	Neurospora crassa	ubiquinol-cytochrome C reductase complex core protein 2 precursor	0.18
1.94	CN808121	35	NP_013458	1b	Carbohydrate metabolism	Saccharomyces cerevisiae	Transaldolase, enzyme in the pentose phosphate pathway; Tal1p	0.11
2.17	AJ273457	36	CAC28787	3a	Ribosomal proteins	Neurospora crassa	probable ribosomal protein L12	0.36
2.31	CN809336	37	EAA33203	9a	Hypothetical Protein	Neurospora crassa	hypothetical protein	0.25
2.33	AJ273011	38	CAA05375	5a	Cell wall	Trichoderma harzianum	b-1,3 exoglucanase	0.45
2.46	CN808187	39	BAD01559	5a	Cell wall	Aspergillus kawachii	cell wall protein	0.39

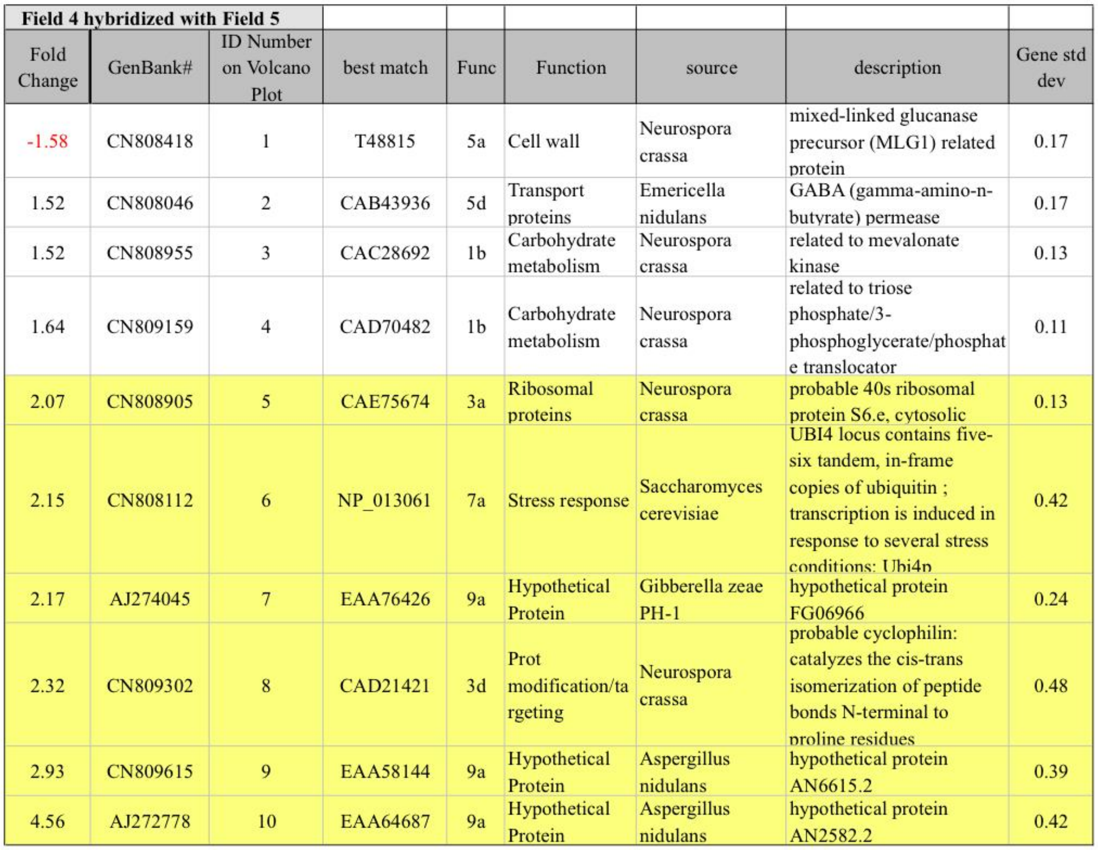
Figure 3.2b **Significant Genes with a fold change of 1.5 or greater (part 2 of 4).**

Field 1 hybridized with Field 2								
Fold Change	GenBank#		best match	Func	Function	source	description	Gene std dev
gnificant genes with a 1.5 > fold change								

Field 2 hybridized with Field 3								
Fold Change	GenBank#		best match	Func	Function	source	description	Gene std dev
gnificant genes with a 1.5 > fold change								

Field 3 hybridized with Field 4								
Fold Change	GenBank#	ID Number on Volcano Plot	best match	Func	Function	source	description	Gene std dev
-6.49	AJ274064	1	CAE76477	6a	Cell cycle	Neurospora crassa	related to cell cycle regulation and aging protein	0.22
-2.80	CN808789	2	AAB69310	9b	Unknown Protein	Metarhizium anisopliae	cystein rich protein	0.29
-1.54	CN808266	3	AAD55983	3c	Proteolysis	Magnaporthe grisea	ubiquitin-conjugating protein	0.23
1.51	AJ273806	4	AAN31395	1a	Aminoacid metabolism	Aspergillus sojae	leucine aminopeptidase	0.13
1.51	CN807972	5	CAD70918	1c	Lipid metabilism	Neurospora crassa	probable fatty acid elongase (FEN1)	0.16
1.52	AJ273384	6	EAA77732	9a	Hypothetical Protein	Gibberella zeae PH-1	hypothetical protein FG09683	0.23
1.55	CN808112	7	NP_013061	7a	Stress response	Saccharomyces cerevisiae	UBI4 locus contains five-six tandem, in-frame copies of ubiquitin ; transcription is induced in response to several stress conditions; Ubi4p	0.25
1.70	AJ274279	8		9b	Unknown Protein		unknown	0.25
1.71	AJ274318	9	T38841	4b	RNA modification	Schizosaccharomyces pombe	probable pre-mRNA splicing factor	0.23
1.75	AJ273517	10	CAB77004	5e	Cellular transportation	Schizosaccharomyces pombe	similar to yeast Synaptobrevin (v-SNARE) homolog; Bos1	0.26
1.77	AJ274289	11		9b	Unknown Protein		unknown	0.26
1.86	AJ272823	12	NP_010430	4c	Transcription	Saccharomyces cerevisiae	transcriptional activator; Swi5p	0.19
2.19	AJ273303	13	T45525	5a	Cell wall	Kluyveromyces marxianus var. lactis	WSC4 homolog, WSC4p is involved in cell wall integritz and stress response	0.36

Figure 3.2c **Significant Genes with a fold change of 1.5 or greater (part 3 of 4).**



Field 4 hybridized with Field 5								
Fold Change	GenBank#	ID Number on Volcano Plot	best match	Func	Function	source	description	Gene std dev
-1.58	CN808418	1	T48815	5a	Cell wall	Neurospora crassa	mixed-linked glucanase precursor (MLG1) related protein	0.17
1.52	CN808046	2	CAB43936	5d	Transport proteins	Emericella nidulans	GABA (gamma-amino-n-butyrate) permease	0.17
1.52	CN808955	3	CAC28692	1b	Carbohydrate metabolism	Neurospora crassa	related to mevalonate kinase	0.13
1.64	CN809159	4	CAD70482	1b	Carbohydrate metabolism	Neurospora crassa	related to triose phosphate/3-phosphoglycerate/phosphate translocator	0.11
2.07	CN808905	5	CAE75674	3a	Ribosomal proteins	Neurospora crassa	probable 40s ribosomal protein S6.e, cytosolic	0.13
2.15	CN808112	6	NP_013061	7a	Stress response	Saccharomyces cerevisiae	UBI4 locus contains five-six tandem, in-frame copies of ubiquitin ; transcription is induced in response to several stress conditions; Ubi4p	0.42
2.17	AJ274045	7	EAA76426	9a	Hypothetical Protein	Gibberella zeae PH-1	hypothetical protein FG06966	0.24
2.32	CN809302	8	CAD21421	3d	Prot modification/targeting	Neurospora crassa	probable cyclophilin: catalyzes the cis-trans isomerization of peptide bonds N-terminal to proline residues	0.48
2.93	CN809615	9	EAA58144	9a	Hypothetical Protein	Aspergillus nidulans	hypothetical protein AN6615.2	0.39
4.56	AJ272778	10	EAA64687	9a	Hypothetical Protein	Aspergillus nidulans	hypothetical protein AN2582.2	0.42

Figure 3.2d **Significant Genes with a fold change of 1.5 or greater (part 4 of 4)**

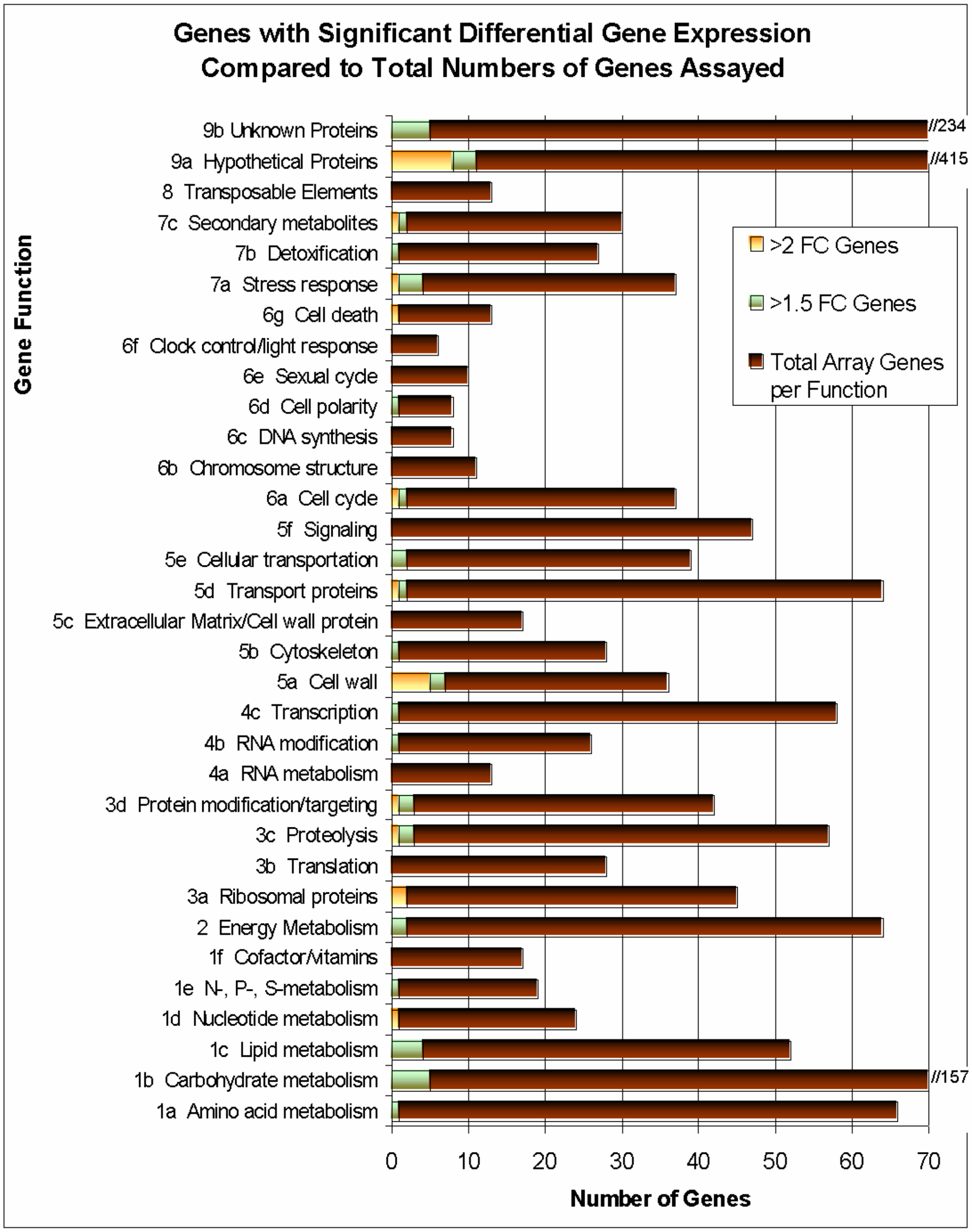


Figure 3.3 **Genes with Significant Differential Gene Expression Compared to Number of Genes Assayed Based on Functional Group.** Thirty three functional groups were assayed. Hypothetical and cell wall genes showed the highest amount of differential gene expression at >1.5 and >2 fold change. No genes involved in pathogenicity, transposable elements, clock control, light control, sexual cycle, DNA synthesis, chromosome structure, extracellular matrix, RNA metabolism, translation or cofactors and vitamins showed any changes in gene expression between the samples.

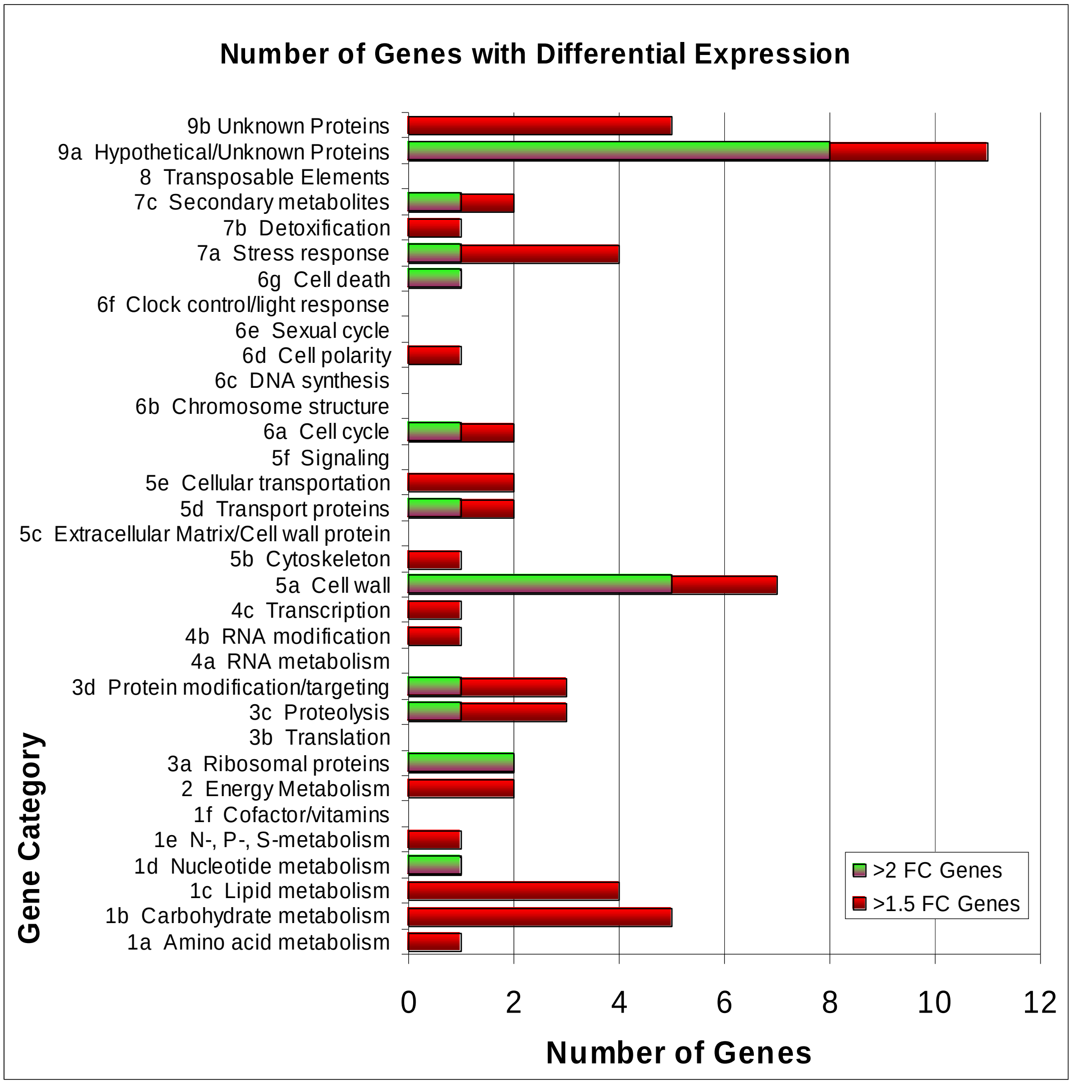


Figure 3.4 **Number of Genes in Each Functional Group with Significant Differential Gene with > 1.5 Fold Change.** The highest number of genes with differential gene expression was for hypothetical proteins, this functional group also contained the largest number of ESTs. Cell wall proteins, ribosomal proteins, transcription, RNA modification, cytoskeleton, transport proteins, cell polarity, stress response, detoxification, secondary metabolites, and unknown proteins all are functional groups that showed significant changes in gene expression. No genes involved in pathogenicity showed altered expression patterns.

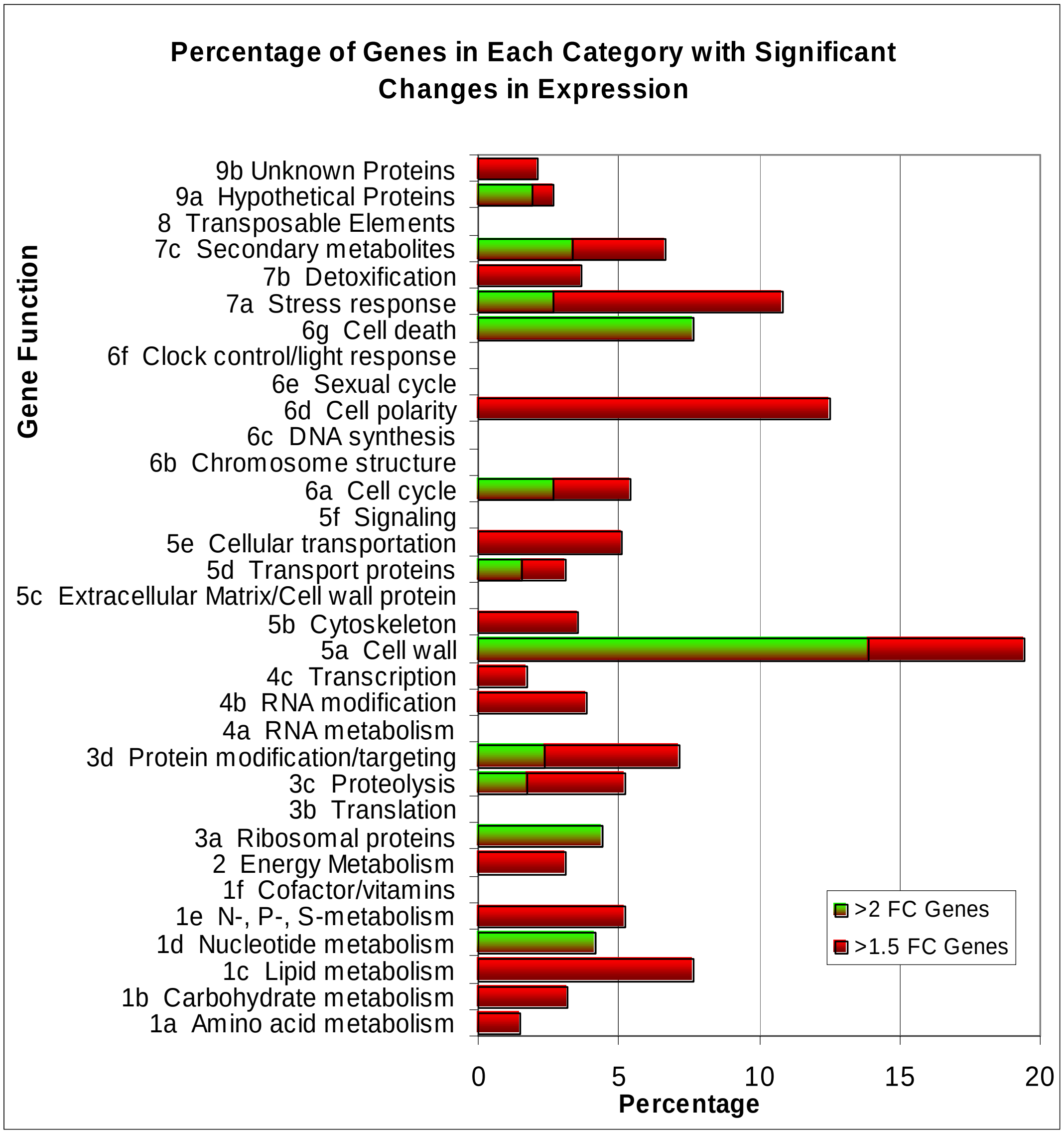


Figure 3.5 **Percentage of Genes in Each Functional Group with Significant Differential Gene with > 1.5 Fold Change.** The highest percentage of genes with differential gene expression were for cell wall, cell polarity and stress response genes. Hypothetical proteins, ribosomal proteins, transcription, RNA modification, cytoskeleton, transport proteins, cell polarity, stress response, detoxification, secondary metabolites, and unknown proteins all are functional groups that showed significant changes in gene expression. No genes involved in pathogenicity showed altered expression patterns.

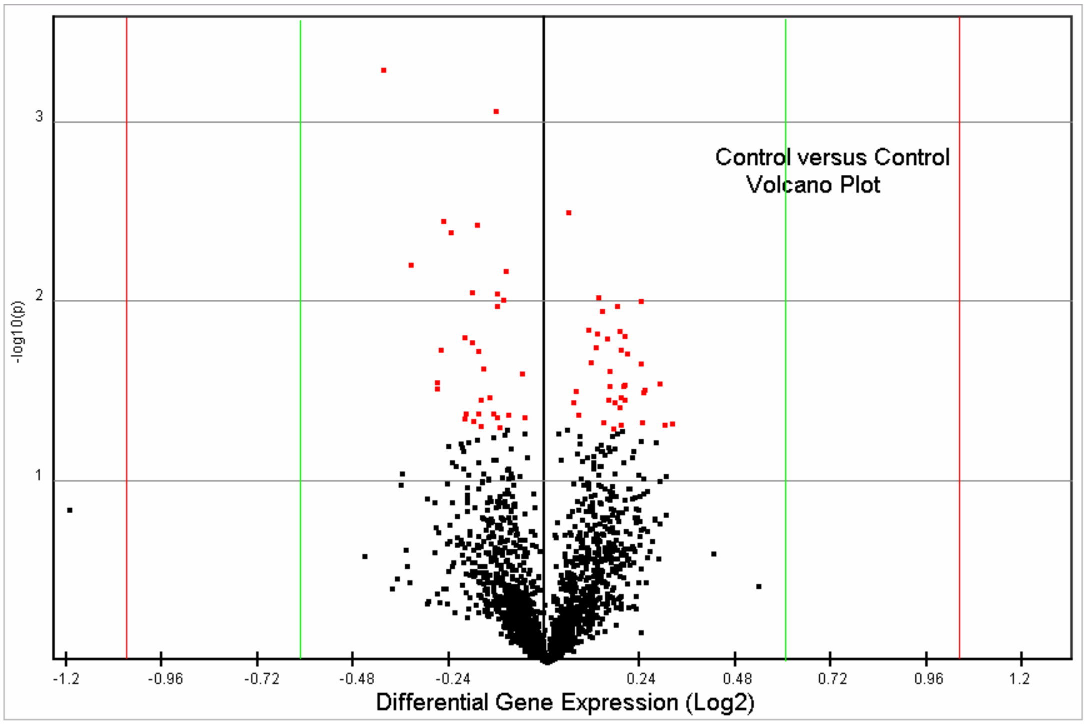


Figure 3.6 **Volcano Plot for Control (input strain) hybridized with Control (input strain).** No genes with a fold change of 1.5 or greater were identified that had significant ($p<0.05$) change in gene expression. The vertical green lines at log2 (.585) denote a fold change of 1.5 and vertical red lines at log2 (1) denote a fold change in gene expression between the two samples of 2. Dots to the left of the black line represent genes that are up-regulated and dots to the right of the right of the black line represents genes that are down-regulated. Genes that were significantly different between the two samples, with a $p < 0.05$ (-log10 1.301), between the two field plots are represented as red dots. Only genes with a fold change of 2.0 (log2 1) or greater were examined in this study. The control slides were performed to determine the amount of background noise and to act as biological and design controls. When the replicates of the controls were compared to each other for microarray analyses, very few changes in differential gene expression were observed. This is what would be expected for the control slides.

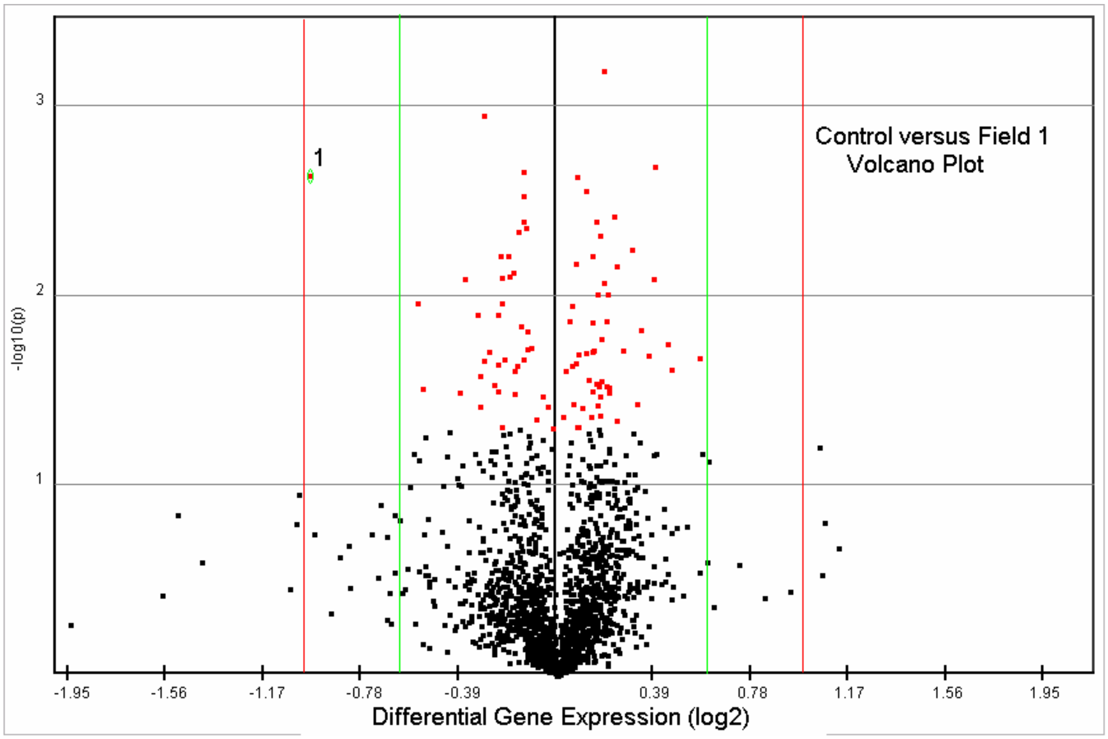


Figure 3.7 **Volcano Plot for Control (input strain) hybridized with Field 1 Recovered Isolates.** One significant gene (green encircled red dot) was down-regulated between a 1.5 and 2 fold change, GenBank #CN808375 (hypothetical protein) with a fold change of -1.99(Labeled 1 on the figure). There were no genes with significant ($p<0.05$) differential expression of 2 fold or greater.

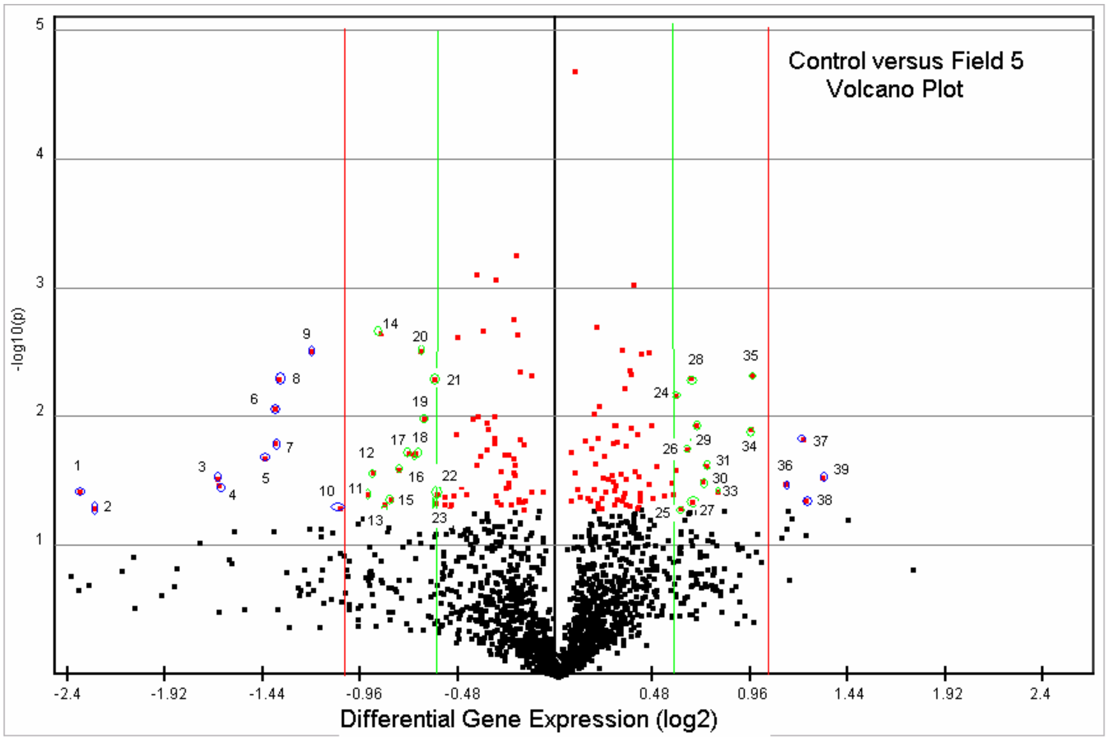


Figure 3.8 **Volcano Plot for Control (input strain) hybridized with Field 5 Recovered Isolates.** Fourteen genes had significant fold changes of >2, the most observed in this assay. The identification of genes numbered in the volcano plot above is listed in Figure 3.2. The genes were 1) GenBank# AJ272832 (unknown function) had a fold change of -5.1156, 2) GenBank# AJ272765 (secondary metabolite, Fum12p required for fumonisin biosynthesis) with a fold change of -4..8654 , 3) GenBank# AJ272811 (putative mitochondrial transport protein) had a fold change of -3.1996, 4) GenBank# AJ272870 (putative function in senescence-associated protein involved in cell death) had a fold change of -3.1856, 5) GenBank# AJ242799 (GEL1 protein involved with the cell wall) had a fold change of -2.7204, 6) GenBank# AJ272761 (unknown function) had a fold change of –2.6334, 7) GenBank#AJ272778 (unknown function) had a fold change of -2.6261, 8) GenBank# CN809178 (a hydrophobic protein of the cell wall) had a fold change of -2.5903, 9) Gen Bank# AJ272786 (a proteolytic protein with a highly conserved ring finger with ubiquitin-protein ligase activity) had a fold change of -2.3159, 10) GenBank# CN809258 (introns-encoded homing endonuclease) had a fold change of -2.1014, 11) GenBank# AJ273457 (putative ribosomal protein L12) had a fold change of 2.1731 , 12) GenBank# CN809336 (unknown function) had a fold change 2.3108 13) GenBank# AJ273011 (Beta-1,3 exoglucanase) had a fold change of 2.3314 and 14) GenBank# CN808187 (cell wall protein) had a fold change of 2.4646. All genes represented by red dots have significant differential expression (p>0.05) between the two RNA populations. Red dots encircled in blue (to the left and right of the vertical red lines) represent 14 genes that had both significant differential expression (p<0.05) and fold changes of 2 or greater. There were 25 significant genes, green encircled red dots between the red and green vertical lines, with that had differential with fold changes between 1.5 and 2.

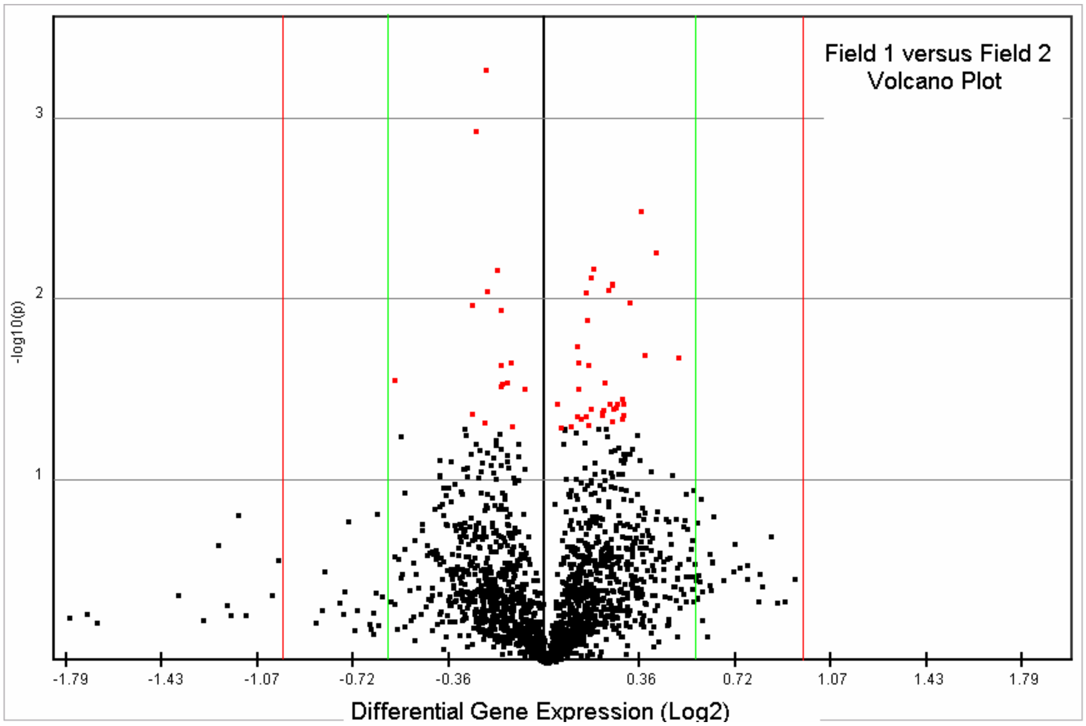


Figure 3.9 **Volcano Plot for Field 1 hybridized with Field 2 Recovered Isolates.** No genes with significant ($p<0.05$) differential expression that showed a fold change of 1.5 or greater.

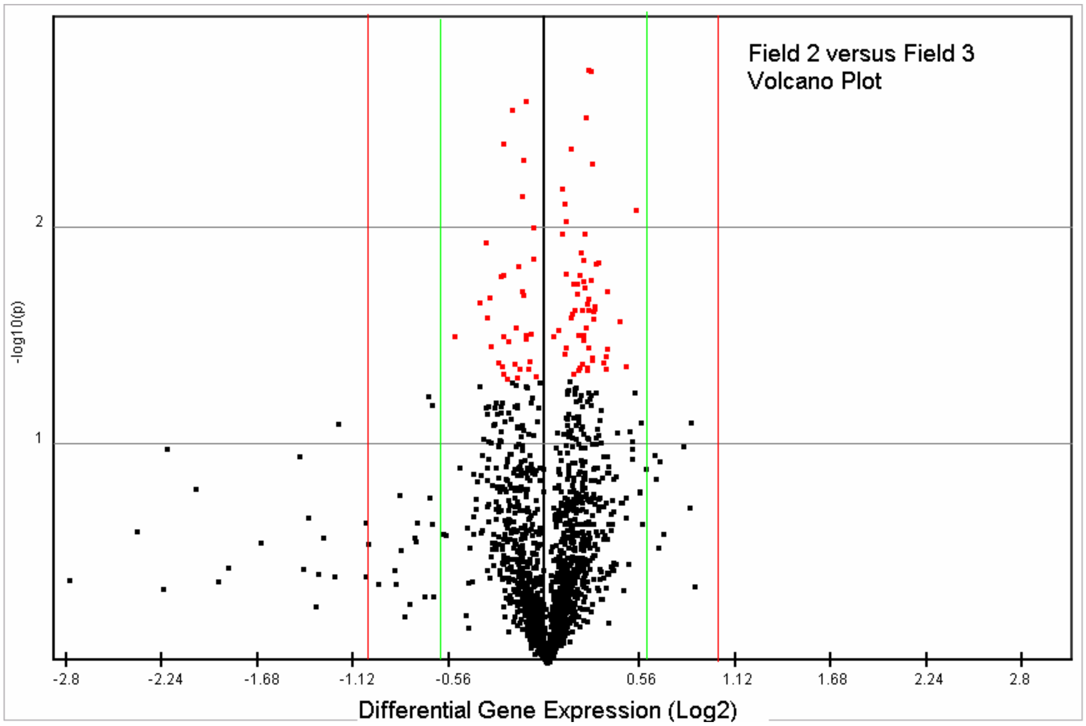


Figure 3.10 **Volcano Plot for Field 2 hybridized with Field 3 Recovered Isolates.** No genes with significant ($p<0.05$) differential expression that showed a fold change of 1.5 or greater.

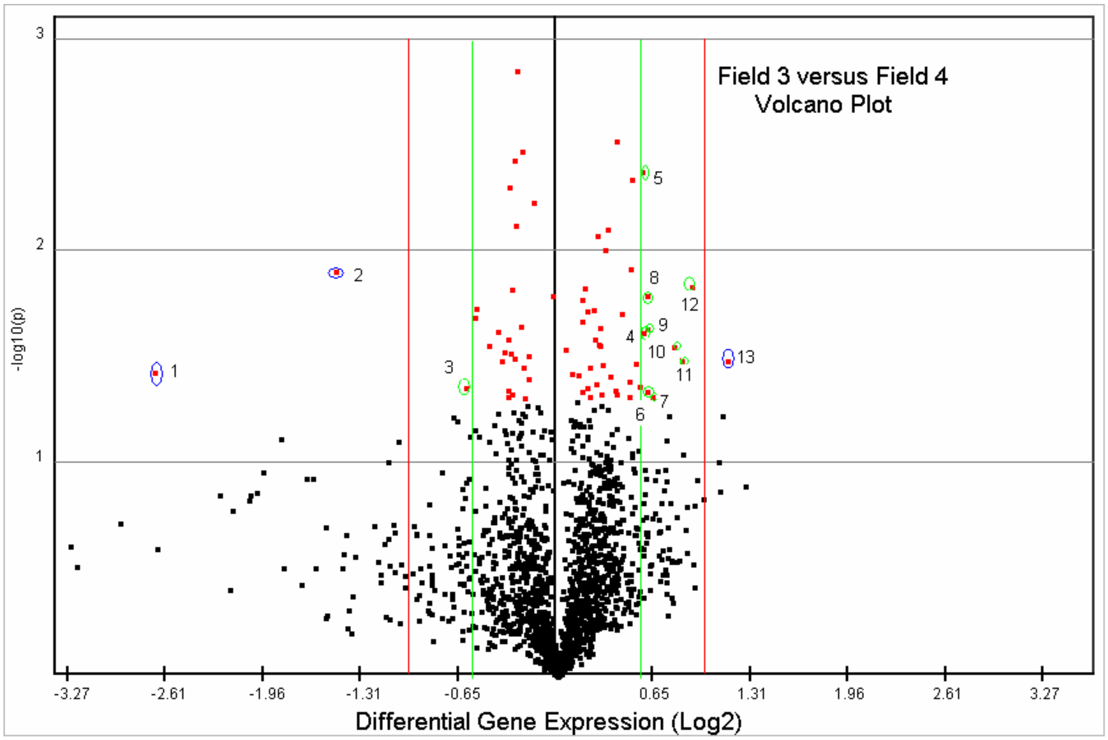


Figure 3.11 **Volcano Plot for Field 3 hybridized with Field 4 Recovered Isolates.** Three genes with a significant fold change of 2 or greater, red dots encircled in blue. The identification of genes numbered in the volcano plot above is listed in Figure 3.2. The three genes were 1) GenBank#AJ274064 (protein associated with the cell cycle regulating and identified as an aging protein) had a fold change of -6.4936 (the highest significant fold change examined in this study) a 2) GenBank# CN808789 (unknown function) had a fold change of -2.8019 and 3) GenBank# AJ273303 (putative functions in cell wall and stress response) had a fold change of 2.1947. Ten genes had significant differentially expressed genes with fold changes from 1.5-2, represented by red dot encircled in green.

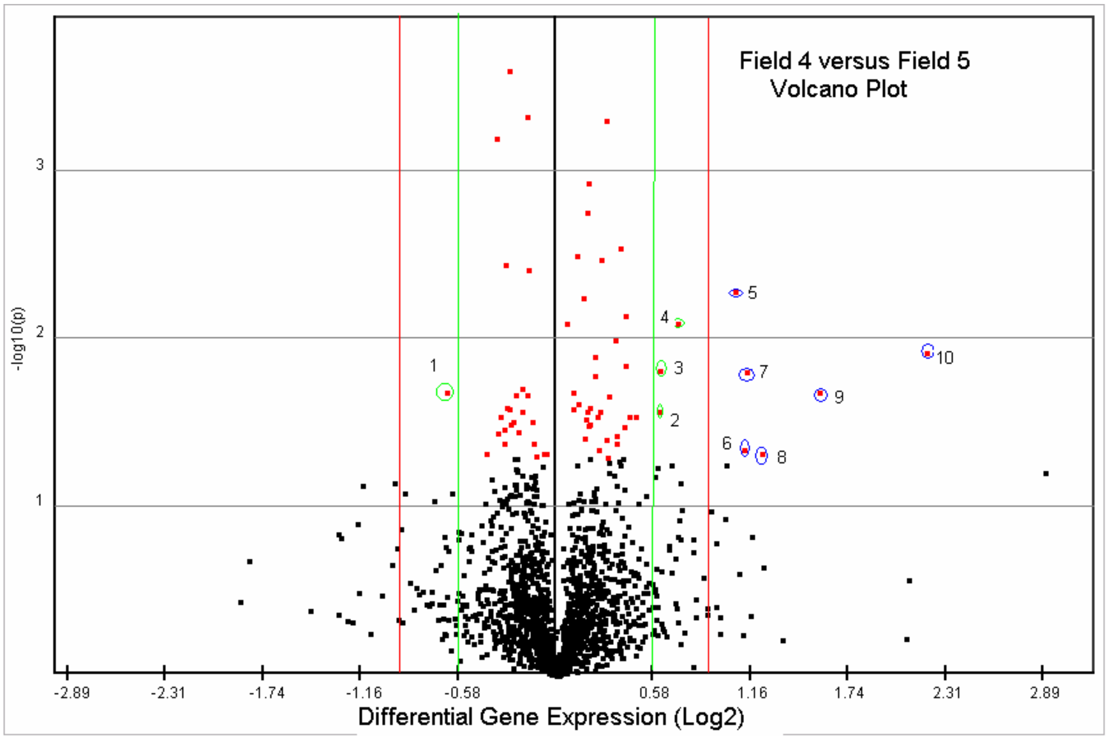
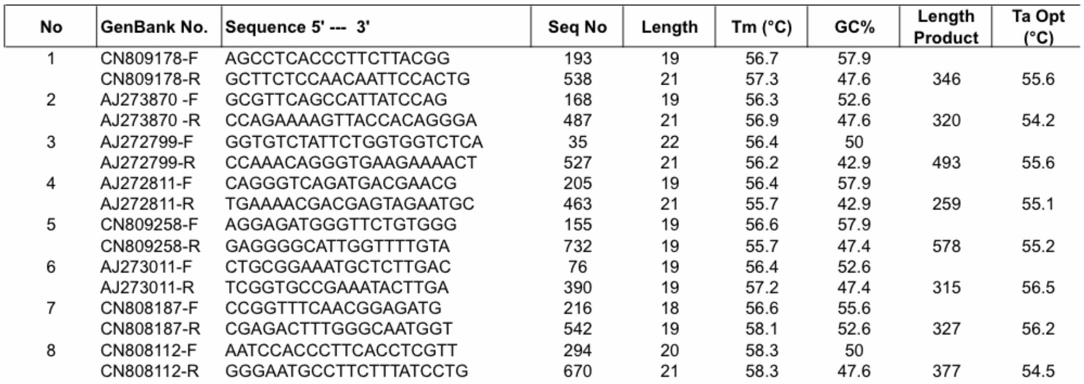


Figure 3.12 **Volcano Plot for Field 4 hybridized with Field 5 Recovered Isolates.** Six genes with a significant fold change of 2 or greater, all were up-regulated and are represented as red dots encircled in blue. The identification of genes numbered in the volcano plot above is listed in Figure 3.2. The six genes were 1) GenBank# CN80895 (a putative cystolic 40s ribosomal protein) had a fold change of 2.0724, 2) GenBank# CN808112 (stress response protein) had a fold change of 2.1542, 3) GenBank# AJ274045 (unknown function) had a fold change of 2.1719, 4) GenBank# CN809302 with a fold change of 2.3156 with a probable function as a cyclophilin, catalyzing the cis-trans isomerization of peptide bonds at the N-terminal to proline, 5) GenBank# CN809615 (unknown function) had a fold change of 2.9344, and 6) GenBank# AJ272778 (unknown function) had a fold change of 4.5575 also with an unknown function. Four genes had significant changes in gene expression with a fold change between 1.5 and 2, represented as red dots encircled in green between the red and green vertical lines.



No	GenBank No.	Sequence 5' --- 3'	Seq No	Length	Tm (°C)	GC%	Length Product	Ta Opt (°C)
1	CN809178-F	AGCCTCACCCTTCTTACGG	193	19	56.7	57.9		
	CN809178-R	GCTTCTCCAACAATTCCACTG	538	21	57.3	47.6	346	55.6
2	AJ273870 -F	GCGTTCAGCCATTATCCAG	168	19	56.3	52.6		
	AJ273870 -R	CCAGAAAAGTTACCACAGGGA	487	21	56.9	47.6	320	54.2
3	AJ272799-F	GGTGTCTATTCTGGTGGTCTCA	35	22	56.4	50		
	AJ272799-R	CCAAACAGGGTGAAGAAAACT	527	21	56.2	42.9	493	55.6
4	AJ272811-F	CAGGGTCAGATGACGAACG	205	19	56.4	57.9		
	AJ272811-R	TGAAAACGACGAGTAGAATGC	463	21	55.7	42.9	259	55.1
5	CN809258-F	AGGAGATGGGTTCTGTGGG	155	19	56.6	57.9		
	CN809258-R	GAGGGGCATTGGTTTTGTA	732	19	55.7	47.4	578	55.2
6	AJ273011-F	CTGCGGAAATGCTCTTGAC	76	19	56.4	52.6		
	AJ273011-R	TCGGTGCCGAAATACTTGA	390	19	57.2	47.4	315	56.5
7	CN808187-F	CCGGTTTCAACGGAGATG	216	18	56.6	55.6		
	CN808187-R	CGAGACTTTGGGCAATGGT	542	19	58.1	52.6	327	56.2
8	CN808112-F	AATCCACCCTTCACCTCGTT	294	20	58.3	50		
	CN808112-R	GGGAATGCCTTCTTTATCCTG	670	21	58.3	47.6	377	54.5

Figure 3.13 **Primers designed for RT-PCR Follow-up**. Eight genes were selected to be examined by RT-PCR based on function and degree of differential expression. Primer lengths were between 17-22 nucleotides and product length were from 259-578 nucleotides.

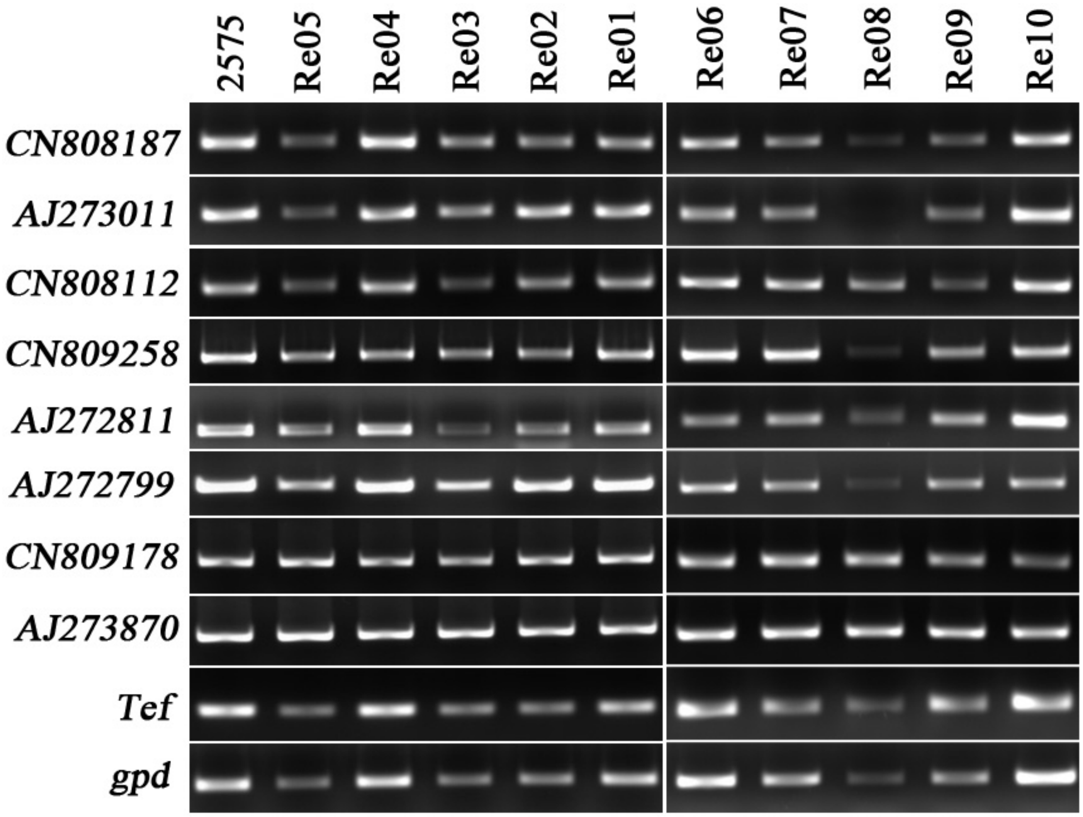
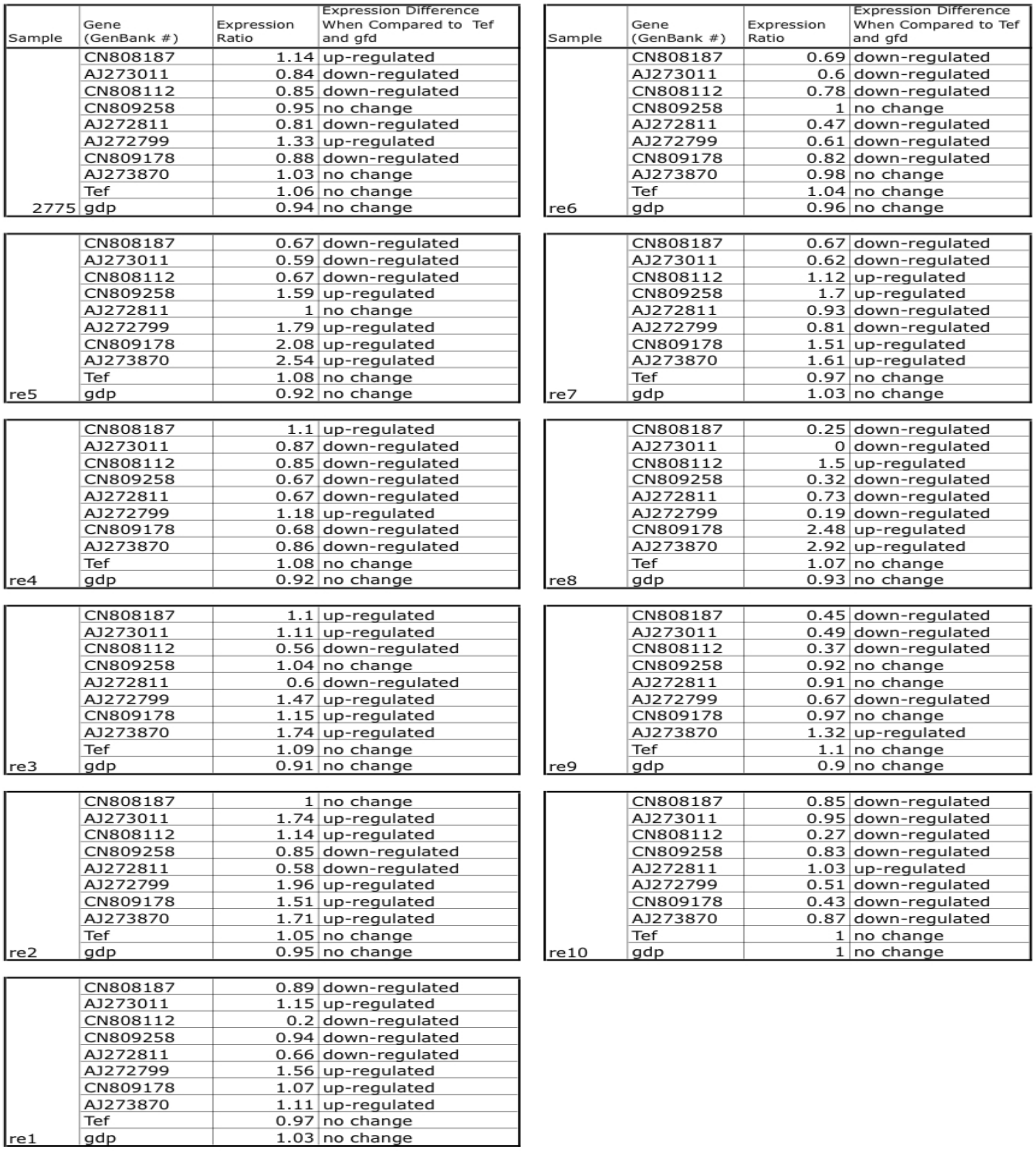


Figure 3.14. **RT PCR Follow-up**. RT PCR confirmed the results from the microarray analyses. RNA intensities from 8 selected genes with significant changes in gene expression, as identified from microarray slides, were examined using RT PCR. Two additional genes *Gpd*, glyceraldehyde 3-phosphate dehydrogenase (Genbank #AY461523) and *Tef*, translation elongation factor 1-α (Genbank #AY445082) were examined as references. *Gpd* and *Tef* have constitutive expression, and are used to compare the expression of the other genes in each column. RNA extracted from the five recovered strains (Re01-Re05) was compared to the input strain (2575) as well as 5 additional recovered strains (Re06-Re10).



Sample	Gene (GenBank #)	Expression Ratio	Expression Difference When Compared to Tef and gfd
2775	CN808187	1.14	up-regulated
	AJ273011	0.84	down-regulated
	CN808112	0.85	down-regulated
	CN809258	0.95	no change
	AJ272811	0.81	down-regulated
	AJ272799	1.33	up-regulated
	CN809178	0.88	down-regulated
	AJ273870	1.03	no change
	Tef	1.06	no change
	gdp	0.94	no change
re5	CN808187	0.67	down-regulated
	AJ273011	0.59	down-regulated
	CN808112	0.67	down-regulated
	CN809258	1.59	up-regulated
	AJ272811	1	no change
	AJ272799	1.79	up-regulated
	CN809178	2.08	up-regulated
	AJ273870	2.54	up-regulated
	Tef	1.08	no change
	gdp	0.92	no change
re4	CN808187	1.1	up-regulated
	AJ273011	0.87	down-regulated
	CN808112	0.85	down-regulated
	CN809258	0.67	down-regulated
	AJ272811	0.67	down-regulated
	AJ272799	1.18	up-regulated
	CN809178	0.68	down-regulated
	AJ273870	0.86	down-regulated
	Tef	1.08	no change
	gdp	0.92	no change
re3	CN808187	1.1	up-regulated
	AJ273011	1.11	up-regulated
	CN808112	0.56	down-regulated
	CN809258	1.04	no change
	AJ272811	0.6	down-regulated
	AJ272799	1.47	up-regulated
	CN809178	1.15	up-regulated
	AJ273870	1.74	up-regulated
	Tef	1.09	no change
	gdp	0.91	no change
re2	CN808187	1	no change
	AJ273011	1.74	up-regulated
	CN808112	1.14	up-regulated
	CN809258	0.85	down-regulated
	AJ272811	0.58	down-regulated
	AJ272799	1.96	up-regulated
	CN809178	1.51	up-regulated
	AJ273870	1.71	up-regulated
	Tef	1.05	no change
	gdp	0.95	no change
re1	CN808187	0.89	down-regulated
	AJ273011	1.15	up-regulated
	CN808112	0.2	down-regulated
	CN809258	0.94	down-regulated
	AJ272811	0.66	down-regulated
	AJ272799	1.56	up-regulated
	CN809178	1.07	up-regulated
	AJ273870	1.11	up-regulated
	Tef	0.97	no change
	gdp	1.03	no change

Sample	Gene (GenBank #)	Expression Ratio	Expression Difference When Compared to Tef and gfd
re6	CN808187	0.69	down-regulated
	AJ273011	0.6	down-regulated
	CN808112	0.78	down-regulated
	CN809258	1	no change
	AJ272811	0.47	down-regulated
	AJ272799	0.61	down-regulated
	CN809178	0.82	down-regulated
	AJ273870	0.98	no change
	Tef	1.04	no change
	gdp	0.96	no change
re7	CN808187	0.67	down-regulated
	AJ273011	0.62	down-regulated
	CN808112	1.12	up-regulated
	CN809258	1.7	up-regulated
	AJ272811	0.93	down-regulated
	AJ272799	0.81	down-regulated
	CN809178	1.51	up-regulated
	AJ273870	1.61	up-regulated
	Tef	0.97	no change
	gdp	1.03	no change
re8	CN808187	0.25	down-regulated
	AJ273011	0	down-regulated
	CN808112	1.5	up-regulated
	CN809258	0.32	down-regulated
	AJ272811	0.73	down-regulated
	AJ272799	0.19	down-regulated
	CN809178	2.48	up-regulated
	AJ273870	2.92	up-regulated
	Tef	1.07	no change
	gdp	0.93	no change
re9	CN808187	0.45	down-regulated
	AJ273011	0.49	down-regulated
	CN808112	0.37	down-regulated
	CN809258	0.92	no change
	AJ272811	0.91	no change
	AJ272799	0.67	down-regulated
	CN809178	0.97	no change
	AJ273870	1.32	up-regulated
	Tef	1.1	no change
	gdp	0.9	no change
re10	CN808187	0.85	down-regulated
	AJ273011	0.95	down-regulated
	CN808112	0.27	down-regulated
	CN809258	0.83	down-regulated
	AJ272811	1.03	up-regulated
	AJ272799	0.51	down-regulated
	CN809178	0.43	down-regulated
	AJ273870	0.87	down-regulated
	Tef	1	no change
	gdp	1	no change

Table 3.1 **RT PCR Densitometry Ratios.** RT PCR data were analyzed by using the densitometer program Un-Scan-It™. Intensities for the two constitutive bands, *Tef* and *Gdp* were averaged. The average calculated average value was then used to determine a ratio of expression for the *Tef* and *Gpd* gene intensity. The range between the *Tef* and *Gdp* intensities was set as the base-line. The intensities for the other 6 genes (indicated by their Genbank identification numbers)were divided by the average *Tef-Gdp* intensity value. Calculated ratio values higher than the *Tef* or *Gdp* ratio values were up-regulated, values in the same range as the *Tef-Gdp* ratios showed no change, and values less than the *Tef-Gdp* ratio were down-regulated. Intensity ratios for the original input samples (2575) were compared to the other 10 recovered isolated (Re1-Re10) to determine the changes in gene expression between samples. Based on comparing the intensity ratios, RT PCR corroborated microarray results.

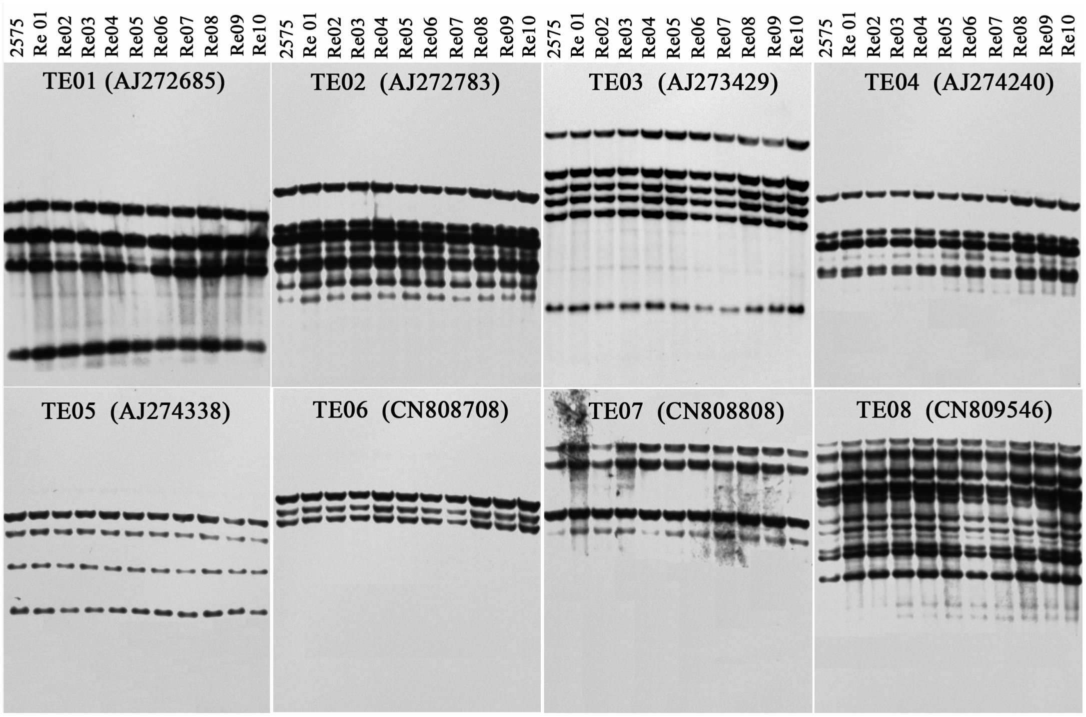


Figure 3.15 **Southern Blot Images for Ten Recovered Isolates (Re)**. Eight transposable elements (TE01-TE08, GenBank#J272685, AJ273429, AJ274240, AJ274338, CN808708, CN808808, and CN809546) were used as probes to test for any changes in chromosome structure. Ten field isolates collected from the field 3.5 years after inoculation. DNA was extracted form these isolates as well the control input lab strain (2575). When compared to the control, there were no observable differences between the 10 recovered isolates (Re). Five of these isolates were used for microarray analyses.

Discussion

Rapid evolution of new phenotypes is usually caused by changes in gene expression (Ben-Shahar *et al.,* 2002; Enard *et al*., 2002). In this study, array based mutation accumulation assays were used to inventory strains recovered from the field site for changes in gene content and altered patterns of gene regulation that can be used to predict any physiological changes (Wang and St. Leger, 2005; Wang *et al.,* 2005b).

Array based assays of mutation rates have not been performed with other fungal species. Therefore, it cannot be said that gene expression divergence in each isolate compared to the input strain is comparatively fast or slow by the standards of other fungi. In absolute terms, the rate at which mutation produces statistically significant new variation in gene expression seemed slow because only 1.31% of the arrayed genome of the 5 isolates were affected. Rates of mutation could be affected by the generation time of *M. anisopliae* in natural settings, and it can take up to three weeks from infection of an insect to sporulation on an insect cadaver. The introduced *M. anisopliae* is not dormant, as *Metarhizium* can be isolated from insects and about 1% of colonies isolated from the field are unstable diploids that spontaneously breakdowns to form aneuploids (Wang, O'Brien, Pava-Ripoll and St. Leger, Unpubl.). Although spontaneous mutations have the potential to generate abundant variation in gene expression, natural variation may be constrained by purifying selection. By comparing the five isolates that were collected after 3.5 years on the field, we attempted to determine if all biological processes or molecular functions were equally mutable or conserved. Genes with altered accumulation of transcripts in isolates 4 and 5 included several involved in cell wall synthesis and ribosomal proteins. However, at 2-fold level of differentiation, genes for secreted enzymes, virulence factors, genes required for saprotrophy and metabolic pathways were highly conserved between all isolates. Therefore for much of the genome, stabilizing selection dominates intrastrain transcriptome evolution of *M. anisopliae.* However, the isolates originated from replicated plots and the soil habitat will likely have been close to those to which the fungus, originally isolated in South Carolina, was already adapted to. At least in *Drosophila*, there is a strong correlation between mutational variance and environmental variance (Rifkin *et al*., 2005).

The mutations in isolates 4 and 5 affected different genes for cell wall proteins, however similar changes in gene regulatory profiles between these two isolates from replicate plots could show selection for certain genes to become canonically overexpressed or silent in turf, indicative of Darwinian selection (rapid evolution and acquisition of new functions) (Rifkin *et al*., 2005). Populations declined over the duration of the field trial indicating that selection pressures were presenting the turf. Cycling of the fungus was indicative that some members of the introduced populations of *Metarhizium* were adapting better to the turf. Changes in expression of genes for cell wall proteins would signify raw material for evolutionary diversification of structural components interphasing with the environment. A caveat to this is that production of a particular product at a specific time and place may be the target of selection rather than gene expression itself. As in enzyme flux models, the selective effect of any particular change in gene expression may be negligible over a range of values but become substantial when the abundance of mRNA becomes rate limiting or when the variation becomes otherwise functional relevant (Rifkin *et al.*, 2005).

Identifying the potential for transposons, insertion elements and other mobile elements in recombinant microorganisms to influence the potential for genetic mobility was identified as a high priority at a recent stakeholder workshop. An expansion of a subset of insertion elements in *M. anisopliae* strain 443 which hybridized to respective sequences from 2575 suggests that evolution could occur in leaps, resulting in consequences for product stability and pathogenic potential. Having obtained the jumping genes as 2575 ESTs, we used these as probes to assay for frequency of jumping events and to identify increases in copy number in rescued colonies by Southern blot analysis. These data showed that *M. anisopliae* displayed no observable chromosomal changes over three and a half years in the field. This genetic stability *of M. anisopliae* is important not only to understand how this fungus adapts, but in a broader sense to examine the stability of transgenic organisms. This is the first time that microarray and transposon analyses have been used to investigate the rate of genetic divergence and long-term genetic stability of a fungus in field conditions.

Chapter 4: Using winter wheat, *Triticum aestivum*, to examine the ability of *Metarhizium anisopliae* to localize in the rhizosphere and promote plant growth.

Introduction

Metarhizium has been extensively studied as a sustainable mycoinsecticide to control insects such as thrips, termites and Japanese beetles (Villani *et al.*, 1994; Maniania *et al.*, 2003; Wright *et al.*, 2005), but the possibility of using *Metarhizium* as a rhizospheric fungus to benefit plant growth and crop yields in other manners has only recently attracted attention (Maniania *et al.*, 2003; Kabaluk and Ericsson, 2007). To begin studying the benefits *Metarhizium* may confer to plants, one must examine the most suitable method of application for the crop at hand. Many options exist for applying *M. anisopliae* including direct application of conidia to seeds, applying the fungus in the form of granules around the crop, or spraying a suspension of spores over the foliage and soil (Bruck, 2005; Jaronski *et al.*, 2007). Understanding *M. anisopliae's* life outside of its host, principally at the rhizosphere, is also needed for development of an effective delivery system for applying this entomopathogen in successful commercial applications (Bruck, 2005).

The ability of *M. anisopliae* to colonize roots was demonstrated with cabbage (Hu and St. Leger 2002) and has since been extended to many plant systems. The roots of spruce grown in containers treated with *M. anisopliae* showed much higher levels of the fungus in areas surrounding the root than in other areas of the container.

M. anisopliae also infected over 70% of the second and third instars of the black vine weevil infesting the contained spruce (Bruck and Donahue, 2007).

Seed treatment using *M. anisopliae* applied to corn, *Zea mays*, was shown to protect crops against herbivory by the wireworm, *Agriotes obscurus* and consequently increased yields (Kabaluk and Ericsson, 2007). Colonization of *M. anisopliae* at the rhizosphere may also have increased the rate of corn seed germination and plant growth. This would allow the plant to establish earlier in the season, thus increasing nutrient uptake by the plant, as plants in the study weighed more than uninoculated controls. What was particularly interesting about the corn experiment was that both corn treated with just conventional insecticides and corn treated with just *M. anisopliae* had increased yields when compared to the control, while corn treated with both insecticides and *M. anisopliae* had the highest yields, implying that the application of *M. anisopliae* conferred more than just protection to insect pests (Kabaluk and Ericsson, 2007). A field trial designed to test the effectiveness of *M. anisopliae* to control onion thrips, *Thrips tabaci*, showed similar results. After three seasons of conidial application, onion bulb yield was greatest in the field treated with *M. anisopliae* (Maniania *et al.*, 2003).

Metarhizium may be more than just an entomopathogen, Metarhizium may act as a biofertilizer

M. anisopliae may provide other benefits to the plant besides protection from herbivory. The relationship between crop losses and disease has been studied in multiple systems. Entomopathogenic fungi have been shown to release a number of bioactive compounds, some of which may act to inhibit the growth and development of insects, bacteria and other fungi (Selitrennikoff, 2001; Leger, 2004; Lee *et al.*, 2005). *M. anisopliae* has antagonist effects on various plant pathogens, including *Fusarium oxysporum* and *Alternaria solani,* which result in mycelial inhibition zones when they are grown together with *M. anisopliae* on nutrient agar (Kang *et al,* 1996). An area that has not been as well investigated is the potential of *M. anisopliae* to act as a biofertilizer by increasing a plant's capacity to obtain nutrients not readily available in the soil.

Biofertilizers are living organisms or byproducts from organisms that increase the accessibility of nutrients and resources such as zinc, nitrogen, water and phosphate that would otherwise be unavailable to the plants accessible (Behl *et al*., 2007). Phosphorus is second only to nitrogen as the most required nutrient for plants. But up to 99% of the phosphorus present in soil is inaccessible to the plants as phosphate is present in an insoluble form (Vassileva *et al*., 1998; Rajankar *et al.*, 2007). The addition of soluble phosphate to soil provides only a short-term solution for plants, as the phosphate is quickly converted into an insoluble form in soil (Omar, 1998). Many plant-associated microorganisms including bacteria and fungi are capable of solubilizing inorganic phosphate, with certain species of fungi having a greater capacity for solubilizing inorganic phosphate (Singal *et al*., 1994; Asea *et al*., 1998; Omar, 1998). Arbuscular mycorrhiza contribute to cycling of phosphorus, creating a sustainable nutrient supply to the plant (Tore *et al*., 1997). Mechanisms utilized by these phosphate-solubilizing microorganisms include producing chelating ions and acids to lower pH (Kang *et al*., 1996; Smiley and Patterson, 1996; Dadarwal, 1997; Yadav and Dadarwal, 1997; Kang *et al.*, 1998; Rajankar *et al.*, 2007). The ability of *M. anisopliae* to perform such cycling has yet to be determined. But it has been reported that *M. anisopliae* is capable of altering its ambient pH by producing organic acids; therefore *Metarhizium* may be able to make insoluble phosphate available to plants thereby acting as a biofertilizer (Singal *et al*, 1994*;* St. Leger *et al.*, 1999).

Biofertilizers not only have been shown to increase plant mass and yields, but they are sustainable and provide a better quality and more uniform crop (Maniania *et al.*, 2003; Behl *et al.*, 2007). Many options exist to increase plants yields, such as applying chemical insecticides and fertilizers, which are expensive and may have unwanted environmental consequences. Another option is to create transgenic plants that are able to grow faster, produce greater yields, and are more resistant to plant pathogens and insect pests. But such plants require time and money to create; in addition, pathogens and pests have been shown to evolve rapidly and regain the ability to target these plants (Broglie *et al.*, 1991; Grover and Gowthaman, 2003). Designing a technique that could be applied to multiple plants systems, which would allow the plant to establish faster therefore out compete weeds and pathogens may be a better long-term, sustainable, environmentally friendly option. This economic and long-term option for increased crop yields may be obtained by understanding the underlying nature of the rhizospheric interaction of potentially mutualistic fungus like *M. anisopliae* with crop plants to develop of *Metarhizium* as a comprehensive plant symbiont.

The rhizosphere is a complex ecosystem with various fungi, bacteria, animals and plants competing for limited resources. Abiotic factors such as soil composition, pH, temperature, and rainfall, as well as biotic factors such as other microorganisms inhabiting the rhizosphere, all play roles in determining rhizospheric competence (Bateman and Kwasna, 1999; Brimecombe *et al*., 2001). While conditions in these lab studies do not recreate the conditions found in nature, they may elucidate some of the complex interactions between various rhizospheric fungi and plants. These experiments set out to examine if *M. anisopliae* can successfully colonize the rhizosphere when conidia were applied to the seeds before planting in both laboratory and field settings, and the effects of such applications on plant growth and yield.

Scope of this research

In previous field studies with corn, *M. anisopliae* applied to seeds significantly increased yields (Kabaluk and Ericsson, 2007). *Metarhizium* could however have a multifactorial influence on plant growth through either protecting the plants against potential pathogens or by acting as a biofertilizer. The current field study with winter wheat showed that treating seeds with the wild type *M. anisopliae* increased the average weight of the harvested seeds by 14.9%. A non-virulent mutant (Δ*Mcl1*) increased seed weight by 3.38%, while the mutant (Δ*Mad2*) that was unable to adhere to root surfaces had no beneficial effect. A previous field study with onions showed a 29.2% or 7 metric tons ha^{-1} increase in yield when *M. anisopliae* was applied to control onion thrips versus when the insecticide dimethoate was used for insect control of thrips (Maniania, 2003). The importance of *M. anisopliae* as an entomopathogen was demonstrated by these field trials. However, other benefits that *M. anisopliae* may confer to the plant were not fully examined in the field. Follow-up experiments using microcosms were designed to test if *M. anisopliae* may act as a biofertilizer, solubilizing insoluble phosphate and thus making phosphate accessible to the plant. Winter wheat was chosen for both the field and microcosm experiments as winter wheat is a well studied system and its yield correlates with factors such as the amount of water and fertilizer applied (Fouly *et al*., 1996; Sieber *et al*;, 1988, Gurha and Srivastava, 2002; Behl *et al.*, 2007).

Materials and Methods

Field trial to examine how the application of M. anisopliae, M. anisopliae ΔMcl (non-virulent) and M. anisopliae ΔMad2 (unable to adhere to root surface) affect crop mass

Seeds of *Triticum aestivum* variety Expedition, a cultivar of red winter wheat, were certified organic, certificate number CC025219 and purchased from Albert Lea Seed House, 1414 West Main P.O. Box 127, Albert Lea, MN 56007 were inoculated prior to planting. The winter wheat seeds were treated with 100 µl per seed of a suspension of *M. anisopliae* conidia (10^8 conidia l^{-1} in 10% filter–sterilized gum arabic) and allowed to air dry in a sterile hood. A total of 20 seeds were planted per square meter plot, for a total application of approximately 10^4 l^{-1} conidia per m^2.

Twenty four hours before seeds were planted in the field in the field, they were treated with four different preparations: 1) GFP labeled *M. anisopliae*, 2) *M. anisopliae* Δ*Mcl*1, the immune evasion non-pathogenic mutant, 3) *M. anisopliae* Δ*Mad2*, the mutant unable to adhere to plant epidermis, and 4) gum arabic solution (control). The seeds for each treatment were sown in five randomly assigned replicate plots (Figure 4.1). Each plot measured one square meter. Seeds were planted every 200 cm, for a total of 20 seeds per plot. One hundred seeds were planted for each of the four treatments. The seeds were planted at the beginning of October to allow time for root and shoot development before first frost. Immediately before planting, samples of soil (from the top 5 cm, the depth at which most fungi are found under natural conditions) were taken from 30 random locations within the application area and control plots to characterize the background levels of *M.*

anisopliae. Samples of non-rhizospheric soil from evenly spaced locations within the grass free paths (taken using a 1-cm soil-core sampler) were also analyzed to monitor dispersal and persistence of transgenic fungi.

The plots for planting were not treated with any chemical fertilizers or insecticides for the duration of the experiment. Ten randomly selected plants per treatment per plot were extricated at the end of the growing season in June. The efficacy of an entomopathogen depends on its ability to prevent damage to the plant and increase mass, so seeds were counted and weighed to measure mass. Random samples from various lengths (distal, medial, and proximal to the seed) of the roots were analyzed for the presence of the fungus. To monitor migration of GFP-2575, samples were obtained by collecting soil loosely adhering to roots (outer rhizosphere), and the roots were sectioned in sterile water to assay fungus in the inner rhizosphere and rhizoplane. An Olympus SZX12 with a GFP filter 470/500 was used to examine all recovered CFUs and plant material.

Average seed weight was analyzed using SAS to perform an analysis of variance (ANOVA). Data were analyzed using proc mixed protocol then Tukey's Studentized Range (HSD) Test was used to compare data sets for the average seeds weights for seeds recovered for plants grown from seeds treated with: *Metarhizium*-GFP conidia, *Metarhizium* Δ*Mcl1* conidia, *Metarhizium* Δ*Mad2* conidia, gum-arabic, and the original weight of the 400 seeds planted.

Figure 4.1. **Schematic of the Random Block Design Used to Assign Samples to Each Plot.** A random block design was used to assign the treated winter wheat seeds to each plot. There was a minimum of a one meter buffer zone between each plot. The control plots were separated from the treated samples by minimum of a seven meter buffer zone. There were five replicates for each treatment: seeds treated with GFP-labeled *M. anisopliae* (GFP 1-5), seeds treated with GFP-labeled *M. anisopliae* ΔMad2 (MAD 1-5), seeds treated with GFP-labeled *M. anisopliae* ΔMcl1 (MCL 1-5) and control seeds treated only with filter-sterilized gum arabic (control). Plots were labeled with color-coded with spray paint for easy identification.

Examination of Metarhizium as a potential biofertilizer

The ability of *M. anisopliae* to act as a biofertilizer by increasing either germination rates and/ or rate of plant growth was examined by using agar plates with insoluble inorganic phosphate salts added. The plates were opaque, but solubilization of the phosphate produced visible clearing zones. The media used to examine clearing caused by solubilizing inorganic phosphate salts consisted of 1.2% wt/volume agar, 10 $g{\cdot}l^{-1}$ glucose, 5 $g{\cdot}l^{-1}$ tri-calcium phosphate, 0.27 $g{\cdot}l^{-1}$ $NH4{\cdot}NO3$, 0.2 $g{\cdot}L^{-1}$ KCl, 0.1 $g{\cdot}l^{-1}$ $MgSO4{\cdot}7H2O$, 1 $mg{\cdot}l^{-1}$ $MnSO4{\cdot}6H2O$, 1 $mg{\cdot}l^{-1}$ $FeSO4{\cdot}7H2O$ and 0.1 $g{\cdot}l^{-1}$ yeast extract (Song *et al.*, 2008). Ten replicates were plated for each of the following fungal strains: *M. anisopliae, M. anisopliae* Δ*Mcl1, M. anisopliae* Δ*Mad2, M. anisopliae* mutant that overproduced oxalic acid, *M. anisopliae* that underproduced oxalic acid and *A. niger* (as the positive control). Fungi were placed on the center of a plate and clearing zones were measured in mm after incubating at 28˚C for 72 hours.

The ability of *M. anisopliae* to function as a biofertilizer in microcosms and make insoluble phosphate available to winter wheat seedlings was tested. Alcohol sterilized seeds were inoculated with 10^4 l^{-1} conidia of *M. anisopliae* and placed in autoclaved soil amended with 50 mg of insoluble tri-calcium phosphate per kg of soil (Schubert and Hayman, 1986). Controls included: 1) untreated seeds grown in calcium phosphate treated soil, 2) seeds treated with *M. anisopliae* conidia prior to planting, grown in soil with 50 mg of soluble phosphate (potassium monophosphate and potassium biphosphate calibrated to a pH of 7.88) per kg of autoclaved soil, 3) seeds without conidia treatment, grown in soil with one gram of soluble phosphate,

4) untreated seeds grown in unamended soil and 5) seeds inoculated with conidia of *M. anisopliae* prior to planting and grown in unlamented soil. Plant growth was measured by examining increase in blade length. Growth rates were compared for these conditions. This experiment was repeated three times, with ten replicates per six conditions. All replicates contained one seed per 50 ml sterile conical tube.

Plants were carefully removed from the 6 microcosms after 16 days. Plants were cleaned of soil, and allowed to air dry for 48 hours. Dried biomass was determined by weighing the air dried plants.

Unpaired two-tailed t tests with a standard p value of 0.05 were used to compare growth rates of the control seeds to the seeds inoculated with Metarhizium, *Metarhizium ΔMcl1* and *Metarhizium ΔMad2*.

Data from the 6 microcosms: 1) control with untreated seed and no soil phosphate, 2) control untreated seed grown in soil amended with insoluble phosphate, 3) control untreated seed grown in soil with soluble phosphate, 4) *Metarhizium* treated seed and no soil treatment, 5) *Metarhizium* treated seed grown in soil with insoluble phosphate and 6) *Metarhizium* treated seed grown in soil with soluble phosphate were analyzed using SAS to perform an analysis of variance (ANOVA). Data were analyzed using proc mixed protocol and repeated measures generating an autocorrelation variation matrix to adjust for repeated measure correlation between the 6 microcosms growth rate. Tukey's Studentized Range (HSD) Test was used to compare data sets for the growth rates for the six microcosms. Biomass was analyzed using SAS to perform an analysis of variance (ANOVA). Data were analyzed using proc mixed protocol and repeated measures generating an autocorrelation variation matrix to adjust for repeated measure correlation between the 6 microcosms growth rate. Tukey's Studentized Range (HSD) Test was used to compare data sets for the biomass for plants recovered from the six microcosms

Results

Results from a field trial examining how the mass of winter wheat is affected by treatment of seeds with M. anisopliae

Prior to commencing the field trial, the soil was tilled multiple times, and roots were removed from the area. Samples of soil were taken before and after the field trial and no CFUs of *M. anisopliae* were observed from soil in the plots and surrounding tilled area. Seed mass was measured as average weight per seed (2400 seeds harvested) (Figure 4.2), and the presence of GFP-expressing fungi on and near the roots was recorded. GFP was not found on any area of the plant besides the roots. The average weight of each seed recovered from the plants grown with *gfp*-2575 treated seeds was 0.037 (±0.001) gram/seed was significantly greater than the control seeds 0.032 (± 0.001) g/seed recovered from plants grown from seeds treated only with filter-sterilized gum arabic control plot (f=157.82, df=4, p<0.0001). The average weight of each seed harvested from plants grown from the *gfp*-2575 treated seeds was also significantly greater than the average weight of seeds harvested from plants grown from Δ*Mcl1* treated seeds was 0.033 (±0.001) g/seed. The average weight of each seed harvested from plants grown from the *gfp*-2575 treated seeds was also significantly greater than the average weight of seeds harvested from plants grown from Δ*Mad2* treated seeds which were 0.032 (± 0.002) g/ seed. While there was an increase in seed weight for the Δ*Mcl1* treated seeds, the increased seed weight was not significant when compared to the control seeds. The average weight of the original 400 seeds planted was 0.031 (± 0.002) g/seed was not significantly different than the average weight of seeds recovered from the control plots.

Seven months after planting the seeds, plants were carefully harvested from the plots, the roots shaken free of loosely adhering soil and examined under UV dissecting scope for GFP fluorescence. None of the plants recovered from any of the plots had *M. anisopliae*-GFP present on the leaf blades or stems. Of the winter wheat plants from the *M. anisopliae*-GFP plots, 28.6% showed ~ 0.1 mm patches of florescence along the roots indicating the presence of transgenic *M. anisopliae.* (Figure 4.3). Of the winter wheat plants from the Δ*Mcl1* plots, 22 % showed similar fluorescent patches. None of the plants recovered from the control or Δ*Mad*2 plots showed fluorescence indicating the absence of transgenic *M. anisopliae.* Seed count and total plant mass were not used as indicators as severe weather caused damage to the wheat blades and caused some seeds to fall from their sheaths. Roots were sectioned into 5 cm lengths and plated onto PDA plates to confirm the presence of *M. anisopliae*-GFP (Figures 4.4 and 4.5). The principal root colonizers growing out on PDA were identified by Karen Rane at The University of Maryland's Plant Diagnostic Laboratory (room 3171 Department of Entomology). They included yeast, *Gliocladium*, *Penicillium*, *Paecilomyces*, *Cladosporium*, and *Alternaria* species. *Alternaria* spp. was the only fungi identified on the wheat seeds, and was present on all seeds harvested. All of these are normal saprophytes of wheat indicating that our field study took place against a commonplace diversity of indigenous fungi. Although on PDA plates these other fungi seemed to be effective competitors with *M. anisopliae*, the greater preponderance of *M. anisopliae* 2575 on freshly harvested roots suggest that in the field soil environment they have a much different effect. They could possibly even synergize the impact of *M. anisopliae*.

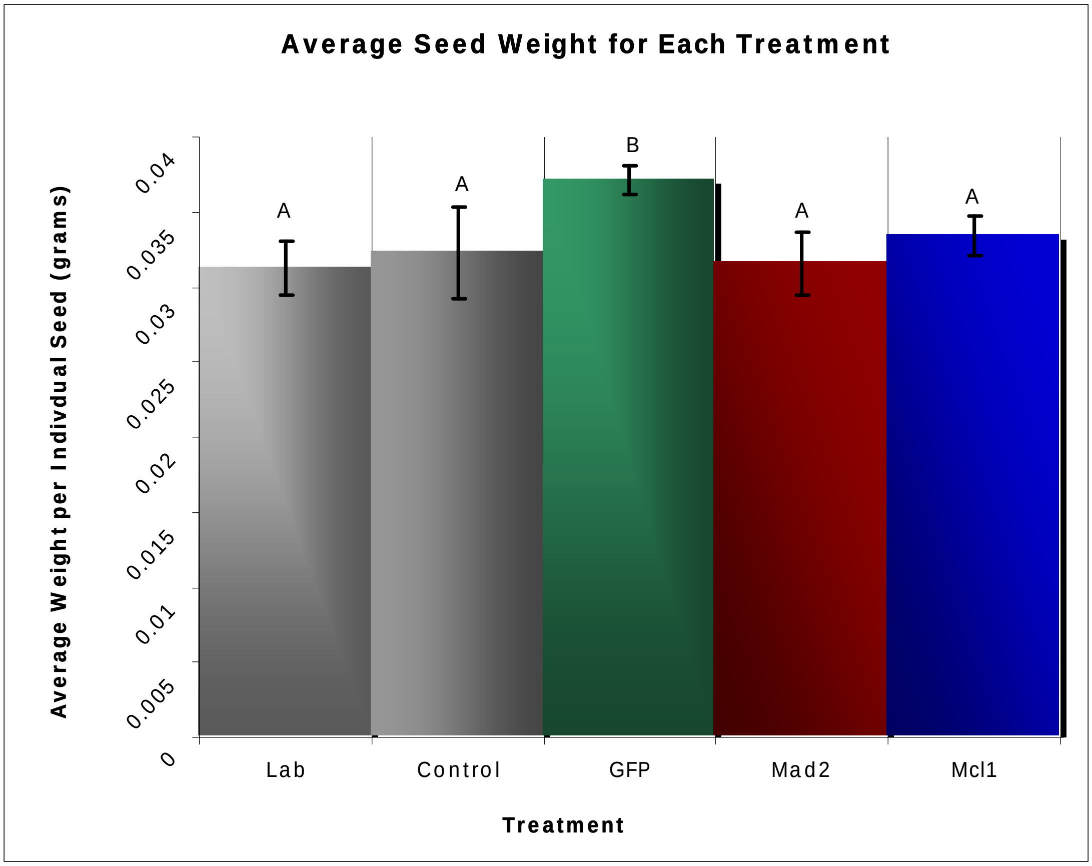


Figure 4.2. **Average Seed Weight for Each Treatment.** Treatment of winter wheat seed with *M. anisopliae* conferred an increase in mass (measured as average weight of each seed). Seeds treated with *M. anisopliae* (GFP) showed significant increase (f=157.82, df=4, p<0.0001) in average seed weight when compared to the control gum-arabic treated seeds (Control), original seeds planted on the field (Lab), the *M. anisopliae* Δ*Mad2* treated seeds (Mad2), or the *M. anisopliae* Δ*Mcl1* treated seeds (Mcl1). The average seed mass for a *M. anisopliae* treated seeds was 0.037 (± 0.001) grams/seed. The average seeds mass for the original seeds planted on the field (lab) was 0.031 (± .002) grams/seed. The average seeds mass for control gum-arabic treated seeds (control) was 0.032 (± .001) grams/seed. The average seeds mass for *M. anisopliae* Δ*Mad2* treated seeds (Mad2) was 0.032 (± .0019) grams/seed. The average seeds mass for *M. anisopliae* Δ*Mcl1* treated seeds (Mcl1) was 0.033 (± .0013) grams/seed.

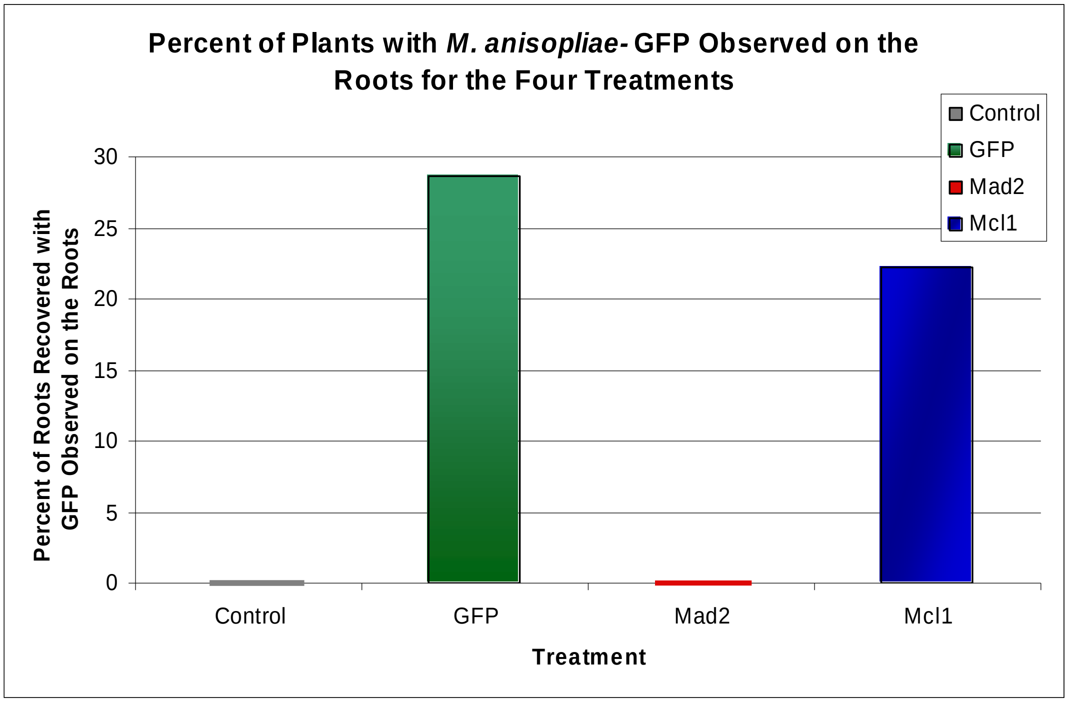
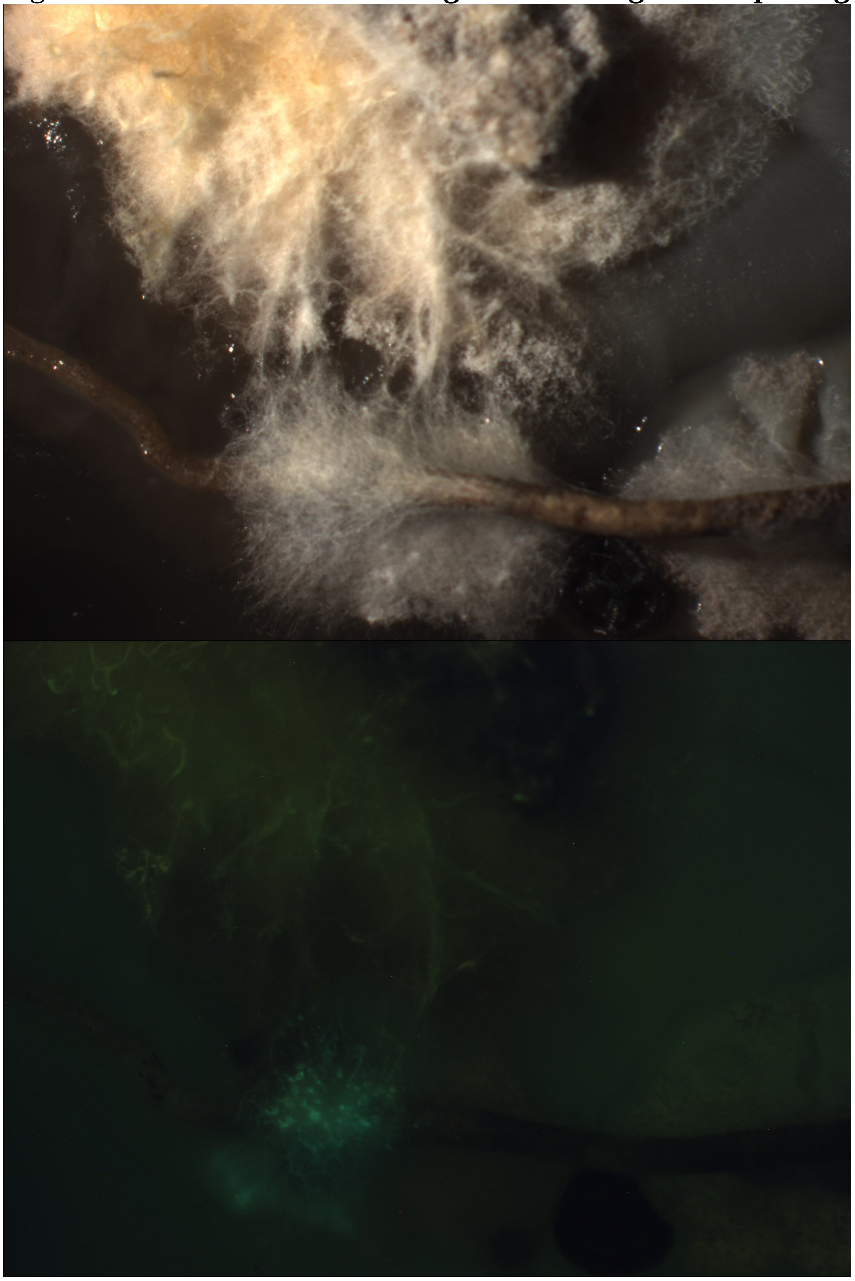


Figure 4.3 **Percent of Plants with *M. anisopliae*-GFP observed on the Roots for the Four Treatments.** Plants were recovered from the field and all loose dirt was removed from the plant. Roots were then examined under a UV dissecting scope and the presence of GFP was examined. No plants recovered from the control gum-arabic treated seeds (control) or *M. anisopliae* Δ*Mad2* treated seeds (Mad2) fluoresced under a UV dissecting scope, indicating that the GFP labeled *M. anisopliae* was not present on the roots. Samples from *M. anisopliae* treated seeds showed 28.6% of the plants recovered from the field had the GFP-labeled *M. anisopliae* present on the roots. Samples from *M. anisopliae* Δ*Mcl1* treated seeds (Mcl1) showed 22.2% of the plants recovered from the field had the GFP-labeled *M. anisopliae* present on the roots.

Figure 4.4 **Roots under normal light and UV light after plating on PDA plates.**



Two photographs showing the root under natural light and under UV light. *M. anisopliae*-GFP was observed, under 40X magnification. *Gliocladium*, *Penicillium*, *Paecilomyces*, *Cladosporium*, and *Alternaria* species were also observed on the root once the root was plated on PDA.

Figure 4.5 **Roots under normal light and UV light after plating on PDA plates.**

Two photographs showing the root under natural light and under UV light for another section of root taken under 40X magnification. *M. anisopliae*-GFP was observed, but with less intensity than samples freshly recovered from the field. *Gliocladium*, *Penicillium*, *Paecilomyces*, *Cladosporium*, and *Alternaria* species were also observed on the root once the root was plated on PDA.

Examination of Metarhizium as a potential biofertilizer in microcosms

While germination rates were not altered by the application of *M. anisopliae*, its presence accelerated initial growth rates. Seeds growing in the presence of *M. anisopliae* and *M. anisopliae* Δ*Mcl1* had significantly faster growth rates than uninoculated control seeds or seeds treated *M. anisopliae* Δ*Mad2* (Figure 4.6 and Figure 4.7). The growth rate was calculated for the first 10 days. After 14 days the growth rates for all conditions began to plateau and leaf blades from all microcosms began to turn brown. The growth rate of *M. anisopliae*-GFP treated seeds [1.61 (±0.16) cm/day] was significantly greater (t=2.4338, p=.0410) than that of the control seeds [1.15 (±0.11) cm/day]. The growth rate of *M. anisopliae*-GFP Δ*Mcl1* treated seeds [1.55 (±0.12) cm/day] was also significantly greater than that of the control seeds (t=2.4641, p=0.0391). The growth rate *M. anisopliae*-GFP Δ*Mad2* [1.31 (±0.12) cm/day] treated seeds were not significantly different (t=.9768, p=0.3573) than the control seeds.

The experimental protocol was repeated for 6 microcosms: 1) control which had no treatment of seed and no soil phosphate, 2) control untreated seed grown in soil with insoluble phosphate, 3) control untreated seed grown in soil with soluble phosphate, 4) *Metarhizium* treated seed and no soil treatment, 5) *Metarhizium* treated seed grown in soil with insoluble phosphate and 6) *Metarhizium* treated seed grown in soil with soluble phosphate. For each treatment blade length was measure over time (Figure 4.8). ANOVA analysis was used to calculate significant differences in blade length growth over time (Table 4.1). For days 3 and 15, there were no significant differences in blade length (f=0.04, p=0.9992) on day 3 (f=2.84,

$p=0.0179$) or on day 15. On days 11 and 13, blade length was significantly ($p<0.05$) higher in the three microcosms containing *Metarhizium* treated seeds and the microcosm containing soluble phosphate and untreated seeds ($f=7.60$, $p<0.001$ for day 11) ($f=10.55$, $p<0.001$ for day 13). Growth rates were calculated for these 6 microcosms at day 13 (Figure 4.9).

After blade length was measured at day 15, plants were carefully removed from their microcosms and cleaned. They were then allowed to air dry. Plant mass was measured (Figure 4.10). SAS was used to perform an ANOVA test. Data were analyzed using proc mixed protocol and repeated measures generating an autocorrelation variation matrix to adjust for repeated measure correlation between the 6 microcosms growth rate. Tukey's Studentized Range (HSD) Test was used to compare data sets for the growth rates for the six microcosms. Average biomass was calculated for each microcosm (Table 4.2).

Samples grown on the plates containing insoluble salts were observed for clearing zones. Preliminary work with plates inoculated with *M. anisopliae*, *M. anisopliae* Δ*Mcl1* and *M. anisopliae* Δ*Mad2* showed clearing zones of 1.06 ± 0.15 mm, 1.00 ± 0.11 mm and 1.06 ± 0.12 mm respectively. Analyzing the data with unpaired t tests showed that the clearing zones for *M. anisopliae* and *M. anisopliae* Δ*Mcl1* were not significantly different ($t=0.5005$, $p= 0.6228$). When clearing zones for *M. anisopliae* and *M. anisopliae* Δ*Mad2* were compared, they were found to be not significantly different ($t= 0.5169$, $p=0.6615$). Clearing zones for *M. anisopliae* ΔM*cl1* and *M. anisopliae* Δ*Mad2* were also not significantly different ($t= 0.8910$, $p= 0.3847$). These fungal strains appear to behave in similar fashions on these plates.

The experiment was repeated using *A. niger* as the positive control (Figure 4.11), the final media pH was 5.3 ± 0.56 and the clearing zone was 11.5 ± 0.34 mm. The initial media pH was 7.2 (Omar, 1998). Plates with *M. anisopliae* had final media pH of 6.4 ± 0.55 and the clearing zone was 1.05 ± 0.16 mm. The *M. anisopliae* mutant that over produced oxalic acid had a final media pH of 5.7 ± 0.11 and the clearing zone was 2.35 ± 0.17 mm. The *M. anisopliae* mutant that under produced oxalic acid had a final media pH was 6.7 ± 0.11 and the clearing zone was 0.15 ± 0.11 mm. There was a strong correlation between pH and clearing zone, which was observed when the log (clearing zone) was plotted against the pH. SAS was used to generate Pearson Correlation Coefficients of -0.90139.

ANOVA analysis of repeated measures generated from SAS indicated that the 4 fungal strains clearing zones were significantly differently from each other ($p<0.05$). The results were from Tukey's Studentized Range (HSD) Test was used to compare data sets and n, f and p value were generated ($f=606.61$, $df=36$, $p<.0001$) .

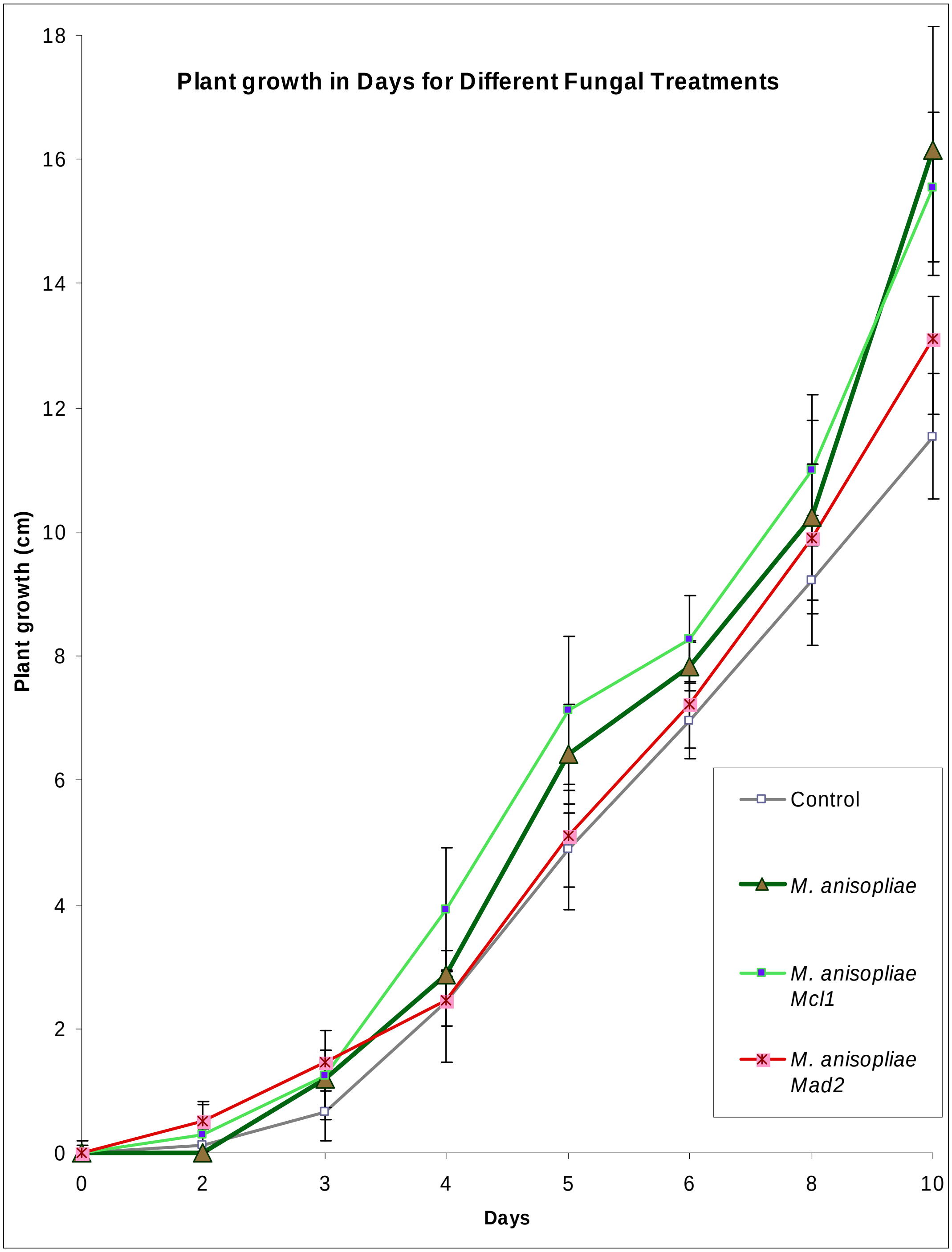


Figure 4.6 **Plant Growth over 10 days for *M. anisopliae* Treatments.** Four microcosms were examined: control untreated seeds, seeds that were treated with conidia from *Metarhizium*, *Metarhizium* Δ*Mad2* and *Metarhizium* Δ*Mcl1*. Germination rates for all conditions were the same and after 14 samples from the four microcosms showed no significant difference in blade length or growth rate.

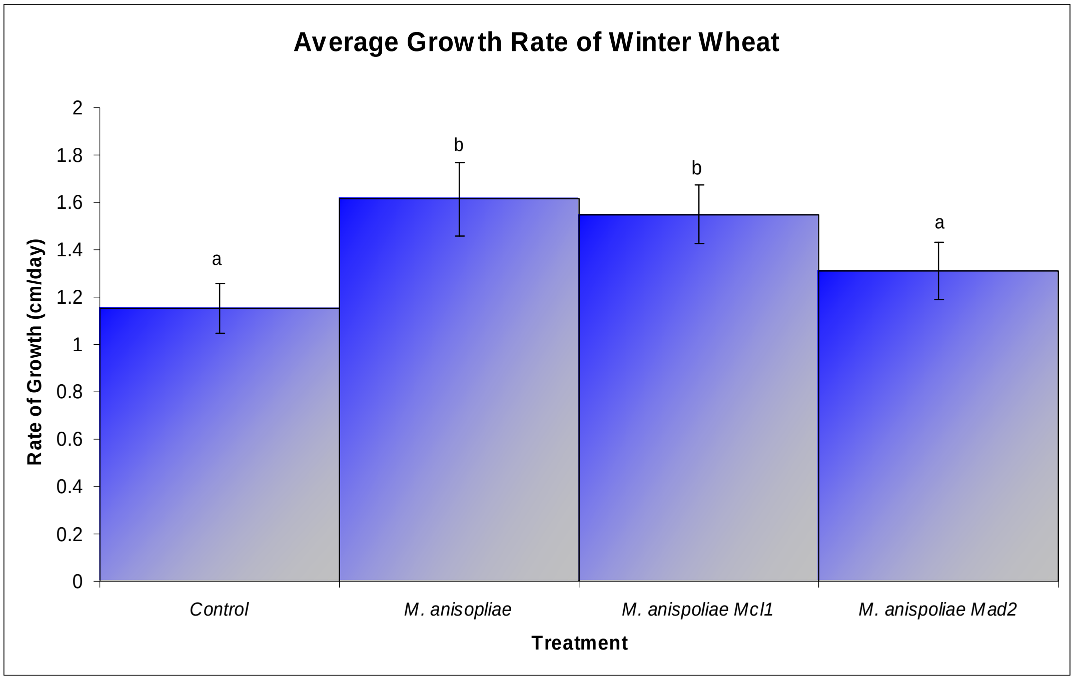


Figure 4.7. **Average Growth Rate of Winter Wheat after 10 days**. Growth rates after 10 days were compared for the various conditions, and the presence of fungal conidia increased growth rates when compared to the control, untreated seeds. Seeds that were treated with *Metarhizium* and *Metarhizium* Δ*Mcl1* prior to planting in the microcosm had significantly ($p<0.05$) faster growth (a) rates than the control and seeds treated with *Metarhizium* Δ*Mad2*(b).

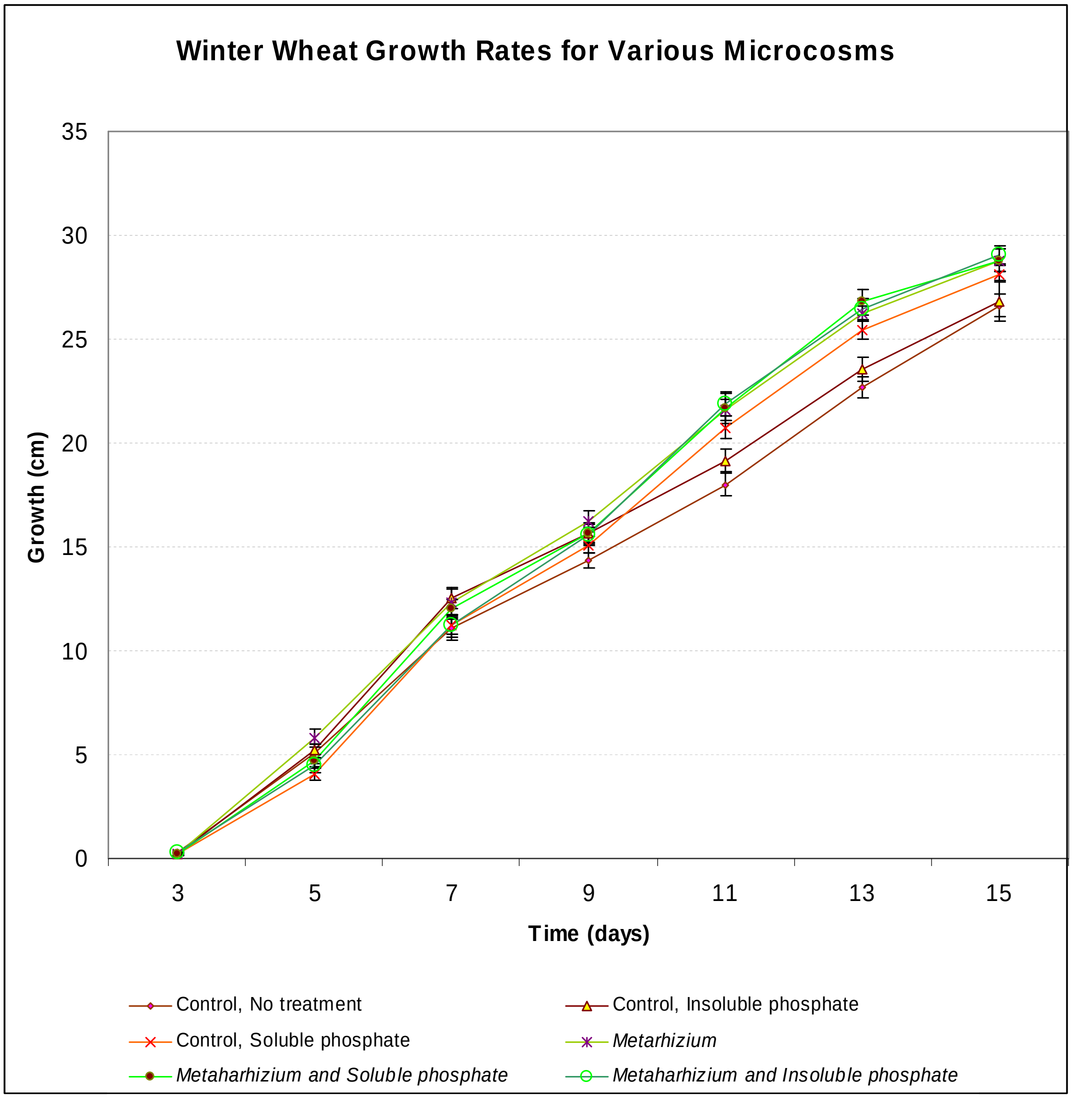


Figure 4.8 **Growth Over 15 Days for Various Microcosms.** Length of winter wheat blades were measured every two and average growth rate was calculated from the linear portion of the graph. At day 3 and day 15, the growth rates for all microcosms were not significantly different. Limited space for roots to grow and develop and limited nutrients may have contributed to the stunted growth after day 15. Leaves in all microcosms began to brown on the edge by day 15. On days 11 and 13, total blade lengths for plants in the following microcosm: *Metarhizium* only, *Metarhizium* and soluble phosphate, *Metarhizium* and insoluble phosphate and the control with soluble phosphate were significantly ($p<0.05$) larger than the blades of grass from the control microcosm.

Days	**3**	**5**	**7**	**9**	**11**	**13**	**15**
Control, no treatment	0.26 ± 0.09 A	5.05 ± 0.49 AB	11.07 ± 0.58 A	14.35 ± 0.35 B	17.98 ± 0.55 C	22.67 ± 0.49 C	26.6 3 ± 0.56 A
Control, Insoluble phosphate	0.23 ± 0.08 A	5.19 ± 0.35 AB	12.53 ± 0.50 A	15.65 ± 0.48 AB	19.16 ± 0.55 BC	23.53 ± 0.58 BC	26.84 ± 0.97 A
Control, Soluble phosphate	0.23 ± 0.06 A	4.08 ± 0.31 B	11.20 ± 0.41 A	15.10 ± 0.40 AB	20.75 ± 0.55 AB	25.43 ± 0.45 AB	28.13 ± 0.40 A
Metarhizium	0.24 ± 0.05 A	5.80 ± 0.42A	12.35 ± 0.65 A	16.24 ± 0.51 A	21.60 ± 0.50 A	26.24 ± 0.35 A	28.80 ± 0.52 A
Metarhizium and Soluble phosphate	0.25 ± 0.08 A	4.69 ± 0.35 AB	12.02 ± 0.47 A	15.63 ±0.56 AB	21.67 ± 0.69 A	26.78 ± 0.65 A	28.78 ± 0.55 A
Metarhizium and Insoluble phosphate	0.26 ± 0.06 A	4.50 ± 0.35 AB	11.22 ± 0.54 A	15.56 ± 0.37 AB	21.89 ± 0.56 A	26.47 ± 0.52 A	29.07 ± 0.43 A
Mean growth (cm)	0.24	4.88	11.74	15.43	20.49	25.14	28.02
f,df,p	0.04, 5, 0.9992	2.61, 5, 0.0275	1.54, 5, 0.1798	1.90, 5, 0.0979	7.60, 5, <.0001	10.55, 5, <.0001	2.84, 5, 0.0179

Table 4.1 **Tukey's Studentized Range (HSD) Test for Growth**. Total blade length was measure for winter wheat in 6 different microcosms at 2 day intervals beginning when germination was observed on day 3. Total blade length was recorded in cm. Significant differences ($p<0.05$) in blade length were not observed until day 5. By day 15 blade length for all microcosms was not significantly different.

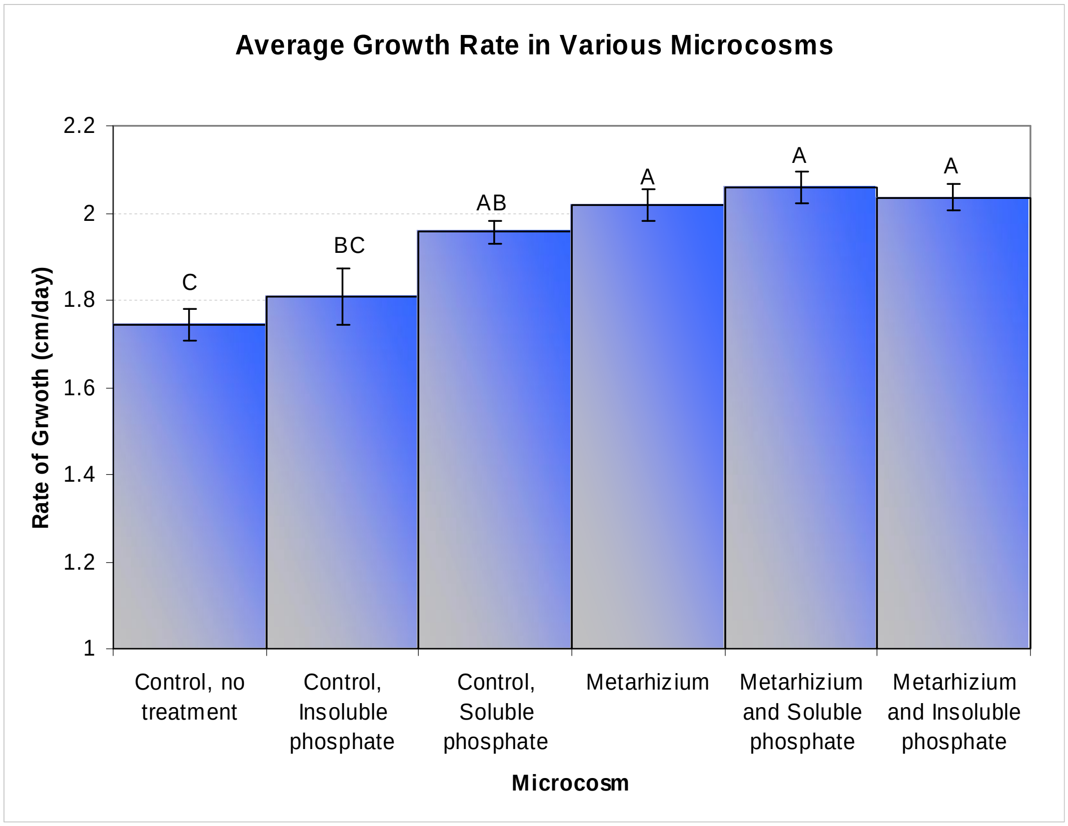


Figure 4.9 **Average Growth Rate for the Six Microcosms.** Growth rates after 13 days were compared for the various conditions. The presence of fungal conidia significantly ($p<0.05$) increased growth rates (A) when compared to the control, untreated seeds (C). Seeds that were treated with *Metarhizium* and also had significantly faster growth (A) rates than the control untreated seeds in soil with no addition phosphate (BC). Untreated seeds gown in soil with soluble phosphate did not have significantly differences in growth rates (AB when compared to the three microcosms containing *Metarhizium* treated seeds. Untreated seeds grown with soluble phosphate added to the soil had significantly different growth rates (AB) than the control untreated seeds grown in unamended soil (C).

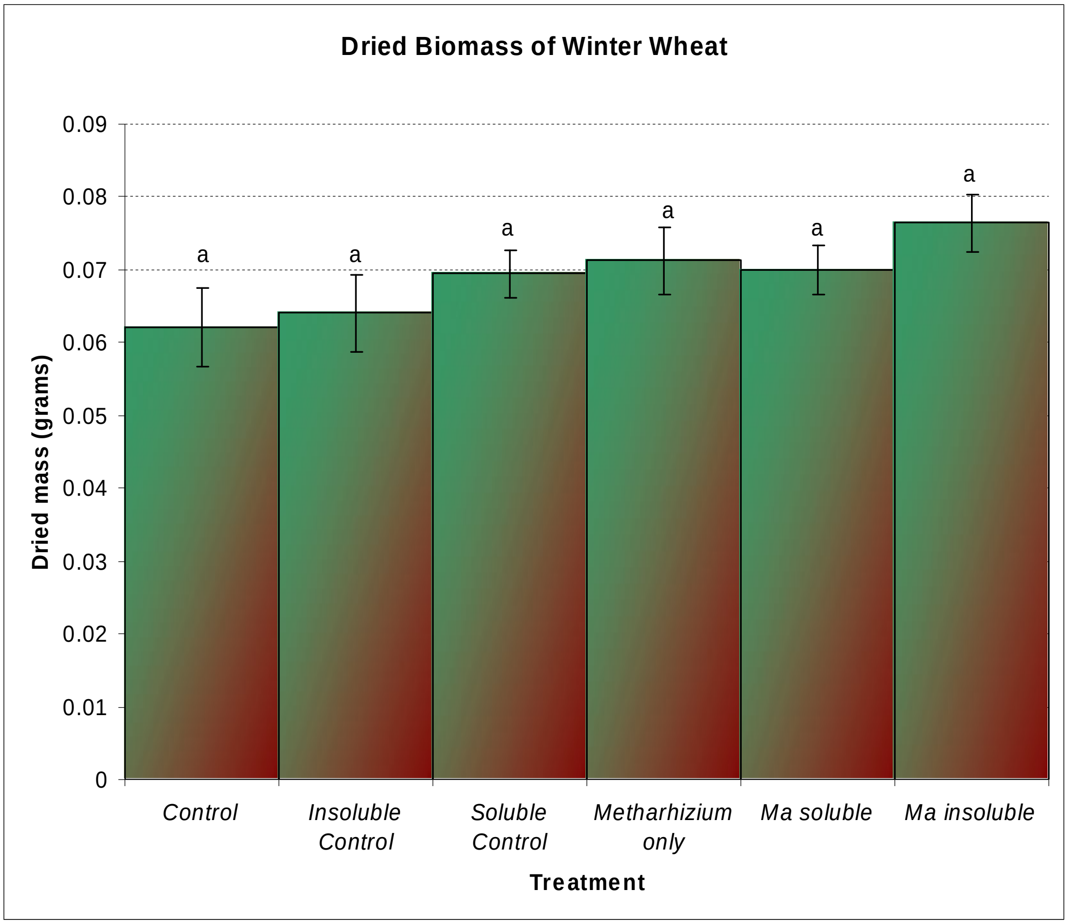


Figure 4.10 **Dried Biomass of Winter Wheat Plants Recovered from Six Microcosms.** Intact winter wheat plants were removed from their microcosm and loose soil as shaken off. Plants were then rinsed with distilled water to remove remaining soil. After the plants were dried for 48 hours. Average plant weight was measured in grams. Samples from the microcosm were not shown to have significantly ($p<0.05$) different masses.

Treatment	**Mean Biomass(g)**
Control, no treatment	0.062 (± 0.0055) A
Control, Insoluble phosphate	0.064 (± 0.0053) A
Control, Soluble phosphate	0.069 (± 0.0033) A
Metarhizium	0.071 (± 0.0046) A
Metarhizium and Soluble phosphate	0.070 (± 0.0033) A
Metarhizium and Insoluble phosphate	0.076 (± 0.0040) A

Table 4.2 **Mean Biomass from Samples Recovered from the Six Microcosms.** Tukey's Studentized Range (HSD) Test for weight for samples compared to the control were f=1.28, df=96, p=0.2800. No significant differences ($p<0.05$) were found in plant biomass.

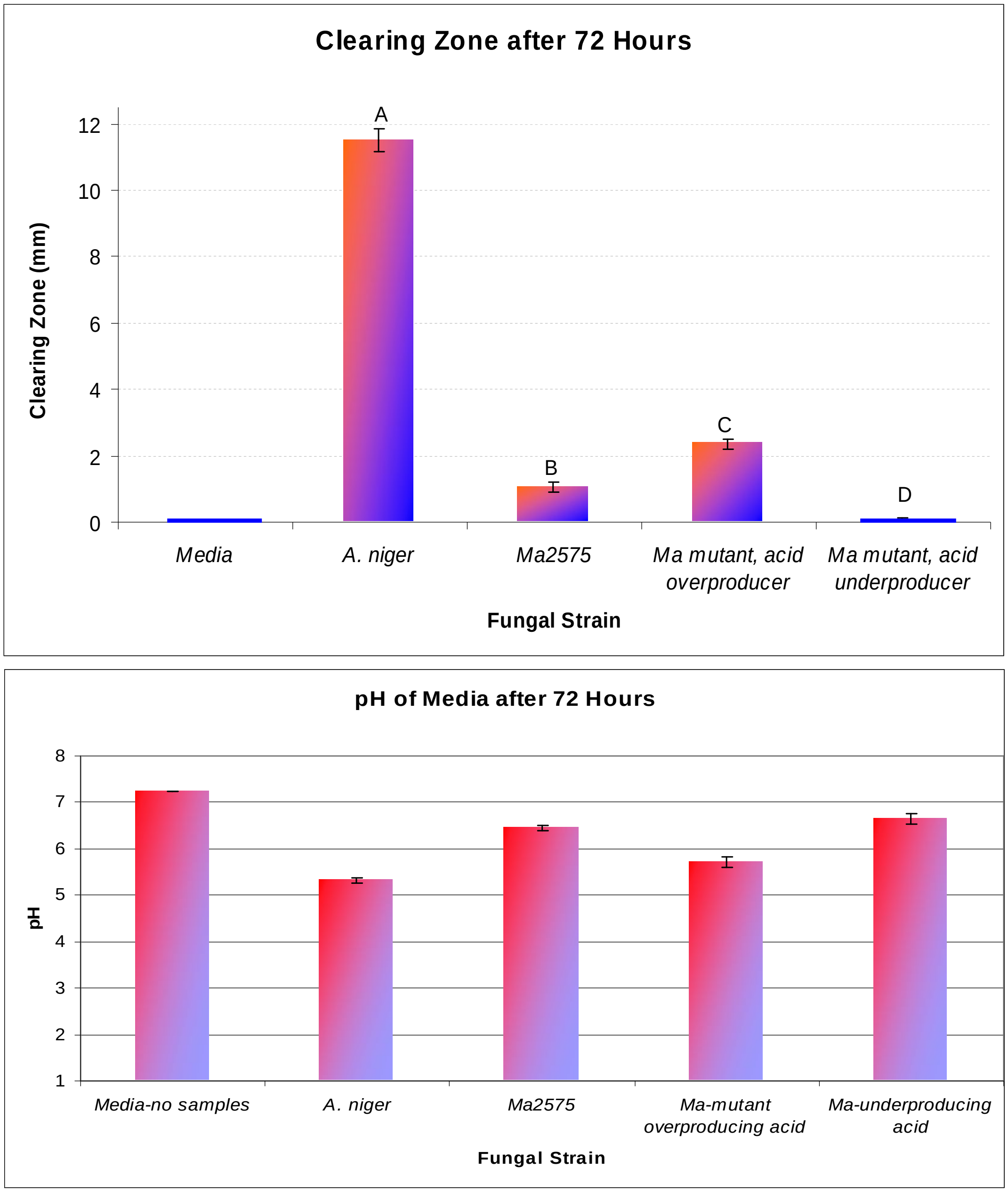


Figure 4.11 **Clearing Zones on Plates with Agar-Based Media and Final Media pH.** Fungal strains were grown on media containing insoluble phosphate. *A. niger* was used as a positive control. Acidic by-products released from the fungi caused the pH of the media to decrease from 7.24 and increased the clearing zone. The relationship is not proportional and it is possible other compounds released by the fungus contribute to the size of the clearing zone. Two *Metarhizium* mutants were used to study the effect of acid production on clearing zone and it was found that the mutant that over produced acid had a larger clearing zone than the mutant that under produced acid. The clearing zones for the 4 fungal strain examined were all significantly ($p<0.05$) different f=606.61, df=36, $p<0.0001$.

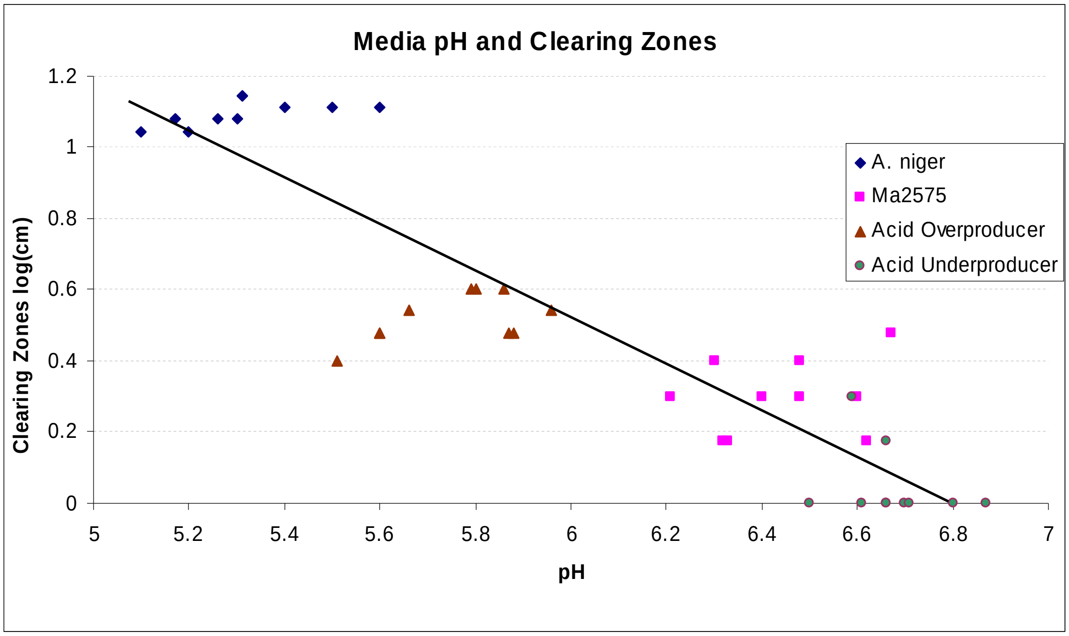


Figure 4.12 **Correlation Between pH and log(Clearing Zone)**. The Pearson Correlation Coefficients was -0.90139. There was a strong correlation between pH and clearing zone. As pH decreased and the media was acidified, insoluble calcium phosphate because soluble and accessible to the fungus, creating a visible clearing zone in the opaque media.

Discussion

Many genotypes of *M. anisopliae* are abundant in grassland soils, and different genotypes are specialized to forest soils (Bidochka 2001, Bidochka *et al*., 2001). The impact of rhizosphere competence by *M. anisopliae* on plant ecology in general could be considerable with implicit co-evolutionary implications. Rhizospheric competence may need to be considered as a feature for selecting fungal strains for biocontrol and this also raises the possibility of managing the rhizosphere microflora to achieve optimal insect control. This would dovetail with attempts in integrated pest management to manipulate the environment of the plant and insect to enhance insect biocontrol (Roberts and Hajek, 1992). In this study we show for the first time that spores of *M. anisopliae* applied to seeds of winter wheat will colonize the root system. It was also demonstrated that the treatment conferred a 14.9% increase in seed weight. This was much greater than we anticipated and would clearly have a profound impact if broadly applicable. If our results were due to insect control then the range of insects present, levels of infestation and soil conditions would likely affect the magnitude of growth promotion. This would need to be established by repeating these experiments under different conditions, soil types and geographical regions.

It should be emphasized that the *M. anisopliae* strain chosen for these studies was not selected by screening multiple strains for rhizospheric competence. It is not known if strain 2575 is a particularly competent strain or if selection could provide a much better colonizer. Presumably, if a good root colonizer were chosen, one that is capable of being transported by the root through the soil profile, then seed treatment would be the most attractive method for introducing the root colonizer into the soil-plant environment where the root colonizer may have the opportunity to be the first colonizer of roots. The seed has already proved an important delivery vehicle for a variety of beneficial microbes for plant growth enhancement and biological disease control, and there is potential of seed-applying multiple biocontrol agents with different disease-suppressive mechanisms (reviewed in Barea *et al.,* 2005).

Species of clavicipitaceous endophytes are already used commercially in turf grass seeds in this manner and our study suggests that *M. anisopliae* may usefully join them. Treating corn seeds with conidia of *M. anisopliae* resulted in a significant increase in plant stand density and yield (Kabaluk and Ericsson, 2007). Only recently has it become evident how poorly the complexities of plant-*Metarhizium* interactions are understood. The inability of the rhizosphere competent, but non-entomopathogenic strain *M. anisopliae* Δ*Mcl1* to significantly promote growth indicates that the efficacy of *M. anisopliae* results largely from suppression of insect pests, at least under the particular field conditions we tested. This presumably results from direct pathogenicity. *M. anisopliae* strain 2575 has a wide host range that includes beetles, lepidopterans and hemipterans. Preplant observations revealed wireworms and various scarab larvae that could be targeting the winter wheat. These would be susceptible to strain 2575, but a direct correlation between treatment effects and insect levels was not established. It is likely that greater effects than those observed could be obtained by screening wild type strains or engineering a strain more virulent to the principal pests that were limiting plant growth.

An insect’s greatest defense mechanism may be avoidance of entomopathogenic fungi (Rath, 2000), and *M. anisopliae* is repellent to many insect species including

Japanese beetle (*Popillia japonica*) in turf grass (Villani *et al.*, 1994). Thus, *M. anisopliae* in the rhizosphere could provide a repellent barrier around roots that would offer effective protection to the plant from root-feeding insect pests, as there is an inevitable time lag following fungal infection and cessation of feeding. The nature of fungal repellency has not been determined, but as *M. anisopliae* Δ*Mcl1* is deficient in a gene that is only expressed in the host hemolymph it is not likely that it would be less repellant than the wild type GFP-2575, and yet MCL1 provided much smaller benefit than the virulent GFP-2575. Therefore, insect repellency could be a useful additional criterion for screening natural strains for inclusion as seed treatments.

The development of appropriate combinations of seed treatments that included insect pathogens would obviously provide a higher level of plant protection and constitutes a very promising research area. However, many soil microbes including rhizospheric *Trichoderma* promote plant growth by a variety of means (Harman and Dozalli 2001; Harman and Shoresh, 2007), so future studies should also focus on the impact of *M. anisopliae* on plant physiology, including stress tolerance, uptake of nutrients, and other biological factors not necessarily related to pest control. The ability of *M. anisopliae* to accelerate growth of seedlings in insect free microcosms is noteworthy as indicative of a possible role for them as biofertilizers. *M. anisopliae* was also capable of producing a ~1 mm clearing zone in media containing insoluble phosphate. Such a clearing zone is indicative that this fungus is capable of solubilizing phosphate (Omar, 1998). The ability of an entomopathogenic rhizospheric fungus like *M. anisopliae* to solubilize phosphate could provide another added dimension to the benefits *M. anisopliae* provides to plant growth, mass and ultimately to yield.

Chapter 5: Conclusions

The research presented here examines the potential uses of *Metarhizium* in agriculture. Understanding how *Metarhizium* behaves outside a suitable insect host, as a saprophyte at the rhizosphere, will allow *Metarhizium* to be optimized for commercial use. The field trials we conducted focused on the symbiotic relationship between *Metarhizium* and the grasses tall fescue and winter wheat. The field trials also gave important insight as to how an introduced strain can persist in the environment and how an introduced strain behaves compared to native strains. It is important to note that CFUs of the introduced fungus were still being recovered from the field four years after its initial application. The question remains how long an introduced strain can persist until no significant titers can be recovered? It is possible that an introduced strain could naturalize to an introduced environment and remain indefinitely. This may be beneficial in terms of long-term insect control.

But there are many environmental and economic reasons why researchers and industry would not seek to permanently establish an engineered microbial agent in the environment. Rhizospheric competence may increase the difficulty of eliminating the pathogen following unexpected environmental effects. There is a possibility that an entomopathogen, such as *Metarhizium,* applied to fields could contaminate neighboring fields (St. Leger and Screen, 2001). Issues of biological safety must be addressed prior to releasing a transgenic entomopathogen into the environment (Barea *et al.,* 2005). In this circumstance, it is essential to understand how long an introduced strain may persist, its behavior in the field, and its genetic stability. A key advantage of microbial control agents is their potential to replicate and persist in the environment providing long-term control. For certain situations, we may want a strain to persist in the environment long enough to kill insect pests, but short enough not to survive more than one season. With this in mind, *Metarhizium* strains with suicide genes, or a knockout mutant such as Δ*Mad1* may be used to reduce the long-term survivorship of *Metarhizium* in the field.

In a turf environment, application of a conidial suspension could be used to establish fungal colonies. Application of conidia directly to wheat seeds was also effective in establishing populations at the rhizosphere. The research presented here demonstrated that both methods of application could be used to establish *Metarhizium* colonies at the rhizosphere. Follow-up experiments could be performed to determine the best application method and concentration of *Metarhizium* conidia to apply. This may be dependent on the type of plant, how quickly the plant germinates or if the plant is already established, time of year, time of day, weather patterns and soil conditions.

To dissect the complex relationship between the plant and *Metarhizium* requires research into how the fungus behaves and also on the physiological interactions between the fungus and the plant. Certain grasses, such as tall fescue, decrease their photosynthetic rates during the hot, dry summer months found in the region of Maryland where these experiments were conducted. Prima facie, reduced photosynthetic rates would result in fewer carbon sources for *M. anisopliae* and fewer herbivorous insects for *Metarhizium* to infect. *Metarhizium* populations consistently showed reduced populations in the summer months through the four years of the field trial. Since there is the possibility that there is a relationship between how a plant responds to environmental cues and fungal populations, by researching a plant's response to changes in the environment, information about maintaining fungal populations may be elucidated. Understanding the complex relationship *Metarhizium* has with its host plant may be just important as the relationship *Metarhizium* has with its insect host. Further experiments should be performed to examine the rhizospheric competence of different plant species with different *Metarhizium* strains. It may be that there are specialized strains of *Metarhizium* specific for different plant species. Knowing which strain of *Metarhizium* is best suited for a specific plant may allow for customizing *Metarhizium* for specific crops.

Release of the knockout mutants Δ*Mcl1* and Δ*Mad2* were conducted to demonstrate whether survivability of *M. anisopliae* depended on *Metarhizium's* saprophytic competence or its ability to cycle through insect populations. Not surprisingly, eliminating either lifestyle option of *Metarhizium*, as an entomopathogen and as a saprophyte, reduced survival compared to the wild-type over four years. This research into the roles played by *Mad2* and *Mcl1* may also allow for customized strains to be created that do not persist in the environment for prolonged periods of time. This could be beneficial if an inundative approach were chosen for applying *Metarhizium* and it were undesirable for the fungus to persist after application. On the other hand, overexpressing these genes may allow them to persist longer at the rhizosphere when applied to field conditions. Other genes besides MCL1 and MAD2 should be examined in future research. Suitable candidates include genes involved in stress response and cell wall proteins that had altered expression patterns in isolates recovered from the field. A better understanding of the role these genes play may provide insight into the role selection plays on *Metarhizium's* saprophytic life and the stability of introduced strains. A key to achieve a successful, reproducible and safe from the risk management point of view, biological control lies in detailed knowledge of the properties of pathogens in their environments. Such knowledge would fulfill what the USDA (Biotechnology Risk Assessment Research Grants Program) Stakeholder Workshop's (2003) considered high priority issue on genetically based containment that would reduce rather than eliminate risk. The workshop proposed reducing the ability of the organism to spread through a lack of saprophytic competence. The current field trial demonstrated that *M. anisopliae* principally survives in field conditions as colonists of roots but high population levels required recycling in insects.

Many beneficial effects to crop growth and yield have been attributed to *Metarhizium* acting as an entomopathogen. But the increased winter wheat seed mass from plants recovered from seeds treated with *Metarhizium* conidia prior to planting may not be completely due to insect control. If *Metarhizium* were acting solely as an entomopathogen, winter wheat seeds treated with *Metarhizium* Δ*Mad2* and wild type *Metarhizium* prior to planting should have netted similar increases in seed mass for field recovered seeds. Yet *Metarhizium* Δ*Mad2* produced no increase in average seed mass, indicating that *Metarhizium* must be associated with the rhizosphere for plant protection by *Metarhizium* to occur. *Metarhizium* Δ*Mcl1* should have provided no benefit to the plant if the fungus were just acting as an entomopathogen. Yet there was a small (but not statistically significant) increase in seed mass from plants grown from

Metarhizium Δ*Mcl1* treated seeds. This indicated *Metarhizium* may also act to benefit the plant in other manners, possible acting as a biofertilizer.

Exactly how *Metarhizium* may be acting as a biofertilizer has only begun to be examined. The ability of an entomopathogenic rhizospheric fungus like *M. anisopliae* to solubilize phosphate could provide an added dimension to the benefits *M. anisopliae* provides to increasing plant growth rate, mass and ultimately yield. The research presented here demonstrated that *Metarhizium* can solubilize insoluble phosphate and increase plant growth rates in microcosms. One experiment demonstrated this could be due to acidic compounds released from by *Metarhizium*. There was a strong negative correlation between final media pH and clearing zone when different mutants of *Metarhizium* were examined. Such acidic byproducts could potentially be detrimental to some plant species, so further research could be done to identify and isolate chelating agents released by *Metarhizium* that may increase the availability of these insoluble nutrients. Phosphate is just one compound *Metarhizium* may help a to plant access. *Metarhizium* may be solubilizing other essential compounds in the soils, such as nitrogen. Future research can examine the range of compounds and mechanisms utilized by *Metarhizium* to increase nutrient availability. If the genes are identified that make *Metarhizium* a viable biofertilizer, it may be possible to overexpress these genes to maximize *Metarhizium's* potential as a biofertilizer. In addition to solubilizing compounds such as phosphate and nitrogen, fungal biofertilizers may use their mycelium to reach regions with more water and nutrients, essentially increasing the reach of a plant's root system. Experiments can be performed to examine the ability of *Metarhizium* to help plants survive various types of drought conditions.

My research also used microarrays to measure the events of microevolution for introduced fungal strains. While there were no specific genes that were altered in all the recovered samples, some trends were observed. Most changes in gene expression effected genes for stress response and cell wall proteins. No changes in the expression of genes involved in pathogenicity were observed. Understanding the genetic stability of an introduced biological control agent is essential before applying a biological control agent to the field so that a researcher would know that the fungus would not mutate rapidly and potentially change its host range. This is especially true if an extremely lethal specialist strain of *Metarhizium* were released for insect control.

Microarray based assays of mutation rates have not been performed with other fungal species, thus, it cannot be said that the changes we observed in gene expression are comparatively fast or slow by the standards of other fungi. A comprehensive effort to measure rates of microevolution in multiple fungal systems would allow the process to be standardized. With no fungal model systems to set a baseline of genetic stability, the changes in gene expression were quantified by measuring the number of genes with altered expression patterns. The USDA-ARS has begun a project to sequence the entire genome of *M. anisopliae* 2575, which is to be completed by September 2009. Once this project is complete, a comprehensive microarray slide of oligo-nucleotide sequences can be generated. At which point more thorough microarray experiments could be performed to define “normalcy” for evolution of changes in gene expression in *M. anisopliae.*

Future research and general conclusions

Hybridizing different strains of *Trichoderma* to optimize combinations of beneficial properties produced the commercially utilized strain of *Trichoderma* T22. It is therefore possible that different strains of *M. anisopliae* which have multiple roles in plant protection and growth could be used to create an optimal plant symbiont. It has been shown that some strains of *M. anisopliae* have antagonistic effects on plant pathogenic fungi, including *Fusarium oxysporum, Botrytis cinerea* and *Alternaria solani* (Kang *et al.*, 1996). Similarly, *B. bassiana* and several species of *Lecanicillium* have dual activities against insects and plant pathogens (Askary *et al.*, 1998; Kim *et al.*, 2001; Ownley *et al.*, 2004, 2008), emphasizing that further studies are needed to understand the true ecology of insect pathogenic fungi. Once the genes that confer the resistance to various insect and plant pathogens have been identified, they may be use to create a transgenic strain of *Metarhizium*. Ideally this hybrid strain of *Metarhizium* would also have the addition of genes for enzymes such as nitrogenase and phosphatase that would allow *Metarhizium* to fix nitrogen and solubilize phosphate respectively. Currently transgenic strains of *Metarhizium* exist that have scorpion toxin genes that increase the speed of kill of insect pests. It may be possible to create a hybrid strain of *Metarhizium* that offered protection to the host plant from insect pests with increased kill rates, protected against other potential pathogens and worked as a biofertilizer. Research should also be conducted to identify crop specific strains and interpret the range of variation for *Metarhizium anisopliae* strains in terms of rhizospheric competence. Using a *Metarhizium* strains with narrow insect host ranges would eliminate the danger to non-target insects and beneficial insects such as pollinators and natural predators. Employing such a strain of *Metarhizium* as part of an integrated pest management system could substantially reduce the use of chemical insecticides and fertilizers.

Appendices

Appendix A

Expressed Sequence Tags Examined using Microarray Analysis

UID	GenBank#	Func	Function	best match	source	description
1A1	AJ272682	1b	Carbohydrate metabolism	AAL52759	Brucella melitensis 16M	glyoxylate induced protein
1A3	AJ272685	8	Transposable Elements	CAA93759	Tolypocladium inflatum	putative transposase
1A5	AJ272688	1a	Aminoacid metabolism	P14010	Emericella nidulans	4-aminobutyrate aminotransferase (GABA transaminase)
1A7	AJ272691	4b	RNA modification	CAA20727	Schizosaccharomyces pombe	ATP-dependent, dead box, RNA helicase
1A9	AJ272697	7c	Secondary metabolites	AAC49319	Aspergillus parasiticus	versicolorin B synthase
1A11	AJ272704	3a	Ribosomal proteins	CAB11701	Schizosaccharomyces pombe	40S ribosomal protein S10
2A1	AJ272847	9	Hyp/Unknown Prot	CAB10133	Schizosaccharomyces pombe	hypothetical protein SPAC29A4.06c
2A3	AJ272851	4b	RNA modification	AAC49909	Schizosaccharomyces pombe	putative splicing factor; Srp1
2A5	AJ272854	9	Hyp/Unknown Prot			unknown
2A7	AJ272857	1a	Aminoacid metabolism	Q9P6I2	Schizosaccharomyces pombe	Glutamate carboxypeptidase-like protein
2A9	AJ272863	3c	Proteolysis	CAB63912	Metarhizium anisopliae var. anisopliae	subtilisin-like protease; Pr1G
2A11	AJ272868	1b	Carbohydrate metabolism	AAH53853	Homo sapiens	PHYHD1 protein: Phytanoyl-CoA dioxygenase
3A1	AJ273039	4c	Transcription	T38818	Schizosaccharomyces pombe	transcription factor btf3 homolog
3A3	AJ273043	3d	Prot modification/targeting	BAA87295	Schizosaccharomyces pombe	Fkbp-type peptidyl prolyl cis-trans isomerase
3A5	AJ273046	9	Hyp/Unknown Prot			unknown
3A7	AJ273051	3c	Proteolysis	AAB84057	Hypocrea jecorina	proteasome regulatory subunit 12
3A9	AJ273056	4c	Transcription	CAB11044	Schizosaccharomyces pombe	probable transcription initiation factor TFIIB subunit
3A11	AJ273058	9	Hyp/Unknown Prot			unknown
SSC1						
4A3	AJ273220	9	Hyp/Unknown Prot			Unknown
4A5	AJ273224	9	Hyp/Unknown Prot			unknown
4A7	AJ273226	5d	Transport proteins	CAB88550	Neurospora crassa	related to carboxylic acid transport protein JEN1
4A9	AJ273230	9	Hyp/Unknown Prot	EAA76527	Gibberella zeae PH-1	hypothetical protein FG09635
4A11	AJ273233	9	Hyp/Unknown Prot			unknown
5A1	AJ273410	9	Hyp/Unknown Prot			unknown
5A3	AJ273412	3c	Proteolysis	CAB64346	Metarhizium anisopliae var. anisopliae	subtilisin-like protease Pr1I

5A5	AJ273415	5e	Cellular transportation	CAA91078	Schizosaccharomyces pombe	probable coiled coil protein
5A7	AJ273420	5a	Cell wall	CAC18170	Neurospora crassa	related to b-1, 3 exoglucanase precursor
5A9	AJ273422	9	Hyp/Unknown Prot			unknown
5A11	AJ273426	9	Hyp/Unknown Prot			unknown
1A2	AJ272684	2	Energy Metabolism	CAA19282	Schizosaccharomyces pombe	SPBC4B4.02c: involved in control of mitochondrial synthesis of Atp6p and Atp8p (ATP synthase subunit 6 and 8)
1A4	AJ272687	2	Energy Metabolism	BAB11407	Arabidopsis thaliana	glycolate oxidase subunit D-like; D-lactate dehydrogenase-like
1A6	AJ272689	9	Hyp/Unknown Prot			unknown
1A8	AJ272694	9	Hyp/Unknown Prot	EAA69263	Gibberella zeae PH-1	hypothetical protein FG00602
1A10	AJ272701	1b	Carbohydrate metabolism	T40935	Schizosaccharomyces pombe	probable UTP-glucose-1-phosphate uridylyltransferase
1A12	AJ272706	3a	Ribosomal proteins	NP_011223	Saccharomyces cerevisiae	ribosomal protein L2A (L5A) (rp8)
2A2	AJ272848	1b	Carbohydrate metabolism	BAA09656	Aspergillus oryzae	tannase
2A4	AJ272852	6c	DNA synthesis	JC6519	Emericella nidulans	DNA topoisomerase I
SSC2						
2A8	AJ272861	3c	Proteolysis	CAB89873	Metarhizium anisopliae var. anisopliae	subtilisin-like protease; Pr1D
2A10	AJ272866	1e	N-, P-, S-metabolism	NP_000447	Homo sapiens	sulfite oxidase
2A12	AJ272869	9	Hyp/Unknown Prot	EAA75284	Gibberella zeae PH-1	hypothetical protein FG05467
3A2	AJ273042	1f	Cofactor and vitamins	AAC59645	Gallus gallus	vitamin D3 hydroxylase associated protein
3A4	AJ273044	3c	Proteolysis	CAB46670	Schizosaccharomyces pombe	19S proteasome regulatory subunit; essential (PMID 12618370);
3A6	AJ273047	9	Hyp/Unknown Prot	EAA55399	Magnaporthe grisea	predicted protein
3A8	AJ273052	3c	Proteolysis	AAB18765	Caulobacter crescentus	ATP dependent protease
3A10	AJ273057	5a	Cell wall	AAF40139	Aspergillus fumigatus	b (1-3) glucanosyltransferase Gel2p
3A12	AJ273061	9	Hyp/Unknown Prot			unknown
4A2	AJ273219	4c	Transcription	AAF93178	Sclerotinia sclerotiorum	putative zinc finger protein Pac1
4A4	AJ273223	4b	RNA modification	CAB92703	Neurospora crassa	related to suppressor protein; SPT23
4A6	AJ273225	1a	Aminoacid metabolism	AAN31395	Aspergillus sojae	leucine aminopeptidase
4A8	AJ273228	9	Hyp/Unknown Prot	NP_595795	Schizosaccharomyces pombe	serine/threonine-protein kinase yeast ksp1 like
4A10	AJ273232	9	Hyp/Unknown Prot	EAA33875	Neurospora crassa	hypothetical protein
4A12	AJ273235	9	Hyp/Unknown Prot			unknown
5A2	AJ273411	1d	Nucleotide metabolism	CAC18138	Neurospora crassa	probable adenylate kinase
5A4	AJ273413	9	Hyp/Unknown Prot			unknown
5A6	AJ273417	9	Hyp/Unknown Prot			Unknown
5A8	AJ273421	1e	N-, P-, S-metabolism	CAD11393	Neurospora crassa	related to phosphatase 2a inhibitor
5A10	AJ273423	9	Hyp/Unknown Prot	S19420	Saccharomyces cerevisiae	hypothetical protein Ycr010cp
5A12	AJ273428	9	Hyp/Unknown Prot			unknown
1C1	AJ272733	9	Hyp/Unknown Prot	EAA70694	Gibberella zeae PH-1	hypothetical protein FG00748

1C3	AJ272736	3d	Prot modification/targeting	CAD70739	Neurospora crassa	probable clathrin-associated adaptor complex medium chain
1C5	AJ272740	1f	Cofactor and vitamins	P39954	Saccharomyces cerevisiae	putative S-adenosyl-L-homocysteine hydrolase; Sah1p
1C7	AJ272745	9	Hyp/Unknown Prot	EAA70045	Gibberella zeae PH-1	hypothetical protein FG10202
1C9	AJ272747	1b	Carbohydrate metabolism	CAC18158	Neurospora crassa	related to ER glucosidase I
1C11	AJ272749	9	Hyp/Unknown Prot			Unknown
2C1	AJ272899	1b	Carbohydrate metabolism	BAA03574	Pseudomonas aeruginosa	endo a-1,4 polygalactosaminidase precusor
2C3	AJ272904	6g	Cell death	AAR25995	Pyrus communis	putative senescence-associated protein
2C5	AJ272906	4b	RNA modification	CAA22814	Schizosaccharomyces pombe	nucleolar protein involved in pre-rRNA processing
2C7	AJ272908	9	Hyp/Unknown Prot			unknown
2C9	AJ272912	6a	Cell cycle	CAB52419	Pyrenopeziza brassicae	putative septin
2C11	AJ272914	3c	Proteolysis	BAB97387	Aspergillus oryzae	aorsin: a novel serine proteinase with trypsin-like specificity at acidic pH
3C1	AJ273079	1b	Carbohydrate metabolism	AAA34350	Candida albicans	a-glucosidase (maltase)
3C3	AJ273082	9	Hyp/Unknown Prot	EAA67473		hypothetical protein FG00676
3C5	AJ273085	9	Hyp/Unknown Prot			Unknown
3C7	AJ273087	4b	RNA modification	AAG52143	Arabidopsis thaliana	putative RNA helicase
3C9	AJ273093	9	Hyp/Unknown Prot	EAA75495	Gibberella zeae PH-1	hypothetical protein FG05259
3C11	AJ273103	1f	Cofactor and vitamins	NP_010332	Saccharomyces cerevisiae	uroporphyrinogen decarboxylase; Hem12p
4C1	AJ273263	6a	Cell cycle	AAB41233	Emericella nidulans	septin B
4C3	AJ273266	1e	N-, P-, S-metabolism	AAB18274	Emericella nidulans	sconCp (sulphur metabolite repression regulation)
4C5	AJ273270	5d	Transport proteins	NP_013993	Saccharomyces cerevisiae	Overexpression Rescues sro7/sop1 in NaCl. Encodes a membrane protein.; Ymr266wp
4C7	AJ273273	9	Hyp/Unknown Prot			unknown
4C9	AJ273275	6c	DNA synthesis	CAB75412	Schizosaccharomyces pombe	mis5 protein (genome replication, s-phase)
4C11	AJ273279	5a	Cell wall	CAB16200	Schizosaccharomyces pombe	probable glucan 1,3-b-glucosidase precursor
5C1	AJ273455	9	Hyp/Unknown Prot			unknown
5C3	AJ273457	3a	Ribosomal proteins	CAC28787	Neurospora crassa	probable ribosomal protein L12
5C5	AJ273459	9	Hyp/Unknown Prot			unknown
5C7	AJ273461	9	Hyp/Unknown Prot	EAA33203	Neurospora crassa	predicted protein
5C9	AJ273465	9	Hyp/Unknown Prot			Unknown
5C11	AJ273470	6b	Chromosome structure	CAA39153	Emericella nidulans	histone H2B
1C2	AJ272734	9	Hyp/Unknown Prot	EAA70547	Gibberella zeae PH-1	hypothetical protein FG02472
1C4	AJ272739	9	Hyp/Unknown Prot			Unknown
1C6	AJ272743	3c	Proteolysis	CAB44652	Metarhizium anisopliae	Trypsin-related protease
1C8	AJ272746	9	Hyp/Unknown Prot	EAA64348	Aspergillus nidulans	hypothetical protein AN9016.2
1C10	AJ272748	1b	Carbohydrate metabolism	NP_596852	Schizosaccharomyces pombe	possibly involved in the degradation of external UDP-glucose by similarity

1C12	AJ272750	9	Hyp/Unknown Prot			unknown
2C2	AJ272901	2	Energy Metabolism	CAA20859	Schizosaccharomyces pombe	ubiquinol-cytochrome c reductase complex subunit
2C4	AJ272905	6f	Clock control/light response	AAC64287	Neurospora crassa	clock-controlled gene-6 protein
2C6	AJ272907	9	Hyp/Unknown Prot	BAB14465	Homo sapiens	hypothetical protein FLJ13150
2C8	AJ272909	5b	Cytosqueloton	CAC28678	Neurospora crassa	related to LAS1 protein (involved in cell morphogenesis, cytoskeletal regulation and bud formation)
2C10	AJ272913	9	Hyp/Unknown Prot			unknown
2C12	AJ272915	1c	Lipid metabilism	NP_035551	Mus musculus	sphingomyelin phosphodiesterase 1
3C2	AJ273081	3c	Proteolysis	CAB44651	Metarhizium anisopliae	Chymotrypsin
3C4	AJ273083	9	Hyp/Unknown Prot			unknown
3C6	AJ273086	9	Hyp/Unknown Prot			unknown
3C8	AJ273089	9	Hyp/Unknown Prot			unknown
3C10	AJ273102	4c	Transcription	XP_331132	Neurospora crassa	WHITE COLLAR 1 PROTEIN (WC1): Transcription regulation; Activator; DNA-binding
3C12	AJ273104	9	Hyp/Unknown Prot	EAA72447	Gibberella zeae PH-1	hypothetical protein FG08750
4C2	AJ273265	3d	Prot modification/targeting	T39856	Schizosaccharomyces pombe	probable chaperonin
4C4	AJ273268	5f	Signaling	AAB84002	Cochliobolus heterostrophus	GTPase activating protein homolog
4C6	AJ273272	9	Hyp/Unknown Prot	EAA52609	Magnaporthe grisea	hypothetical protein MG05301.4
4C8	AJ273274	9	Hyp/Unknown Prot	EAA77587	Gibberella zeae PH-1	hypothetical protein FG06651
4C10	AJ273278	3a	Ribosomal proteins	BAB32661	Sus scrofa	60S ribosomal protein L35
4C12	AJ273280	7b	Detoxification	AAF29805	Emericella nidulans	ABC-transporter
5C2	AJ273456	5f	Signaling	CAD70304	Neurospora crassa	related to 3-phosphoinositide dependent protein kinase-1 (PDK1)
5C4	AJ273458	8	Transposable Elements	CAA93759	Tolypocladium inflatum	putative transposase
5C6	AJ273460	5e	Cellular transportation	P35848	Neurospora crassa	mitochondrial import receptor subunit Tom20 (mitochondrial 20 kDa outer membrane protein)
5C8	AJ273462	4b	RNA modification	NP_014017	Saccharomyces cerevisiae	putative RNA-dependent helicase; Has1p
5C10	AJ273469	3b	Translation	S71745	Schizosaccharomyces pombe	translation initiation factor eIF-4A
5C12	AJ273472	7b	Detoxification	T40370	Schizosaccharomyces pombe	DNA-(apurinic or apyrimidinic site) lyase
1K1	AJ272773	5d	Transport proteins	P34054	Trichoderma harzianum	AMINO-ACID PERMEASE INDA1
1K3	AJ272777	9	Hyp/Unknown Prot	EAA65486	Aspergillus nidulans	hypothetical protein AN1303.2
1K5	AJ272779	9	Hyp/Unknown Prot			unknown
1K7	AJ272782	9	Hyp/Unknown Prot	EAA31092	Neurospora crassa	hypothetical protein
1K9	AJ272785	7b	Detoxification	P54006	Cochliobolus carbonum	ToxD protein, unique to isolates that make the cyclic peptide HC-toxin
1K11	AJ272788	5f	Signaling	BAB20820	Rosellinia necatrix	G protein a subunit
2K1	AJ272933	9	Hyp/Unknown Prot	T49762	Neurospora crassa	hypothetical protein B24B19.340
2K3	AJ272937	9	Hyp/Unknown Prot	EAA68502	Gibberella zeae PH-1	hypothetical protein FG01547
2K5	AJ272940	9	Hyp/Unknown Prot	EAA35095	Neurospora crassa	predicted protein

2K7	AJ272946	3d	Prot modification/targeting	CAA22677	Schizosaccharomyces pombe	t-complex protein 1 a chain homolog
2K9	AJ272951	9	Hyp/Unknown Prot	EAA74151	Gibberella zeae PH-1	hypothetical protein FG05089
2K11	AJ272953	5e	Cellular transportation	D49993	Ajellomyces capsulata	ADP-ribosylation factor
3K1	AJ273126	7a	Stress response	NP_014650	Saccharomyces cerevisiae	cell wall integrity and stress response component 1; Slg1p
3K3	AJ273132	3d	Prot modification/targeting	CAC14219	Yarrowia lipolytica	calnexin
3K5	AJ273136	7b	Detoxification	CAA21170	Schizosaccharomyces pombe	UV excision repair protein rad23 homolog
3K7	AJ273138	9	Hyp/Unknown Prot			unknown
3K9	AJ273145	6a	Cell cycle	BAA13080	Schizosaccharomyces pombe	ran-spi1 binding protein
3K11	AJ273148	9	Hyp/Unknown Prot			unknown
4K1	AJ273306	9	Hyp/Unknown Prot	EAA72080	Gibberella zeae PH-1	hypothetical protein FG08503
4K3	AJ273311	2	Energy Metabolism	P44501	Haemophilus influenzae	D-2-hydroxy-acid dehydrogenase homolog
4K5	AJ273314	9	Hyp/Unknown Prot			unknown
4K7	AJ273316	9	Hyp/Unknown Prot	EAA71689	Gibberella zeae PH-1	hypothetical protein FG03487
4K9	AJ273321	7a	Stress response	BAA34384	Trichophyton mentagrophytes	cyclophilin
4K11	AJ273323	3a	Ribosomal proteins	CAA72004	Drosophila melanogaster	40S ribosomal protein S20
5K1	AJ273497	9	Hyp/Unknown Prot	EAA35194	Neurospora crassa	predicted protein
5K3	AJ273501	1c	Lipid metabilism	CAA63976	Nectria hematococca	C14 sterol reductase
5K5	AJ273504	9	Hyp/Unknown Prot	EAA73258	Gibberella zeae PH-1	hypothetical protein FG04474
5K7	AJ273506	1c	Lipid metabilism	AAG13652	Aspergillus oryzae	phosphatidylglycerol/phosphatidylinositol transfer protein
5K9	AJ273508	1c	Lipid metabilism	CAA49804	Saccharomyces cerevisiae	geranylgeranyltransferase regulatory subunit; Mrs6p
5K11	AJ273510	1b	Carbohydrate metabolism	CAE75687	Neurospora crassa	related to peptidylprolyl isomerase (cyclophilin)-like protein
1K2	AJ272776	4c	Transcription	AAB92597	Ustilago maydis	MADS-box homolog Umc1
1K4	AJ272778	9	Hyp/Unknown Prot	EAA64687	Aspergillus nidulans	hypothetical protein AN2582.2
1K6	AJ272780	9	Hyp/Unknown Prot			unknown
1K8	AJ272783	8	Transposable Elements	CAB91877	Phaeosphaeria nodorum	polyprotein
1K10	AJ272786	3c	Proteolysis	NP_593423	Schizosaccharomyces pombe	Highly conserved ring finger; similar to yeast apc11 SPC(ubiquitin-protein ligase activity)
1K12	AJ272789	9	Hyp/Unknown Prot	EAA73470	Gibberella zeae PH-1	hypothetical protein FG04002
2K2	AJ272934	6a	Cell cycle	AAB63523	Emericella nidulans	replication factor C like protein
2K4	AJ272938	9	Hyp/Unknown Prot			unknown
2K6	AJ272945	9	Hyp/Unknown Prot			unknown
2K8	AJ272947	5a	Cell wall	NP_009554	Saccharomyces cerevisiae	involved in cell wall biogenesis; Ecm15p
2K10	AJ272952	7b	Detoxification	CAA21079	Schizosaccharomyces pombe	b transducin
2K12	AJ272954	5f	Signaling	CAA20650	Schizosaccharomyces pombe	GTPase-activator protein for Rho-like GTPases

3K2	AJ273130	3d	Prot modification/targeting	AAA93078	Neurospora crassa	vacuolar ATPase 98 kDa subunit
3K4	AJ273134	4c	Transcription	AAD38380	Emericella nidulans	positive sulphur transcription regulator METR
3K6	AJ273137	1b	Carbohydrate metabolism	AAG08837	Pseudomonas aeruginosa	phosphomannose isomerase/GDP-mannose WbpW
3K8	AJ273139	9	Hyp/Unknown Prot			unknown
3K10	AJ273147	6a	Cell cycle	AAA67467	Schizosaccharomyces pombe	EF-hand protein
3K12	AJ273149	1c	Lipid metabilism	S52745	Ajellomyces capsulatus	stearoyl-CoA desaturase
4K2	AJ273310	9	Hyp/Unknown Prot	EAA68764	Gibberella zeae PH-1	hypothetical protein FG00415
4K4	AJ273312	1a	Aminoacid metabolism	BAC73470	Streptomyces avermitilis	putative dioxygenase
4K6	AJ273315	9	Hyp/Unknown Prot			unknown
4K8	AJ273317	9	Hyp/Unknown Prot	T39513	Schizosaccharomyces pombe	hypothetical protein SPBC1604.01
4K10	AJ273322	2	Energy Metabolism	CAA39949	Neurospora crassa	NADH dehydrogenase (ubiquinone)
4K12	AJ273324	9	Hyp/Unknown Prot			Unknown
5K2	AJ273500	9	Hyp/Unknown Prot	EAA54472	Magnaporthe grisea	hypothetical protein MG02457.4
5K4	AJ273503	9	Hyp/Unknown Prot			unknown
5K6	AJ273505	9	Hyp/Unknown Prot	EAA76593	Gibberella zeae PH-1	hypothetical protein FG07034
5K8	AJ273507	9	Hyp/Unknown Prot			unknown
5K10	AJ273509	9	Hyp/Unknown Prot	EAA77012	Gibberella zeae PH-1	hypothetical protein FG09172
5K12	AJ273511	9	Hyp/Unknown Prot	EAA67248	Gibberella zeae PH-1	hypothetical protein FG00908
1G1	AJ272812	4c	Transcription	T39291	Schizosaccharomyces pombe	hypothetical C2H2 zinc finger protein
1G3	AJ272816	9	Hyp/Unknown Prot			unknown
1G5	AJ272820	9	Hyp/Unknown Prot	EAA49119	Magnaporthe grisea	hypothetical protein MG00777.4
1G7	AJ272822	9	Hyp/Unknown Prot			Unknown
1G9	AJ272824	5f	Signaling	AAF74764	Magnaporhte grisea	MAS1 protein
1G11	AJ272826	6c	DNA synthesis	CAA22533	Schizosaccharomyces pombe	replication factor-a protein 1
2G1	AJ272974	4c	Transcription	XP_330158	Neurospora crassa	related to prefoldin subunit 1: contain a DNA binding leucine zipper motif
2G3	AJ272977	4c	Transcription	CAC18195	Neurospora crassa	similar to Zn-finger transcription factors; Azf1p
2G5	AJ272980	9	Hyp/Unknown Prot			unknown
2G7	AJ272985	9	Hyp/Unknown Prot			unknown
2G9	AJ272993	9	Hyp/Unknown Prot			unknown
2G11	AJ272997	7a	Stress response	NP_955904	Danio rerio	DnaJ (Hsp40) homolog, subfamily C, member 3
3G1	AJ273174	7a	Stress response	CAD21185	Neurospora crassa	related to Hsp90 associated co-chaperone
3G3	AJ273177	9	Hyp/Unknown Prot			unknown
3G5	AJ273179	2	Energy Metabolism	CAA19299	Schizosaccharomyces pombe	SPCC1494.01: putative 2 OG-Fe(II) oxygenase superfamily
3G7	AJ273181	9	Hyp/Unknown Prot			Unknown
3G9	AJ273184	9	Hyp/Unknown Prot	EAA77116	Gibberella zeae PH-1	hypothetical protein FG09559
3G11	AJ273189	2	Energy Metabolism	NP_009751	Saccharomyces cerevisiae	probable carrier protein, mitochondrial; Rim2p
4G1	AJ273366	6d	Cell polarity	BAA26003	Lentinula edodes	similar to adenylyl cyclase associated protein

4G3	AJ273375	1c	Lipid metabilism	AAF19399	Candida albicans	diphosphomevalonate decarboxylase; MVD1
4G5	AJ273377	3a	Ribosomal proteins	CAB52808	Schizosaccharomyces pombe	60S ribosomal protein L11
4G7	AJ273380	3d	Prot modification/targeting	P78695	Neurospora crassa	78 kDa glucose-regulated protein homolog precursor (grp 78) (immunoglobulin heavy chain binding protein homolog) (BIP)
4G9	AJ273384	9	Hyp/Unknown Prot	EAA77732	Gibberella zeae PH-1	hypothetical protein FG09683
4G11	AJ273386	9	Hyp/Unknown Prot			Unknown
5G1	AJ273534	7a	Stress response	AAA82183	Neurospora crassa	70 kDa heat shock protein
5G3	AJ273540	3c	Proteolysis	CAD11898	Metarhizium anisopliae	subtilisin-like serine protease PR1C
5G5	AJ273542	6a	Cell cycle	NP_193034	Arabidopsis thaliana	auxin-responsive protein, putative
5G7	AJ273550	4b	RNA modification	CAB16225	Schizosaccharomyces pombe	probable ATP-dependent RNA helicase
5G9	AJ273553	3d	Prot modification/targeting	CAB98247	Neurospora crassa	related to ubiquitin-activating enzyme homolog
5G11	AJ273558	5c	Extracellular Matrix/Cell wall protein	AAD44757	Filobasidiella neoformans	capsular associated protein
1G2	AJ272814	9	Hyp/Unknown Prot	EAA76224	Gibberella zeae PH-1	hypothetical protein FG06711
1G4	AJ272818	4c	Transcription	CAA93904	Schizosaccharomyces pombe	probable GCS1/GLO3/SPS18 family zinc finger protein
1G6	AJ272821	5f	Signaling	AAK12335	Schizosaccharomyces pombe	LAMMER kinase-like protein
1G8	AJ272823	4c	Transcription	NP_010430	Saccharomyces cerevisiae	transcriptional activator; Swi5p
1G10	AJ272825	4b	RNA modification	NCECX5	Escherichia coli	exodeoxyribonuclease V
1G12	AJ272827	9	Hyp/Unknown Prot			unknown
2G2	AJ272975	3a	Ribosomal proteins	AAM94277	Chlamys farreri	small nuclear ribonucleoprotein D2-like protein
2G4	AJ272978	6e	Sexual cycle	CAB97299	Neurospora crassa	related to tol protein (mediator of mating-type-associated vegetative incompatibility)
2G6	AJ272983	4c	Transcription	NP_057161	Homo sapiens	Bcl-2 inhibitor of transcription; CGI-147 protein
2G8	AJ272989	9	Hyp/Unknown Prot	EAA69971	Gibberella zeae PH-1	hypothetical protein FG10273
2G10	AJ272996	4a	RNA metabolism	BAB12227	Aspergillus oryzae	RNA polymerase II largest subunit
2G12	AJ272999	9	Hyp/Unknown Prot	EAA77604	Gibberella zeae PH-1	hypothetical protein FG06668
3G2	AJ273176	5d	Transport proteins	NP_012390	Saccharomyces cerevisiae	putative phosphatidylinositol transfer protein; Sfh5p
3G4	AJ273178	9	Hyp/Unknown Prot	NP_011515	Saccharomyces cerevisiae	Ygr001cp
3G6	AJ273180	7c	Secondary metabolites	Q51792	Pseudomonas fluorescens	phenazine biosynthesis-like protein; Phzf
3G8	AJ273183	1a	Aminoacid metabolism	AAC37502	Neurospora crassa	acetylglutamate synthase
3G10	AJ273187	9	Hyp/Unknown Prot	EAA67730	Gibberella zeae PH-1	hypothetical protein FG01966
3G12	AJ273192	3a	Ribosomal proteins	P49148	Alternaria alternata	60S acidic ribosomal protein P1
4G2	AJ273371	1a	Aminoacid metabolism	AAN31395	Aspergillus sojae	leucine aminopeptidase
4G4	AJ273376	9	Hyp/Unknown Prot			unknown
4G6	AJ273378	9	Hyp/Unknown Prot	EAA71583	Gibberella zeae PH-1	hypothetical protein FG08277
4G8	AJ273381	9	Hyp/Unknown Prot	EAA67596	Gibberella zeae PH-1	hypothetical protein FG01213

4G10	AJ273385	1d	Nucleotide metabolism	NP_504373	Caenorhabditis elegans	endonuclease/exonuclease/phosphatase family precursor (38.1 kD)
4G12	AJ273389	9	Hyp/Unknown Prot	BAB15008	Homo sapiens	hypothetical protein FLJ21144
5G2	AJ273535	7a	Stress response	CAA67431	Emericella nidulans	heat shock protein 70
5G4	AJ273541	9	Hyp/Unknown Prot	EAA35191	Neurospora crassa	hypothetical protein
5G6	AJ273547	1b	Carbohydrate metabolism	AAA34344	Candida maltosa	glutathion-dependent formaldehyde dehydrogenase
5G8	AJ273551	5d	Transport proteins	P46031	Saccharomyces cerevisiae	peptide transporter; Ptr2-A
5G10	AJ273557	2	Energy Metabolism	AAB53772	Ajellomyces capsulatus	plasma membrane ATPase
5G12	AJ273561	1d	Nucleotide metabolism	AAC07700	Aquifex aeolicus	methylenetetrahydrofolate dehydrogenase
6A1	AJ273588	5e	Cellular transportation	AAF56002	Drosophila melanogaster	clathrin-associated protein, AP-50 gene product
6A3	AJ273590	3a	Ribosomal proteins	A53793	Podospora anserina	ribosomal protein S12
6A5	AJ273593	9	Hyp/Unknown Prot			unknown
6A7	AJ273597	6a	Cell cycle	CAE76120	Neurospora crassa	related to muscle-derived protein (neurite-outgrowth-promoting)
6A9	AJ273605	5b	Cytosqueloton	CAB90777	Schizosaccharomyces pombe	homolog of yeast SLA2 protein-involved in membrane cytoskeleton assembly, cell polarization, internalization phase of endocytosis
6A11	AJ273607	1b	Carbohydrate metabolism	CAC10088	Neurospora crassa	related to n-alkane-inducible cytochrome P450
7A1	AJ273792	2	Energy Metabolism	CAB91241	Neurospora crassa	alcohol dehydrogenase
7A3	AJ273795	3a	Ribosomal proteins	S30133	Saccharomyces cerevisiae	ribosomal protein L14A
7A5	AJ273807	9	Hyp/Unknown Prot			unknown
7A7	AJ273805	5d	Transport proteins	ZP_00141773	Pseudomonas aeruginosa	Cation transport ATPase
7A9	AJ273808	1b	Carbohydrate metabolism	T31685	Aspergillus niger	b-D-galactosidase (lactase)
7A11	AJ273812	5d	Transport proteins	NP_509429	Caenorhabditis elegans	transmembrane 9 superfamily member 2 (75.3 kD) (XJ38):transporter
8A1	AJ274000	9	Hyp/Unknown Prot			unknown
8A3	AJ274002	2	Energy Metabolism	AAQ87554	Rhizobium sp. NGR234	probable Oxidoreductase
8A5	AJ274006	9	Hyp/Unknown Prot			unknown
8A7	AJ274008	3c	Proteolysis	CAB44652	Metarhizium anisopliae	trypsin-related protease
8A9	AJ274012	9	Hyp/Unknown Prot			Unknown
8A11	AJ274014	5f	Signaling	AAB17101	Trichoderma harzianum	14.3.3 protein homolog (TH1433), putative kinesin regulator
9A1	AJ274192	7a	Stress response	AAD08909	Trichophyton rubrum	heat shock protein 70
9A3	AJ274194	1b	Carbohydrate metabolism	CAA81260	Pichia stipitis	transketolase
9A5	AJ274196	1a	Aminoacid metabolism	AAA85391	Yarrowia lipolytica	ATP phosphoribosyltransferase
9A7	AJ274198	3d	Prot modification/targeting	AAG09748	Penicillium chrysogenum	peroxin-1, involved in peroxisome biogenesis
9A9	AJ274202	8	Transposable Elements	AAK16925	Nectria haematococca	restless-like transposase
9A11	AJ274205	5d	Transport proteins	CAD29596	Aspergillus fumigatus	oligonucleotide transporter

10A1	CN807919	1b	Carbohydrate metabolism	AAL09458	Coccidioides posadasii	glucanosyltransferase GEL-1
10A3	CN807930	9	Hyp/Unknown Prot	EAA52857	Magnaporthe grisea	hypothetical protein MG05985
10A5	CN807932	6f	Clock control/light response	Q01302	Neurospora crassa	Clock-controlled protein 6
10A7	CN807936	9	Hyp/Unknown Prot	EAA69727	Gibberella zeae PH-1	hypothetical protein FG02096
10A9	CN807938	9	Hyp/Unknown Prot	NP_745666	Pseudomonas putida	monooxygenase, putative
10A11	CN807943	1b	Carbohydrate metabolism	NP_596551	Schizosaccharomyces pombe	putative mannose-1-phosphate guanyltransferase
6A2	AJ273589	4c	Transcription	AAC24605	Homo sapiens	hypothetical Kruppel-type Zinc Finger Protein, F18547_1
6A4	AJ273592	1e	N-, P-, S-metabolism	BAA08305	Neurospora crassa	thioredoxin
6A6	AJ273595	9	Hyp/Unknown Prot			unknown
6A8	AJ273600	9	Hyp/Unknown Prot			unknown
6A10	AJ273606	9	Hyp/Unknown Prot	EAA28422	Neurospora crassa	predicted protein
6A12	AJ273609	9	Hyp/Unknown Prot			unknown
7A2	AJ273794	5f	Signaling	AAD17221	Metarhizium anisopliae	cAMP-dependent protein kinase catalytic subunit
7A4	AJ273797	9	Hyp/Unknown Prot	T40098	Schizosaccharomyces pombe	hypothetical protein SPBC2A9.07c
7A6	AJ273802	3c	Proteolysis	NP_011504	Saccharomyces cerevisiae	proteasome subunit YC7a/Y8 (protease yscE subunit 7)
7A8	AJ273806	1a	Aminoacid metabolism	AAN31395	Aspergillus sojae	leucine aminopeptidase
7A10	AJ273810	5b	Cytosqueloton	CAA60953	Saccharomyces cerevisiae	nonessential type-I transmembrane protein homologous to intracellular lectins, Emp47
7A12	AJ273816	9	Hyp/Unknown Prot	EAA68782	Gibberella zeae PH-1	hypothetical protein FG00433
8A2	AJ274001	9	Hyp/Unknown Prot	EAA36088	Neurospora crassa	hypothetical protein
8A4	AJ274003	1c	Lipid metabilism	CAA06156	Zea mays	cytochrome P450 monooxygenase
8A6	AJ274007	1b	Carbohydrate metabolism	O00093	Emericella nidulans	3-phytase B precursor (myo-inositol-hexaphosphate 3-phosphohydrolase B)
8A8	AJ274010	1b	Carbohydrate metabolism	NP_593363	Schizosaccharomyces pombe	putative glycosyl transferase
8A10	AJ274013	9	Hyp/Unknown Prot			unknown
8A12	AJ274019	5a	Cell wall	AAF80600	Trichoderma atroviride	glucan 1,3-b-glucosidase GLUC78 precursor
9A2	AJ274193	9	Hyp/Unknown Prot	EAA75991	Gibberella zeae PH-1	hypothetical protein FG06984
9A4	AJ274195	9	Hyp/Unknown Prot			unknown
9A6	AJ274197	9	Hyp/Unknown Prot	EAA77757	Gibberella zeae PH-1	hypothetical protein FG09708
9A8	AJ274200	1a	Aminoacid metabolism	CAC18796	Emericella nidulans	proline oxidase
9A10	AJ274203	8	Transposable Elements	AAA88791	Fusarium oxysporum	pol polyprotein homolog
9A12	AJ274206	1a	Aminoacid metabolism	CAA20731	Schizosaccharomyces pombe	probable asparagine synthase
10A2	CN807921	7c	Secondary metabolites	ZP_00175938	Crocosphaera watsonii	Non-ribosomal peptide synthetase modules and related proteins
10A4	CN807931	5e	Cellular transportation	NP_588516	Schizosaccharomyces pombe	putative potassium channel subunit
10A6	CN807933	9	Hyp/Unknown Prot			Unknown
10A8	CN807937	5c	Extracellular Matrix/Cell wall protein	AAL47843	Fusarium oxysporum	extracellular matrix protein precursor

10A10	CN807940	9	Hyp/Unknown Prot			Unknown
10A12	CN807954	1e	N-, P-, S-metabolism	CAE76366	Neurospora crassa	probable sulfate adenylyltransferase
6C1	AJ273635	9	Hyp/Unknown Prot	EAA57086	Magnaporthe grisea	hypothetical protein MG08055.4
6C3	AJ273641	1f	Cofactor and vitamins	CAE76129	Neurospora crassa	related to indoleamine 2, 3-dioxygenase
6C5	AJ273644	1c	Lipid metabilism	BAA74775	Homo sapiens	phosphatidylinositol glycan, class L (Pig-L)
6C7	AJ273648	1c	Lipid metabilism	CAB97289	Neurospora crassa	putative d (24)-sterol C-methyltransferase
6C9	AJ273655	9	Hyp/Unknown Prot			unknown
6C11	AJ273658	9	Hyp/Unknown Prot			unknown
7C1	AJ273844	9	Hyp/Unknown Prot	EAA68724	Gibberella zeae PH-1	hypothetical protein FG00492
7C3	AJ273847	5c	Extracellular Matrix/Cell wall protein	AAA20128	Magnaporthe grisea	hydrophobin-like protein
7C5	AJ273850	9	Hyp/Unknown Prot			unknown
7C7	AJ273855	1b	Carbohydrate metabolism	CAC18200	Neurospora crassa	probable glycogen synthase kinase 3 a
7C9	AJ273863	9	Hyp/Unknown Prot			unknown
7C11	AJ273867	9	Hyp/Unknown Prot	CAC09402	Neurospora crassa	hypothetical protein
8C1	AJ274047	6a	Cell cycle	NP_011459	Saccharomyces cerevisiae	homolog of S. pombe SDS23; localizes to spindle pole body; Sds23p
8C3	AJ274050	7b	Detoxification	BAA96543	Tursiops truncatus	p67-phox, part of NADPH oxidase that produces superoxide ions in response to microbial infection
8C5	AJ274055	9	Hyp/Unknown Prot	EAA73783	Gibberella zeae PH-1	hypothetical protein FG05608
8C7	AJ274058	1b	Carbohydrate metabolism	NP_012104	Saccharomyces cerevisiae	invertase (sucrose hydrolyzing enzyme)
8C9	AJ274062	9	Hyp/Unknown Prot	EAA70485	Gibberella zeae PH-1	hypothetical protein FG02410
8C11	AJ274064	6a	Cell cycle	CAE76477	Neurospora crassa	related to cell cycle regulation and aging protein
9C1	AJ274228	9	Hyp/Unknown Prot			unknown
9C3	AJ274235	4c	Transcription	AAB04132	Fusarium solani f. sp. Pisi	cutinase G-box binding protein
9C5	AJ274238	9	Hyp/Unknown Prot			unknown
9C7	AJ274240	8	Transposable Elements	CAB91877	Phaeosphaeria nodorum	polyprotein
9C9	AJ274243	2	Energy Metabolism	NP_013139	Saccharomyces cerevisiae	subunit VIb of cytochrome c oxidase; Cox12p
9C11	AJ274249	3b	Translation	T38786	Schizosaccharomyces pombe	translation intiation factor eif-3
10C1	CN807990	5d	Transport proteins	NP_011569	Saccharomyces cerevisiae	high affinity methionine permease; Mup1p
10C3	CN808315	5c	Extracellular Matrix/Cell wall protein	T49444	Neurospora crassa	related to lustrin A
10C5	CN807981	1b	Carbohydrate metabolism	AAL09829	Coccidioides posadasii	beta-glucosidase 5
10C7	CN807983	1a	Aminoacid metabolism	AAG36950	Aspergillus nidulans	phe-inhibited DAHP (3-deoxy-D-arabino-heptulosonate-7-phosphate) synthase
10C9	CN808016	5e	Cellular transportation	CAD15482	Ralstoni*Alternaria solani*anacearum	putative signal peptide protein
10C11	CN808019	7b	Detoxification	CAD71220	Neurospora crassa	related to chloroperoxidase

6C2	AJ273636	1c	Lipid metabilism	CAA93903	Schizosaccharomyces pombe	putative phosphatidylinositol 4-kinase
6C4	AJ273643	3a	Ribosomal proteins	CAA20835	Schizosaccharomyces pombe	60S ribosomal protein L27-A
6C6	AJ273646	1b	Carbohydrate metabolism	ZP_00182529	Exiguobacterium sp.	COG2084: 3-hydroxyisobutyrate dehydrogenase and related beta-hydroxyacid dehydrogenases
6C8	AJ273651	9	Hyp/Unknown Prot	T50959	Neurospora crassa	probable endopolyphosphatase (EC 3.6.1.10) B24P7.140 precursor
6C10	AJ273657	5f	Signaling	NP_013943	Saccharomyces cerevisiae	SRPK1-like Kinase
6C12	AJ273659	5b	Cytosqueloton	T40661	Schizosaccharomyces pombe	reduced viability upon starvation protein 161 homolog, implicated in cell growth and cytoskeletal or ganisation
7C2	AJ273845	5c	Extracellular Matrix/Cell wall protein	CAA12162	Aspergillus fumigatus	IgE-binding protein
7C4	AJ273849	5d	Transport proteins	CAB91174	Schizosaccharomyces pombe	putative MFS allantoate permease
7C6	AJ273853	3b	Translation	AAC43437	Escherichia coli	peptide chain release factor 1
7C8	AJ273858	7a	Stress response	AAS80314	Nectria haematococca	SesB: stress-sensitive B
7C10	AJ273865	9	Hyp/Unknown Prot	EAA77836	Gibberella zeae PH-1	hypothetical protein FG07238
7C12	AJ273869	2	Energy Metabolism	AAC49575	Ustilago maydis	indole-3-acetaldehyde dehydrogenase
8C2	AJ274049	5f	Signaling	AAN64312	Magnaporthe grisea	MAC1 interacting protein 1; ACI1
8C4	AJ274053	9	Hyp/Unknown Prot			unknown
8C6	AJ274057	6a	Cell cycle	CAB98235	Neurospora crassa	related to trfA protein
8C8	AJ274060	1d	Nucleotide metabolism	CAB88582	Neurospora crassa	probable sugar transporter
8C10	AJ274063	9	Hyp/Unknown Prot	EAA48521	Magnaporthe grisea	hypothetical protein MG00179.4
8C12	AJ274066	1a	Aminoacid metabolism	NP_009845	Saccharomyces cerevisiae	aminopeptidase yscIII; Ape3p
9C2	AJ274229	9	Hyp/Unknown Prot	EAA73487	Gibberella zeae PH-1	predicted protein
9C4	AJ274237	9	Hyp/Unknown Prot			unknown
9C6	AJ274239	1c	Lipid metabilism	CAA42906	Penicillium chrysogenum	lysophospholipase precursor
9C8	AJ274241	9	Hyp/Unknown Prot	EAA72912	Gibberella zeae PH-1	hypothetical protein FG03172
9C10	AJ274245	9	Hyp/Unknown Prot			Unknown
9C12	AJ274250	6a	Cell cycle	CAB93660	Podospora anserina	Ami1 protein, involved in nuclear migration events
10C2	CN807992	9	Hyp/Unknown Prot	EAA67432	Gibberella zeae PH-1	hypothetical protein FG02585
10C4	CN807993	9	Hyp/Unknown Prot	EAA71040	Gibberella zeae PH-1	hypothetical protein FG04362
10C6	CN808011	9	Hyp/Unknown Prot			Unknown
10C8	CN808013	9	Hyp/Unknown Prot	EAA64876	Aspergillus nidulans	hypothetical protein AN2044
10C10	CN808018	1c	Lipid metabilism	AAF19345	Nicotiana tabacum	diacylglycerol acylCoA acyltransferase
10C12	CN807949	7a	Stress response	NP_011835	Saccharomyces cerevisiae	cell wall integrity and stress response component 4; Wsc4p
6K1	AJ273681	3a	Ribosomal proteins	CAA21187	Schizosaccharomyces pombe	40S ribosomal protein S2
6K3	AJ273687	7a	Stress response	NP_013352	Saccharomyces cerevisiae	stress-induced yeast MPV17 homolog; Ylr251wp (is a closely related peroxisomal protein involved in the development of early-onset glomerulosclerosis

6K5	AJ273692	9	Hyp/Unknown Prot			unknown
6K7	AJ273695	5e	Cellular transportation	CAB76027	Schizosaccharomyces pombe	putative clathrin-associated protein (AP)
6K9	AJ273699	4a	RNA metabolism	T38507	Schizosaccharomyces pombe	probable cysteinyl-tRNA synthetase
6K11	AJ273703	5d	Transport proteins	CAC04436	Neurospora crassa	related to Nup98-Nup96 precursor
7K1	AJ273890	1c	Lipid metabilism	BAB93117	Saccharomyces kluyveri	putative delta 8-sphingolipid desaturase
7K3	AJ273897	9	Hyp/Unknown Prot	EAA73656	Gibberella zeae PH-1	hypothetical protein FG04330
7K5	AJ273901	5e	Cellular transportation	AAO59280	Botryotinia fuckeliana	kinesin: play a role in organelle transport
7K7	AJ273906	9	Hyp/Unknown Prot	EAA67769	Gibberella zeae PH-1	hypothetical protein FG02539
7K9	AJ273910	2	Energy Metabolism	A35935	Neurospora crassa	NADH dehydrogenase (ubiquinone) 31K chain precursor
7K11	AJ273912	4c	Transcription	BAA95967	Aspergillus oryzae	transcription activator
8K1	AJ274096	1b	Carbohydrate metabolism	XP_326808	Neurospora crassa	related to UDP-N-acetylglucosamine-peptide-N-acetylglucosaminyltransferase
8K3	AJ274102	3c	Proteolysis	AAB68600	Metarhizium anisopliae	carboxypeptidase
8K5	AJ274105	9	Hyp/Unknown Prot	EAA35967	Neurospora crassa	hypothetical protein
8K7	AJ274110	9	Hyp/Unknown Prot	EAA71239	Gibberella zeae PH-1	hypothetical protein FG03206
8K9	AJ274121	9	Hyp/Unknown Prot			unknown
8K11	AJ274124	1c	Lipid metabilism	CAB91111	Yarrowia lipolytica	triacylglycerol lipase
9K1	AJ274287	9	Hyp/Unknown Prot	CAA30054	Kluyveromyces marxianus var. lactis	hypothetical protein (LAC12 3' region)
9K3	AJ274289	9	Hyp/Unknown Prot			unknown
9K5	AJ274297	3d	Prot modification/targeting	T39449	Schizosaccharomyces pombe	probable importin b-4 subunit
9K7	AJ274301	9	Hyp/Unknown Prot			unknown
9K9	AJ274303	4b	RNA modification	BAB03001	Arabidopsis thaliana	glycine-rich RNA binding protein-like
9K11	AJ274307	1a	Aminoacid metabolism	AAF82115	Emericella nidulans	cobalamin-independent methionine synthase
10K1	CN808063	5f	Signaling	AAF74764	Magnaporthe grisea	MAS1 protein: appressorium formation
10K3	CN808073	1e	N-, P-, S-metabolism	A49848	Neurospora crassa	nitrite reductase [NAD(P)H] (EC 1.7.1.4)
10K5	CN808085	9	Hyp/Unknown Prot			Unknown
10K7	CN808182	1b	Carbohydrate metabolism	NP_518315	Ralstoni*Alternaria solani*anacearum	Probale zinc-dependent alcohol dehydrogenase oxidoreductase protein
10K9	CN808098	1b	Carbohydrate metabolism	CAD70754	Neurospora crassa	probable beta (1-3) glucanosyltransferase gel3p
10K11	CN808107	3d	Prot modification/targeting	AAH44087	Xenopus laevis	MGC52717 protein: involved intra-mitochondrial protein sorting, trafficking
6K2	AJ273685	9	Hyp/Unknown Prot	EAA68629	Gibberella zeae PH-1	hypothetical protein FG01163
6K4	AJ273690	9	Hyp/Unknown Prot	EAA77673	Gibberella zeae PH-1	hypothetical protein FG09811
6K6	AJ273694	9	Hyp/Unknown Prot	BAB34228	Escherichia coli	hypothetical C4-type zinc finger protein TraR-family
6K8	AJ273697	9	Hyp/Unknown Prot			unknown
6K10	AJ273702	3b	Translation	NP_012581	Saccharomyces cerevisiae	anaerobically expressed form of translation initiation factor eIF-5A

6K12	AJ273704	5e	Cellular transportation	CAC18232	Neurospora crassa	probable Erv25 protein (component of the COPII-coated vesicles)
7K2	AJ273896	1d	Nucleotide metabolism	CAB46409	Streptomyces coelicolor	probable large secreted protein, similar to extracellular nucleases
7K4	AJ273900	1b	Carbohydrate metabolism	AAP21783	Malus x domestica	L-galactose dehydrogenase
7K6	AJ273904	1f	Cofactor and vitamins	T38920	Schizosaccharomyces pombe	methylenetetrahydrofolate reductase 2
7K8	AJ273907	3a	Ribosomal proteins	CAA21088	Schizosaccharomyces pombe	60S ribosomal protein L10A
7K10	AJ273911	9	Hyp/Unknown Prot	EAA69244	Gibberella zeae PH-1	hypothetical protein FG00583
7K12	AJ273917	9	Hyp/Unknown Prot	EAA75502	Gibberella zeae PH-1	hypothetical protein FG05266
8K2	AJ274099	9	Hyp/Unknown Prot	EAA68588	Gibberella zeae PH-1	hypothetical protein FG00552
8K4	AJ274103	9	Hyp/Unknown Prot			unknown
8K6	AJ274108	1c	Lipid metabilism	CAB91731	Neurospora crassa	phosphatidic acid-preferring phospholipase A1 related protein
SSC6						
8K10	AJ274122	2	Energy Metabolism	NP_822542	Streptomyces avermitilis	putative oxidoreductase
8K12	AJ274125	4a	RNA metabolism	NP_013105	Saccharomyces cerevisiae	component of RNA polymerase transcription factor TFIIH; Ssl1p
9K2	AJ274288	9	Hyp/Unknown Prot	EAA70296	Gibberella zeae PH-1	hypothetical protein FG10674
9K4	AJ274292	9	Hyp/Unknown Prot			unknown
9K6	AJ274298	2	Energy Metabolism	AAA20440	Aspergillus parasiticus	pyruvate decarboxylase
9K8	AJ274302	1a	Aminoacid metabolism	AAB03015	Escherichia coli	similar to 3-hydroxyisobutyrate dehydrogenases
9K10	AJ274305	3c	Proteolysis	ZP_00218124	Burkholderia cepacia	Putative intracellular protease/amidase
9K12	AJ274309	9	Hyp/Unknown Prot	T49865	Neurospora crassa	hypothetical protein B24P11.210
10K2	CN808070	5e	Cellular transportation	EAA74131	Gibberella zeae PH-1	ADP,ATP carrier protein (ADP/ATP translocase)(adenine nucleotide translocater)(ANT)
10K4	CN808077	9	Hyp/Unknown Prot			Unknown
10K6	CN808183	1b	Carbohydrate metabolism	CAE76318	Neurospora crassa	related to glyoxal oxidase precursor
10K8	CN808094	3c	Proteolysis	NP_588321	Schizosaccharomyces pombe	putative Zn-protease
10K10	CN808105	1f	Cofactor and vitamins	BAA33011	Fusarium oxysporum	flavohemoglobin (Dihydropteridine reductase)
10K12	CN808108	4c	Transcription	NP_633330	Methanosarcina mazei	HTH DNA-binding protein
6G1	AJ273735	9	Hyp/Unknown Prot	CAB40775	Arabidopsis thaliana	putative protein
6G3	AJ273743	3b	Translation	Q94HF1	Oryza sativa	Eukaryotic translation initiation factor 3 subunit 11 (eIF-3 p25)
6G5	AJ273750	1b	Carbohydrate metabolism	AAH67265	Homo sapiens	Lectin, mannose-binding 2-like
6G7	AJ273760	4b	RNA modification	T40623	Schizosaccharomyces pombe	probable polyA-binding protein
6G9	AJ273764	9	Hyp/Unknown Prot			unknown
6G11	AJ273769	6a	Cell cycle	P87072	Neurospora crassa	calcineurin B subunit (protein phosphatase 2B regulatory subunit)
7G1	AJ273949	9	Hyp/Unknown Prot	EAA70450	Gibberella zeae PH-1	hypothetical protein FG00857
7G3	AJ273952	1b	Carbohydrate metabolism	NP_012863	Saccharomyces cerevisiae	aldolase; Fba1p

7G5	AJ273956	1a	Aminoacid metabolism	CAB85685	Agaricus bisporus	aminopeptidase
7G7	AJ273961	9	Hyp/Unknown Prot	EAA36134	Neurospora crassa	hypothetical protein
7G9	AJ273965	9	Hyp/Unknown Prot	EAA50508	Magnaporthe grisea	hypothetical protein MG04267.4
7G11	AJ273967	9	Hyp/Unknown Prot	CAB91329	Neurospora crassa	hypothetical protein B21J21.70
8G1	AJ274150	9	Hyp/Unknown Prot	EAA73796	Gibberella zeae PH-1	hypothetical protein FG10770
8G3	AJ274152	1a	Aminoacid metabolism	BAA61829	Pseudomonas putida	salicylate hydroxylase
8G5	AJ274156	5c	Extracellular Matrix/Cell wall protein	AAA33418	Metarhizium anisopliae	hydrophobin-like protein ssgA precursor
8G7	AJ274160	2	Energy Metabolism	AAG05038	Pseudomonas aeruginosa	probable short-chain dehydrogenase
8G9	AJ274163	9	Hyp/Unknown Prot	AAK16925	Nectria haematococca	restless-like transposase
8G11	AJ274168	3c	Proteolysis	AAA20876	Aspergillus niger	pepsinogen
9G1	AJ274329	3c	Proteolysis	AAD29675	Metarhizium anisopliae	trypsin-related protease, try 2
9G3	AJ274332	2	Energy Metabolism	AAD16178	Aspergillus oryzae	pyruvate decarboxylase
9G5	AJ274336	9	Hyp/Unknown Prot	EAA67364	Gibberella zeae PH-1	hypothetical protein FG00333
9G7	AJ274338	8	Transposable Elements	CAA96388	Fusarium poae	gag-like polyprotein
9G9	AJ274342	9	Hyp/Unknown Prot			unknown
9G11	AJ274347	1b	Carbohydrate metabolism	EAA75069	Gibberella zeae PH-1	Formate dehydrogenase (NAD-dependent formate dehydrogenase) (FDH)
10G1	CN808152	9	Hyp/Unknown Prot	EAA68463	Gibberella zeae PH-1	hypothetical protein FG00450
10G3	CN808158	1c	Lipid metabilism	AAN27998	Leptosphaeria maculans	sterol delta 5,6-desaturase ERG3
10G5	CN808163	1b	Carbohydrate metabolism	AAB22823	Saccharomyces cerevisiae	fructose-2,6-bisphosphatase: required for glucose metabolism
10G7	CN808165	5d	Transport proteins	CAE76088	Neurospora crassa	related to neutral amino acid permease
10G9	CN808173	2	Energy Metabolism	NP_539404	Brucella melitensis	ATP synthase beta subunit/transription termination factor rho
10G11	CN808176	1b	Carbohydrate metabolism	EAA67345	Gibberella zeae PH-1	Ketol-acid reductoisomerase, mitochondrial precursor
6G2	AJ273736	9	Hyp/Unknown Prot	EAA48669	Magnaporthe grisea	hypothetical protein MG00327.4
6G4	AJ273749	2	Energy Metabolism	CAA25104	Neurospora crassa	ADP, ATP carrier protein (ADP/ATP translocase)
6G6	AJ273755	6a	Cell cycle	CAB83164	Schizosaccharomyces pombe	cut14 protein, involved in chromosome segregation
6G8	AJ273762	2	Energy Metabolism	NP_011169	Saccharomyces cerevisiae	dihydrolipoamide dehydrogenase precursor
6G10	AJ273765	5e	Cellular transportation	CAA21305	Schizosaccharomyces pombe	probable mitochondrial protein import protein
6G12	AJ273770	3b	Translation	CAC28833	Neurospora crassa	probable translation elongation factor EF-Tu precursor, mitochondrial; Tuf1p
7G2	AJ273950	1d	Nucleotide metabolism	CAB63906	Metarhizium anisopliae var. anisopliae	DNase1 protein
7G4	AJ273955	1b	Carbohydrate metabolism	P16928	Emericella nidulans	acetyl-coenzyme A synthetase
7G6	AJ273960	9	Hyp/Unknown Prot	EAA75018	Gibberella zeae PH-1	hypothetical protein FG10761
7G8	AJ273964	6a	Cell cycle	AAC50544	Homo sapiens	cullin 1

7G10	AJ273966	5e	Cellular transportation	NP_596668	Schizosaccharomyces pombe	vesicle transport v-snare protein
7G12	AJ273972	9	Hyp/Unknown Prot	EAA52967	Magnaporthe grisea	hypothetical protein MG06095.4
8G2	AJ274151	9	Hyp/Unknown Prot	EAA68994	Gibberella zeae PH-1	hypothetical protein FG01697.1
8G4	AJ274154	2	Energy Metabolism	CAB92021	Neurospora crassa	putative b-succinyl CoA synthetase precursor
8G6	AJ274159	5f	Signaling	EAA33006	Neurospora crassa	RAS-RELATED PROTEIN RAB1BV
8G8	AJ274161	9	Hyp/Unknown Prot	EAA71197	Gibberella zeae PH-1	hypothetical protein FG04176
8G10	AJ274166	9	Hyp/Unknown Prot	EAA70462	Gibberella zeae PH-1	hypothetical protein FG00869.1
8G12	AJ274170	7a	Stress response	AAG03845	Pseudomonas aeruginosa	probable cold-shock protein
9G2	AJ274331	9	Hyp/Unknown Prot			unknown
9G4	AJ274333	1a	Aminoacid metabolism	AAP73942	Rhodococcus erythropolis	putative peptidase
9G6	AJ274337	9	Hyp/Unknown Prot			Unknown
9G8	AJ274340	3c	Proteolysis	CAA21278	Schizosaccharomyces pombe	ubi4-ubiquitin family protein
9G10	AJ274343	3c	Proteolysis	AAB68600	Metarhizium anisopliae	carboxypeptidase
9G12	AJ274355	9	Hyp/Unknown Prot			unknown
10G2	CN808156	5d	Transport proteins	NP_013231	Saccharomyces cerevisiae	Low-affinity zinc transporter of the plasma membrane
10G4	CN808162	3c	Proteolysis	CAB63907	Metarhizium anisopliae	Subtilisin-like protease PR1H
10G6	CN808164	1c	Lipid metabilism	CAA59939	Ajellomyces capsulatus	delta-9 fatty acid desaturase; stearoyl-CoA desaturase
10G8	CN808171	9	Hyp/Unknown Prot	EAA72775	Gibberella zeae PH-1	hypothetical protein FG04394
10G10	CN808066	9	Hyp/Unknown Prot			Unknown
10G12	CN808178	1d	Nucleotide metabolism	CAB63906	Metarhizium anisopliae	DNase1 protein
1B1	AJ272709	4c	Transcription	CAD21367	Neurospora crassa	transcription elongation complex subunit (CDC68)
1B3	AJ272715	1a	Aminoacid metabolism	NP_200424	Arabidopsis thaliana	transducin family protein: nucleotide binding, pre-mRNA processing and cytoskeleton assembly: hydrolysis of guanosine 3',5'-monophosphate
1B5	AJ272720	2	Energy Metabolism	CAA28860	Neurospora crassa	cytochrome C1, heme protein precursor
1B7	AJ272722	9	Hyp/Unknown Prot			unknown
1B9	AJ272726	2	Energy Metabolism	CAB91450	Neurospora crassa	cytochrome C oxidase chain V precursor
1B11	AJ272730	9	Hyp/Unknown Prot	EAA75055	Gibberella zeae PH-1	hypothetical protein FG06113
2B1	AJ272870	9	Hyp/Unknown Prot	EAA77264	Gibberella zeae PH-1	hypothetical protein FG07892
2B3	AJ272874	1b	Carbohydrate metabolism	AAK48000	Mycobacterium tuberculosis CDC1551	3-ketosteroid-delta-1-dehydrogenase, putative
2B5	AJ272880	3d	Prot modification/targeting	NP_595018	Schizosaccharomyces pombe	putative amidase
2B7	AJ272884	4c	Transcription	CAA04448	Emericella nidulans	SamB protein, Zn finger-like domain, developmental regulator
2B9	AJ272891	9	Hyp/Unknown Prot	EAA76368	Gibberella zeae PH-1	hypothetical protein FG06846
2B11	AJ272894	5e	Cellular transportation	CAA83551	Cricetulus griseus	coatomer complex epsilon chain

3B1	AJ273062	5e	Cellular transportation	CAA62944	Saccharomyces cerevisiae	P24B protein precursor, involved in secretory protein transport
3B3	AJ273064	9	Hyp/Unknown Prot	EAA73899	Gibberella zeae PH-1	hypothetical protein FG05186
3B5	AJ273066	7b	Detoxification	AAF79420	Arabidopsis thaliana	F18O14.4, similar to thaumatin-like proteins
3B7	AJ273068	1a	Aminoacid metabolism	CAA19028	Schizosaccharomyces pombe	phosphoribosylanthranilate transferase
3B9	AJ273071	3d	Prot modification/targeting	CAD70873	Neurospora crassa	probable multiprotein bridging factor MBF1
3B11	AJ273076	7a	Stress response	CAA20737	Schizosaccharomyces pombe	Chaperonin; hsp78p
4B1	AJ273238	1a	Aminoacid metabolism	JX0152	Fusarium solani	D-amino-acid oxidase
4B3	AJ273241	9	Hyp/Unknown Prot	EAA28451	Neurospora crassa	hypothetical protein
4B5	AJ273243	9	Hyp/Unknown Prot			unknown
4B7	AJ273246	9	Hyp/Unknown Prot	EAA72091	Gibberella zeae PH-1	hypothetical protein FG08514
4B9	AJ273254	6g	Cell death	NP_011747	Saccharomyces cerevisiae	Possible role in cell aging; Phb2p (prohibitin antiproliferative protein)
4B11	AJ273261	9	Hyp/Unknown Prot	EAA77661	Gibberella zeae PH-1	hypothetical protein FG09799
5B1	AJ273429	8	Transposable Elements	BAA32244	Fusarium oxysporum	transposase-like protein
5B3	AJ273432	3a	Ribosomal proteins	EAA68463	Gibberella zeae PH-1	hypothetical protein FG00450
5B5	AJ273438	6c	DNA synthesis	CAE76388	Neurospora crassa	related to DNA polymerase V
5B7	AJ273440	9	Hyp/Unknown Prot			unknown
5B9	AJ273446	6a	Cell cycle	NP_009840	Saccharomyces cerevisiae	probable G-protein, -transducin type; Ybr281cp
5B11	AJ273453	1b	Carbohydrate metabolism	CAB91480	Neurospora crassa	probable branching enzyme (be1)
1B2	AJ272713	9	Hyp/Unknown Prot	EAA67250	Gibberella zeae PH-1	hypothetical protein FG01872
1B4	AJ272716	4c	Transcription	NP_079788	Mus musculus	RIKEN cDNA 2310008M20: Predicted Zn-finger protein
1B6	AJ272721	1c	Lipid metabilism	CAA93797	Schizosaccharomyces pombe	isopentenyl-diphosphate d-isomerase
1B8	AJ272723	5f	Signaling	BAB88922	Magnaporthe grisea	neuronal calcium sensor 1
1B10	AJ272728	6a	Cell cycle	CAD70986	Neurospora crassa	related to sporulation protein SPO72
1B12	AJ272732	9	Hyp/Unknown Prot	EAA59972	Aspergillus nidulans	hypothetical protein AN3764.2
2B2	AJ272873	1b	Carbohydrate metabolism	AAF80600	Trichoderma harzianum	glucan 1,3-beta-glucosidase GLUC78 precursor
2B4	AJ272879	1e	N-, P-, S-metabolism	AAB31768	Asperigllus ficuum	acid phosphatase
2B6	AJ272883	9	Hyp/Unknown Prot	EAA68070	Gibberella zeae PH-1	hypothetical protein FG10168
2B8	AJ272888	9	Hyp/Unknown Prot			unknown
2B10	AJ272893	9	Hyp/Unknown Prot	EAA47045	Magnaporthe grisea	hypothetical protein MG10856.4
2B12	AJ272897	5e	Cellular transportation	T39073	Schizosaccharomyces pombe	synaptobrevin homolog1
3B2	AJ273063	9	Hyp/Unknown Prot	EAA71440	Gibberella zeae PH-1	hypothetical protein FG08579
3B4	AJ273065	9	Hyp/Unknown Prot	EAA75055	Gibberella zeae PH-1	hypothetical protein FG06113
3B6	AJ273067	9	Hyp/Unknown Prot			unknown
3B8	AJ273069	5e	Cellular transportation	CAA53509	Drosophila melanogaster	b-adaptin 1
3B10	AJ273072	9	Hyp/Unknown Prot	EAA74132	Gibberella zeae PH-1	hypothetical protein FG06022
3B12	AJ273078	4c	Transcription	P78706	Neurospora crassa	Transcriptional repressor rco-1
4B2	AJ273239	9	Hyp/Unknown Prot	EAA72148	Gibberella zeae PH-1	predicted protein

4B4	AJ273242	9	Hyp/Unknown Prot			unknown
4B6	AJ273245	3d	Prot modification/targeting	ZP_00085927	Pseudomonas fluorescens PfO-1	Histone acetyltransferase HPA2 and related acetyltransferases
4B8	AJ273248	2	Energy Metabolism	JC1111	Neurospora crassa	H+-transporting ATP synthase a chain
4B10	AJ273260	4c	Transcription	T37669	Schizosaccharomyces pombe	hypothetical zinc-finger protein
4B12	AJ273262	9	Hyp/Unknown Prot	T41257	Schizosaccharomyces pombe	hypothetical protein SPCC285.11
5B2	AJ273431	6a	Cell cycle	AAD45191	Homo sapiens	cullin homolog 4A
5B4	AJ273435	3a	Ribosomal proteins	S25374	Candida maltosa	40S ribosomal protein S13 (S15)
5B6	AJ273439	7c	Secondary metabolites	AAC32591	Pinus taeda	phenylcoumaran benzylic ether reductase
5B8	AJ273442	9	Hyp/Unknown Prot	EAA74036	Gibberella zeae PH-1	hypothetical protein FG06071
5B10	AJ273447	3a	Ribosomal proteins	CAA91100	Schizosaccharomyces pombe	40S ribosomal protein S6
5B12	AJ273454	9	Hyp/Unknown Prot			unknown
1D1	AJ272753	2	Energy Metabolism	CAC18222	Neurospora crassa	H+-transporting ATPase lipid-binding protein
1D3	AJ272756	2	Energy Metabolism	CAA26308	Neurospora crassa	cytochrome c reductase iron-sulfur subunit
1D5	AJ272758	9	Hyp/Unknown Prot			unknown
1D7	AJ272764	6a	Cell cycle	AAM46085	Candida albicans	putative regulatory protein to increase pseudohyphae formation
1D9	AJ272766	1b	Carbohydrate metabolism	AAB18921	Dictyostelium discoideum	similar to human 70 kD a-glucosidase, ModA
1D11	AJ272769	5b	Cytosqueloton	NP_492270	Caenorhabditis elegans	tubulin-specific chaperone d (1I940)
2D1	AJ272916	1b	Carbohydrate metabolism	AAO27746	Fusarium sporotrichioides	putative methyltransferase
2D3	AJ272919	3c	Proteolysis	BAA04974	Aspergillus phoenicis	carboxypeptidase CPDS precursor
2D5	AJ272922	7a	Stress response	CAB88238	Schizosaccharomyces pombe	SPBC1711.08: stress-regulated co-chaperone; activator of Hsp90p
2D7	AJ272925	6a	Cell cycle	CAD62574	Schizosaccharomyces pombe	apc2:anaphase-promoting complex; required for cyclin degradation; involved in metaphase-anaphase transition; cullin family
2D9	AJ272929	1c	Lipid metabilism	AAG06814	Pseudomonas aeruginosa	probable enoyl CoA-hydratase/isomerase
2D11	AJ272931	1a	Aminoacid metabolism	AAL06650	Streptomyces globisporus	truncated methionine synthase-like protein
3D1	AJ273110	1c	Lipid metabilism	Q10137	Schizosaccharomyces pombe	putative Sec14 cytosolic factor (phosphatidyl-inositol/phosphatidyl-choline transfer protein)
3D3	AJ273113	9	Hyp/Unknown Prot			unknown
3D5	AJ273115	3b	Translation	NP_009365	Saccharomyces cerevisiae	GTPase, required for general translation initiation by promoting Met-tRNAiMet binding to ribosomes and ribosomal subunit joining; homolog of bacterial IF2; Fun12p
3D7	AJ273118	5d	Transport proteins	NP_015520	Saccharomyces cerevisiae	oligopeptide transporter, Opt2, Ypr194cp

3D9	AJ273121	1b	Carbohydrate metabolism	AAB52507	Aspergillus terreus	phytase
SSC3						
4D1	AJ273283	9	Hyp/Unknown Prot			unknown
4D3	AJ273287	9	Hyp/Unknown Prot			unknown
4D5	AJ273289	9	Hyp/Unknown Prot	EAA35738	Neurospora crassa	hypothetical protein
4D7	AJ273291	3a	Ribosomal proteins	CAB93015	Schizosaccharomyces pombe	60s ribosomal protein l21
4D9	AJ273295	9	Hyp/Unknown Prot			unknown
4D11	AJ273299	1a	Aminoacid metabolism	CAB63543	Schizosaccharomyces pombe	putative aminotransferase class-III pyridoxal-phosphate
5D1	AJ273476	3b	Translation	BAA19665	Homo sapiens	KIAA0235 protein, putative translational repressor
5D3	AJ273478	9	Hyp/Unknown Prot	EAA77237	Gibberella zeae PH-1	hypothetical protein FG07378
5D5	AJ273481	3c	Proteolysis	CAB63909	Metarhizium anisopliae var. anisopliae	metalloprotease MEP1
5D7	AJ273483	5f	Signaling	AAB05822	Thermomonospora curvata	putative serine/threonine protein kinase; PkwA
5D9	AJ273487	2	Energy Metabolism	AAG06306	Pseudomonas aeruginosa	probable short-chain dehydrogenase
5D11	AJ273493	9	Hyp/Unknown Prot			Unknown
1D2	AJ272754	9	Hyp/Unknown Prot	EAA61491	Aspergillus nidulans	hypothetical protein AN9200.2
1D4	AJ272757	9	Hyp/Unknown Prot	EAA67775	Gibberella zeae PH-1	hypothetical protein FG02545
1D6	AJ272761	9	Hyp/Unknown Prot	EAA69714	Gibberella zeae PH-1	hypothetical protein FG02083
1D8	AJ272765	7c	Secondary metabolites	AAN74815	Gibberella moniliformis	Fum12p, required for fumonisin biosynthesis
1D10	AJ272767	6e	Sexual cycle	AAM95965	Aspergillus nidulans	ESDC, required for sexual development
1D12	AJ272771	9	Hyp/Unknown Prot			unknown
2D2	AJ272917	9	Hyp/Unknown Prot	CAC01464	Streptomyces coelicolor	putative NLP/P60 family protein
2D4	AJ272920	9	Hyp/Unknown Prot	EAA51141	Magnaporthe grisea	predicted protein
2D6	AJ272924	9	Hyp/Unknown Prot	EAA34747	Neurospora crassa	hypothetical protein
2D8	AJ272928	1b	Carbohydrate metabolism	CAD21296	Neurospora crassa	related to cel1 (cellulase) protein precursor
2D10	AJ272930	7c	Secondary metabolites	CAA61605	Metarhizium anisopliae	peptide synthetase
2D12	AJ272932	9	Hyp/Unknown Prot	EAA55296	Magnaporthe grisea	hypothetical protein MG06953.4
3D2	AJ273111	9	Hyp/Unknown Prot			unknown
3D4	AJ273114	1b	Carbohydrate metabolism	CAA70511	Aspergillus tubingensis	ferulic acid esterase A
3D6	AJ273116	3d	Prot modification/targeting	CAA10978	Hypocrea jecorina	protein disulfide isomerase
3D8	AJ273120	9	Hyp/Unknown Prot			Unknown
3D10	AJ273123	3b	Translation	AAC08410	Podospora anserina	translation release factor subunit 1
3D12	AJ273125	5a	Cell wall	NP_013314	Saccharomyces cerevisiae	Protein with similarity to Crh1p, which is a putative glycosidase of the cell wall; Crr1p (cell wall organization and biogenesis)
4D2	AJ273285	9	Hyp/Unknown Prot	EAA67384	Gibberella zeae PH-1	hypothetical protein FG01587
4D4	AJ273288	5d	Transport proteins	CAE76383	Neurospora crassa	related to HOL1 protein, ion transporter
4D6	AJ273290	9	Hyp/Unknown Prot			Unknown

4D8	AJ273292	3b	Translation	CAC04442	Neurospora crassa	probable translation release factor erf3
4D10	AJ273296	7c	Secondary metabolites	AAR90253	Botryotinia fuckeliana	polyketide synthase
4D12	AJ273303	5a	Cell wall	T45525	Kluyveromyces marxianus var. lactis	WSC4 homolog, WSC4p is involved in cell wall integritz and stress response
5D2	AJ273477	9	Hyp/Unknown Prot	CAA97357	Schizosaccharomyces pombe	putative vesicle associated membarne protein
5D4	AJ273480	9	Hyp/Unknown Prot			unknown
5D6	AJ273482	9	Hyp/Unknown Prot	EAA68277	Gibberella zeae PH-1	predicted protein
5D8	AJ273486	9	Hyp/Unknown Prot	EAA76389	Gibberella zeae PH-1	hypothetical protein FG06745
5D10	AJ273489	6a	Cell cycle	S24395	Emericella nidulans	protein-tyrosine-phosphatase, cdc25 homolog nimT (M phase inducer)
5D12	AJ273495	1a	Aminoacid metabolism	EAA31846	Neurospora crassa	probable branched-chain amino acids aminotransferase
1F1	AJ272790	1d	Nucleotide metabolism	AAH14552	Homo sapiens	TPK1 protein: Thiamin pyrophosphokinase, catalytic domain, functioning in nucleotide biosynthesis
1F3	AJ272794	9	Hyp/Unknown Prot	AAK22505	Caulobacter crescentus	conserved hypothetical protein
1F5	AJ272798	9	Hyp/Unknown Prot			unknown
1F7	AJ272803	9	Hyp/Unknown Prot	EAA69263	Gibberella zeae PH-1	hypothetical protein FG00602
1F9	AJ272808	9	Hyp/Unknown Prot			unknown
1F11	AJ272810	6e	Sexual cycle	CAC05511	Schizosaccharomyces pombe	sexual differentiation process protein isp4
2F1	AJ272955	6e	Sexual cycle	BAA32410	Neurospora crassa	krev-1, RAS superfamily, involved in sexual cycle
2F3	AJ272958	9	Hyp/Unknown Prot			unknown
2F5	AJ272960	6b	Chromosome structure	B35072	Saccharomyces cerevisiae	nonhistone chromosomal protein NHP6B
2F7	AJ272962	9	Hyp/Unknown Prot			unknown
2F9	AJ272969	3d	Prot modification/targeting	AAA18831	Saccharomyces cerevisiae	Vps10p (putative vacuolar protein sorting)
2F11	AJ272971	3b	Translation	BAA31571	Schizosaccharomyces pombe	elongation factor 1b
3F1	AJ273151	3a	Ribosomal proteins	CAA65433	Podospora anserina	ribosomal protein S7
3F3	AJ273157	1a	Aminoacid metabolism	NP_010753	Saccharomyces cerevisiae	protein arginine methyltransferase, Rmt2
3F5	AJ273162	6d	Cell polarity	NP_014106	Saccharomyces cerevisiae	GTPase-activating multicopy Suppressor of Bud Emergence; Msb3p
3F7	AJ273164	3a	Ribosomal proteins	AAC08586	Yarrowia lipolytica	40S ribosomal protein S4 (S7)
3F9	AJ273167	5e	Cellular transportation	CAA96756	Saccharomyces cerevisiae	14 KDa protein found on ER-derived vesicles; Erv14p
3F11	AJ273171	4c	Transcription	AAG09789	Candida albicans	repressed by TUP1 protein 4; Rbt4p
4F1	AJ273330	3b	Translation	CAA78282	Candida albicans	translation elongation factor eEF-3
4F3	AJ273342	2	Energy Metabolism	AAB09569	Dictyostelium discoideum	similar to Na+,K+ ATPase; IonA
4F5	AJ273345	9	Hyp/Unknown Prot	EAA76296	Gibberella zeae PH-1	hypothetical protein FG09507
4F7	AJ273352	9	Hyp/Unknown Prot			Unknown
4F9	AJ273357	9	Hyp/Unknown Prot	EAA70688	Gibberella zeae PH-1	hypothetical protein FG00742

4F11	AJ273364	5a	Cell wall	EAA31914	Neurospora crassa	related to secretion protein rsec8
5F1	AJ273512	1e	N-, P-, S-metabolism	BAA86934	Aspergillus oryzae	glutaminase A
5F3	AJ273514	6b	Chromosome structure	CAA25760	Neurospora crassa	histone H4
5F5	AJ273516	6g	Cell death	CAD70452	Neurospora crassa	related to protein involved in authophagy (APG17)
5F7	AJ273518	3a	Ribosomal proteins	T41535	Schizosaccharomyces pombe	60S ribosomal protein L46
5F9	AJ273523	1a	Aminoacid metabolism	AAF97598	Acremonium chrysogenum	cystathionine-gamma-lyase
5F11	AJ273528	9	Hyp/Unknown Prot			unknown
1F2	AJ272793	2	Energy Metabolism	AAC64860	Kluyveromyces lactis	F1F0-ATP synthase subunit 4
1F4	AJ272796	5f	Signaling	AAD02822	Cryphonectria parasitica	mitogen-activated protein kinase kinase; CPK1
1F6	AJ272799	5a	Cell wall	AAC35942	Aspergillus fumigatus	GEL1 protein
1F8	AJ272805	9	Hyp/Unknown Prot			unknown
1F10	AJ272809	1a	Aminoacid metabolism	CAB91256	Neurospora crassa	putative ornithine carbamoyl transferase precursor
1F12	AJ272811	5d	Transport proteins	CAB96004	Schizosaccharomyces pombe	mitochondrial carrier protein; yeast yor222w homolog
2F2	AJ272957	3b	Translation	NP_009365	Saccharomyces cerevisiae	96 kDa protein; Fun12p
2F4	AJ272959	9	Hyp/Unknown Prot	EAA75204	Gibberella zeae PH-1	hypothetical protein FG05633
2F6	AJ272961	9	Hyp/Unknown Prot			unknown
2F8	AJ272967	4c	Transcription	AAA96824	Fusarium solani f. sp. pisi	cutinase transcription factor 1
2F10	AJ272970	9	Hyp/Unknown Prot	EAA48020	Magnaporthe grisea	hypothetical protein MG09150.4
2F12	AJ272972	2	Energy Metabolism	BAA19073	Aspergillus niger	NADP-dependent isocitrate dehydrogenase precursor
3F2	AJ273155	4a	RNA metabolism	NP_014980	Saccharomyces cerevisiae	cytoplasmic alanyl-tRNA synthetase gene; Ala1p
3F4	AJ273159	9	Hyp/Unknown Prot	EAA76041	Gibberella zeae PH-1	hypothetical protein FG07068
3F6	AJ273163	9	Hyp/Unknown Prot	NP_011782	Saccharomyces cerevisiae	hypothetical protein, Ygr266wp
3F8	AJ273165	4c	Transcription	NP_015196	Saccharomyces cerevisiae	transcription initiation factor TFIIF small subunit; Anc1p
3F10	AJ273170	1c	Lipid metabilism	NP_010570	Saccharomyces cerevisiae	diacylglycerol Pyrophosphate Phosphatase; Dpp1p
SSC4						
4F2	AJ273336	9	Hyp/Unknown Prot			unknown
4F4	AJ273344	9	Hyp/Unknown Prot	EAA73009	Gibberella zeae PH-1	hypothetical protein FG08048
4F6	AJ273346	9	Hyp/Unknown Prot	EAA26657	Neurospora crassa	predicted protein
4F8	AJ273356	5f	Signaling	CAC07966	Leishmania mexicana	putative mitogen-activated protein kinase kinase 2
SSC5						
4F12	AJ273365	9	Hyp/Unknown Prot	EAA72298	Gibberella zeae PH-1	hypothetical protein FG04096
5F2	AJ273513	5e	Cellular transportation	NP_593476	Schizosaccharomyces pombe	putative COPII-coated vesicle proteins; emp24-like
5F4	AJ273515	7c	Secondary metabolites	Q12609	Emericella nidulans	probable sterigmatocystin biosynthesis P450 monoxygenase; Stcf
5F6	AJ273517	5e	Cellular transportation	CAB77004	Schizosaccharomyces pombe	similar to yeast Synaptobrevin (v-SNARE) homolog; Bos1
5F8	AJ273519	9	Hyp/Unknown Prot			unknown
5F10	AJ273526	9	Hyp/Unknown Prot	EAA70677	Gibberella zeae PH-1	hypothetical protein FG00731

5F12	AJ273529	3a	Ribosomal proteins	CAA20680	Schizosaccharomyces pombe	60S ribosomal protein L19B
1H1	AJ272830	5d	Transport proteins	AAA80167	Candida albicans	peptide transporter
1H3	AJ272833	2	Energy Metabolism	AAF82789	Cladosporium fulvum	aldehyde dehydrogenase
1H5	AJ272836	6a	Cell cycle	P25694	Saccharomyces cerevisiae	cell cycle protein Cdc48p, transitional endoplasmic reticulum ATPase
1H7	AJ272838	9	Hyp/Unknown Prot			unknown
1H9	AJ272840	1b	Carbohydrate metabolism	BAB12235	Aspergillus oryzae	phosphoglucomutase
1H11	AJ272845	9	Hyp/Unknown Prot			unknown
2H1	AJ273000	5d	Transport proteins	NP_013231	Saccharomyces cerevisiae	low-affinity zinc transport protein; Zrt2p
2H3	AJ273010	2	Energy Metabolism	S13025	Neurospora crassa	NADH dehydrogenase (ubiquinone) 40K chain
2H5	AJ273016	9	Hyp/Unknown Prot			unknown
2H7	AJ273021	9	Hyp/Unknown Prot	EAA77550	Gibberella zeae PH-1	hypothetical protein FG07317
2H9	AJ273029	6a	Cell cycle	CAB90139	Schizosaccharomyces pombe	cell division control protein 5; Myb family DNA-binding
2H11	AJ273036	7a	Stress response	AAR36902	Chaetomium globosum	small heat shock protein
3H1	AJ273194	9	Hyp/Unknown Prot	AAD34105	Homo sapiens	CGI-110 protein
3H3	AJ273201	9	Hyp/Unknown Prot	EAA73888	Gibberella zeae PH-1	hypothetical protein FG05175
3H5	AJ273204	9	Hyp/Unknown Prot			unknown
3H7	AJ273210	3b	Translation	S55900	Schizosaccharomyces pombe	Psi protein, DNAJ-like protein homolog
3H9	AJ273213	5e	Cellular transportation	CAA91773	Schizosaccharomyces pombe	SPAC24B11.08c: COPII-coated vesicle component; involved in ER to golgi transport (predicted); involved in the membrane fusion stage of transport
3H11	AJ273215	9	Hyp/Unknown Prot	EAA51141	Magnaporthe grisea	predicted protein
4H1	AJ273391	9	Hyp/Unknown Prot	EAA70078	Gibberella zeae PH-1	hypothetical protein FG10235
4H3	AJ273393	4c	Transcription	AAB37245	Neurospora crassa	transcriptional repressor rco-1
4H5	AJ273396	5d	Transport proteins	CAA88589	Mus musculus	ATP-binding cassette transporter, sub-family D (ALD)
4H7	AJ273400	9	Hyp/Unknown Prot	EAA69429	Gibberella zeae PH-1	hypothetical protein FG02262
4H9	AJ273405	1b	Carbohydrate metabolism	ZP_00013678	Rhodospirillum rubrum	Glycerophosphoryl diester phosphodiesterase
4H11	AJ273408	9	Hyp/Unknown Prot			Unknown
5H1	AJ273562	1b	Carbohydrate metabolism	ZP_00213858	Burkholderia cepacia R18194	COG3618: Predicted metal-dependent hydrolase of the TIM-barrel fold
5H3	AJ273564	9	Hyp/Unknown Prot	EAA78620	Gibberella zeae PH-1	hypothetical protein FG11307
5H5	AJ273568	6e	Sexual cycle	CAE76537	Neurospora crassa	probable isp4 protein: sexual differentiation process protein
5H7	AJ273573	3d	Prot modification/targeting	CAE85618	Neurospora crassa	probable positive effector protein GCN20
5H9	AJ273577	5a	Cell wall	BAD01559	Aspergillus kawachii	cell wall protein
5H11	AJ273586	9	Hyp/Unknown Prot	EAA72097	Gibberella zeae PH-1	hypothetical protein FG08520
1H2	AJ272832	9	Hyp/Unknown Prot	EAA54957	Magnaporthe grisea	hypothetical protein MG05748.4
1H4	AJ272835	9	Hyp/Unknown Prot			unknown
1H6	AJ272837	9	Hyp/Unknown Prot			unknown
1H8	AJ272839	9	Hyp/Unknown Prot			unknown

1H10	AJ272841	9	Hyp/Unknown Prot	EAA30143	Neurospora crassa	predicted protein
1H12	AJ272846	1a	Aminoacid metabolism	NP_825573	Streptomyces avermitilis	putative dioxygenase
2H2	AJ273004	4a	RNA metabolism	NP_915251	Oryza sativa	similar to methionyl-tRNA synthetase
2H4	AJ273011	5a	Cell wall	CAA05375	Trichoderma harzianum	b-1,3 exoglucanase
2H6	AJ273019	3c	Proteolysis	AAD00154	Metarhizium anisopliae	ubiquitin conjugating enzyme
2H8	AJ273026	2	Energy Metabolism	CAC18180	Neurospora crassa	conserved hypothetical protein
2H10	AJ273034	9	Hyp/Unknown Prot	EAA57352	Magnaporthe grisea	hypothetical protein MG08321.4
2H12	AJ273037	9	Hyp/Unknown Prot	EAA67589	Gibberella zeae PH-1	hypothetical protein FG01206
3H2	AJ273196	5f	Signaling	CAD21237	Neurospora crassa	probable GTP-binding protein Drab11: RAS-related signalling pathway
3H4	AJ273203	9	Hyp/Unknown Prot			unknown
3H6	AJ273205	1b	Carbohydrate metabolism	CAA67930	Candida albicans	glycolipid 2-a-mannosyltransferase (a-1,2-mannosyltransferase)
3H8	AJ273212	9	Hyp/Unknown Prot			unknown
3H10	AJ273214	3a	Ribosomal proteins	CAA20689	Schizosaccharomyces pombe	60S ribosomal protein L18
3H12	AJ273217	9	Hyp/Unknown Prot	EAA69070	Gibberella zeae PH-1	hypothetical protein FG02385
4H2	AJ273392	9	Hyp/Unknown Prot			unknown
4H4	AJ273394	9	Hyp/Unknown Prot	EAA75010	Gibberella zeae PH-1	hypothetical protein FG10753
4H6	AJ273398	5d	Transport proteins	NP_009333	Saccharomyces cerevisiae	putative permease; Seo1p
4H8	AJ273403	2	Energy Metabolism	CAC27837	Gibberella fujikuroi	NAD+ dependent glutamate dehydrogenase
4H10	AJ273407	9	Hyp/Unknown Prot			Unknown
4H12	AJ273409	9	Hyp/Unknown Prot			unknown
5H2	AJ273563	5d	Transport proteins	CAB65616	Schizosaccharomyces pombe	MFS transporter of unknown specificity
5H4	AJ273567	6g	Cell death	AAN41258	Podospora anserina	IDI-7: autophagy protein induced by cell death
5H6	AJ273569	9	Hyp/Unknown Prot	EAA69527	Gibberella zeae PH-1	hypothetical protein FG00976
5H8	AJ273575	9	Hyp/Unknown Prot	EAA31030	Neurospora crassa	predicted protein
5H10	AJ273581	9	Hyp/Unknown Prot			unknown
5H12	AJ273587	9	Hyp/Unknown Prot			unknown
6B1	AJ273610	9	Hyp/Unknown Prot	EAA75645	Gibberella zeae PH-1	hypothetical protein FG06000
6B3	AJ273615	9	Hyp/Unknown Prot			unknown
6B5	AJ273617	9	Hyp/Unknown Prot			unknown
6B7	AJ273622	5e	Cellular transportation	BAA84625	Oryza sativa	hypothetical protein, similar to syntaxin related protein; AtVam3p
6B9	AJ273625	9	Hyp/Unknown Prot			unknown
6B11	AJ273629	5d	Transport proteins	AAS68344	Emericella nidulans	exportin 1
7B1	AJ273817	9	Hyp/Unknown Prot			unknown
7B3	AJ273822	5b	Cytosqueloton	AAC83184	Yarrowia lipolytica	actin binding protein Sla2p
7B5	AJ273828	5c	Extracellular Matrix/Cell wall protein	T37645	Schizosaccharomyces pombe	hypothetical serine rich protein
7B7	AJ273832	5b	Cytosqueloton	AAH63355	Xenopus tropicalis	Thioredoxin domain containing protein 5
7B9	AJ273835	5f	Signaling	CAA22592	Schizosaccharomyces pombe	SPAC637: with Pleckstrin homology domain, found in eukaryotic signalling proteins

7B11	AJ273839	1b	Carbohydrate metabolism	CAA64380	Candida albicans	glucosamine fructose-6-phosphate aminotransferase
8B1	AJ274022	9	Hyp/Unknown Prot	EAA48254	Magnaporthe grisea	hypothetical protein MG10317.4
8B3	AJ274027	6d	Cell polarity	NP_055063	Homo sapiens	RING box protein 1, regulator of cullins 1
8B5	AJ274031	6a	Cell cycle	NP_009872	Saccharomyces cerevisiae	involved in cell division and spore germination; Krr1p
8B7	AJ274033	2	Energy Metabolism	AAG03537	Pseudomonas aeruginosa	probable oxidoreductase
8B9	AJ274037	9	Hyp/Unknown Prot	AAG03445	Pseudomonas aeruginosa	hypothetical protein
8B11	AJ274041	3c	Proteolysis	AAC98480	Homo sapiens	lysosomal pepstatin insensitive protease
9B1	AJ274207	9	Hyp/Unknown Prot			unknown
9B3	AJ274214	4c	Transcription	CAC88374	Hypocrea jecorina	transcription factor
9B5	AJ274216	5a	Cell wall	T40872	Schizosaccharomyces pombe	probable cell wall biogenesis protein, yeast Ecm33 homolog
9B7	AJ274219	1c	Lipid metabilism	T40392	Schizosaccharomyces pombe	probable 3-b-hydroxysteroid dehydrogenase/d 5-4-isomerase
9B9	AJ274223	9	Hyp/Unknown Prot	EAA72792	Gibberella zeae PH-1	hypothetical protein FG04411
9B11	AJ274226	3c	Proteolysis	AAB68600	Metarhizium anisopliae	carboxypeptidase
10B1	CN807950	2	Energy Metabolism	AAQ74977	Podospora anserina	NADPH oxidase isoform 2
10B3	CN807925	5b	Cytosqueloton	NP_013441	Saccharomyces cerevisiae	Involved in cytoskeletal organization and cellular growth; Vrp1p
10B5	CN807959	1a	Aminoacid metabolism	CAD70830	Neurospora crassa	related to phosphatidylserine decarboxylase
10B7	CN807972	1c	Lipid metabilism	CAD70918	Neurospora crassa	probable fatty acid elongase (FEN1)
10B9	CN807975	9	Hyp/Unknown Prot	EAA76359	Gibberella zeae PH-1	hypothetical protein FG06837
10B11	CN807989	5f	Signaling	AAD01641	Magnaporthe grisea	pathogenicity transmembrance protein
6B2	AJ273614	4c	Transcription	NP_014121	Saccharomyces cerevisiae	CCR4 Associated Factor 120 kDa; Caf120p (regulation of transcription from Pol II promoter)
6B4	AJ273616	9	Hyp/Unknown Prot	EAA71198	Gibberella zeae PH-1	hypothetical protein FG04177
6B6	AJ273620	1e	N-, P-, S-metabolism	S57160	Saccharomyces cerevisiae	probable sulfite reductase domain, YJR137c
6B8	AJ273623	1b	Carbohydrate metabolism	CAB82861	Phaeosphaeria avenaria	b-glucosidase
6B10	AJ273627	9	Hyp/Unknown Prot			unknown
6B12	AJ273630	1b	Carbohydrate metabolism	AAF34579	Hypocrea jecorina	1,2-a-D-mannosidase
7B2	AJ273819	6b	Chromosome structure	CAF05892	Neurospora crassa	related to nucleolar 100K polymyositis-scleroderma protein
7B4	AJ273824	6b	Chromosome structure	CAA25761	Neurospora crassa	histone H3
7B6	AJ273829	7b	Detoxification	CAA21951	Candida albicans	rehydrin-like protein
7B8	AJ273834	1b	Carbohydrate metabolism	BAA05869	Gluconacetobacter liquefaciens	L-sorbosone dehydrogenase
7B10	AJ273837	9	Hyp/Unknown Prot	EAA73009	Gibberella zeae PH-1	hypothetical protein FG08048
7B12	AJ273840	9	Hyp/Unknown Prot	EAA76679	Gibberella zeae PH-1	hypothetical protein FG09360
8B2	AJ274024	3d	Prot modification/targeting	AAC31955	Drosophila melanogaster	ER lumen protein retaining receptor

8B4	AJ274029	5c	Extracellular Matrix/Cell wall protein	BAA90540	Pinctada maxima	N66 matrix protein
8B6	AJ274032	3d	Prot modification/targeting	AAC37215	Acanthamoeba castellanii	protein disulfide isomerase (PDI) like protein
8B8	AJ274036	4a	RNA metabolism	CAB91819	Neurospora crassa	arginine-tRNA ligase
8B10	AJ274040	3a	Ribosomal proteins	CAA72204	Blumeria graminis	60S ribosomal protein L29 (L27A)
8B12	AJ274045	9	Hyp/Unknown Prot	EAA76426	Gibberella zeae PH-1	hypothetical protein FG06966
9B2	AJ274212	9	Hyp/Unknown Prot	EAA60601	Aspergillus nidulans FGSC A4	hypothetical protein AN8808.2
9B4	AJ274215	9	Hyp/Unknown Prot	EAA70752	Gibberella zeae PH-1	hypothetical protein FG00806
9B6	AJ274217	9	Hyp/Unknown Prot			unknown
9B8	AJ274221	1e	N-, P-, S-metabolism	AAF28889	Aspergillus terreus	3'phosphoadenylyl thiosulfotransferase
9B10	AJ274225	9	Hyp/Unknown Prot	EAA69710	Gibberella zeae PH-1	hypothetical protein FG02079
9B12	AJ274227	1b	Carbohydrate metabolism	CAB91484	Neurospora crassa	probable oxoglutarate dehydrogenase precursor
10B2	CN807951	9	Hyp/Unknown Prot			Unknown
10B4	CN807958	9	Hyp/Unknown Prot			Unknown
10B6	CN807971	2	Energy Metabolism	CAD21046	Neurospora crassa	ubiquinol-cytochrome C reductase complex core protein 2 precursor
10B8	CN807974	1a	Aminoacid metabolism	CAF05873	Neurospora crassa	glycine hydroxymethyltransferase, cytosolic
10B10	CN808640	6e	Sexual cycle	CAD21276	Neurospora crassa	required for sexual development
10B12	CN808160	3b	Translation	AAR16425	Metarhizium anisopliae	translation elongation factor 1 alpha
6D1	AJ273660	3a	Ribosomal proteins	NP_014332	Saccharomyces cerevisiae	ribosomal protein L9B (L8B) (rp24)
6D3	AJ273662	7a	Stress response	AAA33589	Neurospora crassa	30 kDa heat shock protein
6D5	AJ273664	1f	Cofactor and vitamins	NP_014819	Saccharomyces cerevisiae	ferrochelatase (protoheme ferrolyase); Hem15p
6D7	AJ273669	1b	Carbohydrate metabolism	BAA09832	Rhodotorula minuta	isobutene-forming enzyme and benzoate 4-hydroxylase
6D9	AJ273674	9	Hyp/Unknown Prot	EAA68477	Gibberella zeae PH-1	hypothetical protein FG00464
6D11	AJ273678	3c	Proteolysis	S67665	Saccharomyces cerevisiae	ubiquitin-specific proteinase; Ubp1
7D1	AJ273870	6g	Cell death	BAB33421	Pisum sativum	putative senescence-associated protein
7D3	AJ273874	1a	Aminoacid metabolism	CAA04757	Rhodotorula graminis	D-mandelate dehydrogenase
7D5	AJ273876	3b	Translation	CAB11076	Schizosaccharomyces pombe	probable eukaryotic translation initiation factor 2 b
7D7	AJ273878	1a	Aminoacid metabolism	CAA58037	Hordeum vulgare	probable coproporphyrinogen oxidase precursor
7D9	AJ273883	1e	N-, P-, S-metabolism	NP_015190	Saccharomyces cerevisiae	iron-sulfur cluster nifU-like protein; Isu1p
7D11	AJ273887	1a	Aminoacid metabolism	CAB91739	Neurospora crassa	probable glutamate decarboxylase
8D1	AJ274068	6f	Clock control/light response	CAA63964	Neurospora crassa	white collar-1 (wc-1), regulator of blue light responses
8D3	AJ274072	4b	RNA modification	NP_596034	Schizosaccharomyces pombe	putative splicing factor
8D5	AJ274080	9	Hyp/Unknown Prot			unknown

8D7	AJ274086	5e	Cellular transportation	AAA91301	Emericella nidulans	nuclear migration protein NudF
8D9	AJ274088	1f	Cofactor and vitamins	ZP_00139273	Pseudomonas aeruginosa	Conserved protein/domain typically associated with flavoprotein oxygenases, DIM6/NTAB family
8D11	AJ274092	9	Hyp/Unknown Prot			unknown
9D1	AJ274252	5e	Cellular transportation	T43682	Schizosaccharomyces pombe	nucleoporin
9D3	AJ274266	1a	Aminoacid metabolism	T11611	Schizosaccharomyces pombe	probable X-Pro aminopeptidase
9D5	AJ274269	9	Hyp/Unknown Prot	EAA74631	Gibberella zeae PH-1	conserved hypothetical protein
9D7	AJ274274	4a	RNA metabolism	CAD21174	Neurospora crassa	related to arginine-tRNA-protein transferase
9D9	AJ274278	5e	Cellular transportation	CAA21224	Schizosaccharomyces pombe	protein transport protein sec23 homolog
9D11	AJ274285	3a	Ribosomal proteins	AAB02772	Neurospora crassa	putative ribosome-associated protein
10D1	CN808036	1d	Nucleotide metabolism	XP_372959	Homo sapiens	similar to rRNA intron-encoded homing endonuclease
10D3	CN808047	7c	Secondary metabolites	ZP_00087429	Pseudomonas fluorescens	Isopenicillin N synthase and related dioxygenases
10D5	CN808052	1c	Lipid metabilism	AAR23260	Chaetomium globosum	dienelactone hydrolase
10D7	CN808056	1a	Aminoacid metabolism	NP_009590	Saccharomyces cerevisiae	Nuclear SAM-dependent mono- and asymmetric arginine dimethylating methyltransferase
10D9	CN808059	9	Hyp/Unknown Prot			Unknown
10D11	CN808061	2	Energy Metabolism	P23704	Neurospora crassa	ATP synthase beta chain, mitochondrial precursor
6D2	AJ273661	9	Hyp/Unknown Prot			unknown
6D4	AJ273663	3c	Proteolysis	CAB44651	Metarhizium anisopliae	chymotrypsin
6D6	AJ273668	4a	RNA metabolism	T38536	Schizosaccharomyces pombe	probable DNA-directed RNA polymerase I 49K chain
6D8	AJ273671	9	Hyp/Unknown Prot	CAB92824	Streptomyces coelicolor	hypothetical protein
6D10	AJ273675	9	Hyp/Unknown Prot	EAA70572	Gibberella zeae PH-1	hypothetical protein FG01263
6D12	AJ273680	1c	Lipid metabilism	NP_827376	Streptomyces avermitilis	NADH-dependent, putative enoyl-ACP reductase II
7D2	AJ273873	1f	Cofactor and vitamins	CAB11664	Schizosaccharomyces pombe	probable thiamine biosynthetic bifunctional enzyme
7D4	AJ273875	2	Energy Metabolism	PXNCV7	Neurospora crassa	vacuolar H+-transporting ATPase 67K chain
7D6	AJ273877	1d	Nucleotide metabolism	DAA01288	Danio rerio	TPA: replicase/helicase/endonuclease
7D8	AJ273882	9	Hyp/Unknown Prot	EAA75447	Gibberella zeae PH-1	hypothetical protein FG11237
7D10	AJ273884	9	Hyp/Unknown Prot	EAA75666	Gibberella zeae PH-1	predicted protein
7D12	AJ273888	5d	Transport proteins	NP_010034	Saccharomyces cerevisiae	probable sugar transport protein, Ydl247wp
8D2	AJ274069	4b	RNA modification	CAB91802	Neurospora crassa	probable rrm-type RNA binding protein
8D4	AJ274076	1b	Carbohydrate metabolism	ZP_00197580	Mesorhizobium sp. BNC1	Gluconolactonase
8D6	AJ274084	1b	Carbohydrate metabolism	NP_112319	Rattus norvegicus	methylmalonate semialdehyde dehydrogenase gene
8D8	AJ274087	9	Hyp/Unknown Prot	EAK80799	Ustilago maydis 521	hypothetical protein UM00005

8D10	AJ274089	4c	Transcription	NP_782382	Clostridium tetani	transcriptional regulator
8D12	AJ274093	9	Hyp/Unknown Prot			Unknown
9D2	AJ274261	6a	Cell cycle	NP_012056	Schizosaccharomyces pombe	Kontroller Of Growth; Kog1p; guanine nucleotide binding protein
9D4	AJ274267	9	Hyp/Unknown Prot	EAA67175	Gibberella zeae PH-1	hypothetical protein FG00617
9D6	AJ274270	4c	Transcription	CAB11077	Schizosaccharomyces pombe	hypothetical zinc-finger protein
9D8	AJ274277	5c	Extracellular Matrix/Cell wall protein	CAB97468	Neurospora crassa	related to spore coat protein SP96 precursor
9D10	AJ274279	9	Hyp/Unknown Prot			unknown
9D12	AJ274286	1d	Nucleotide metabolism	CAB97316	Neurospora crassa	probable AMP deaminase
10D2	CN808042	1b	Carbohydrate metabolism	XP_328193	Neurospora crassa	Alcohol dehydrogenase I
10D4	CN808048	1c	Lipid metabilism	AAC09237	Neurospora crassa	annexin XIV: calcium-dependent, phospholipid binding protein, inhibit phospolipase activity
10D6	CN808055	5f	Signaling	AAF75278	Neurospora crassa	SOM1 protein: involved in the cAMP-dependent protein kinase pathway controlling growth polarity
10D8	CN808046	5d	Transport proteins	CAB43936	Emericella nidulans	GABA (gamma-amino-n-butyrate) permease
10D10	CN808060	1f	Cofactor and vitamins	AAG23338	Neurospora crassa	thiamine biosynthesis protein NMT-1
10D12	CN808045	9	Hyp/Unknown Prot	EAA72664	Gibberella zeae PH-1	hypothetical protein FG08636
6F1	AJ273707	9	Hyp/Unknown Prot			unknown
6F3	AJ273710	7a	Stress response	BAA33011	Fusarium oxysporum	flavohemoglobin
6F5	AJ273712	9	Hyp/Unknown Prot	EAA70770	Gibberella zeae PH-1	hypothetical protein FG11630
6F7	AJ273715	9	Hyp/Unknown Prot	EAA76232	Gibberella zeae PH-1	hypothetical protein FG09610
6F9	AJ273723	4a	RNA metabolism	CAA90500	Schizosaccharomyces pombe	tryptophanyl-tRNA synthetase, cytoplasmic
6F11	AJ273731	9	Hyp/Unknown Prot			unknown
7F1	AJ273923	9	Hyp/Unknown Prot	EAA30764	Neurospora crassa	hypothetical protein
7F3	AJ273926	5f	Signaling	AAA96531	Fusarium solani f. sp. pisi	Ran1-like protein kinase
7F5	AJ273929	9	Hyp/Unknown Prot	EAA74303	Gibberella zeae PH-1	hypothetical protein FG10850
7F7	AJ273933	1a	Aminoacid metabolism	NP_014827	Saccharomyces cerevisiae	phosphoserine transaminase; Ser1p
7F9	AJ273940	9	Hyp/Unknown Prot	EAA52448	Magnaporthe grisea	hypothetical protein MG05140.4
7F11	AJ273946	9	Hyp/Unknown Prot	EAA76515	Gibberella zeae PH-1	hypothetical protein FG09623
8F1	AJ274129	1b	Carbohydrate metabolism	AAL09829	Coccidioides posadasii	beta-glucosidase 5
8F3	AJ274132	9	Hyp/Unknown Prot	EAA74903	Gibberella zeae PH-1	hypothetical protein FG11080
8F5	AJ274136	3d	Prot modification/targeting	CAB91698	Neurospora crassa	related to BCS1 protein precursor
8F7	AJ274139	3a	Ribosomal proteins	AAK17096	Emericella nidulans	ribosomal protein L37
8F9	AJ274141	9	Hyp/Unknown Prot	EAA35177	Neurospora crassa	hypothetical protein
8F11	AJ274145	9	Hyp/Unknown Prot			unknown
9F1	AJ274310	5d	Transport proteins	AAA33607	Neurospora crassa	phosphate permease
9F3	AJ274312	3c	Proteolysis	Q96FW1	Homo sapiens	Ubiquitin-specific processing protease OTUB1
9F5	AJ274319	3a	Ribosomal proteins	S49366	Candida albicans	40S ribosomal protein S3AE (S1)

9F7	AJ274321	2	Energy Metabolism	AAA87901	Neurospora crassa	vacuolar ATPase 26 kDa subunit
9F9	AJ274325	6f	Clock control/light response	AAC64287	Neurospora crassa	clock-controlled gene-6 protein
9F11	AJ274327	5a	Cell wall	CAB98243	Neurospora crassa	related to chitinase 3 precursor protein
10F1	CN808111	2	Energy Metabolism	BAA11408	Cylindrocarpon lichenicola	cytochrome P450nor1
10F3	CN808120	9	Hyp/Unknown Prot	EAA71197	Gibberella zeae PH-1	hypothetical protein FG04176
10F5	CN808123	4c	Transcription	P40467	Saccharomyces cerevisiae	Putative 108.8 kDa transcriptional regulatory protein in FKH1-STH1 intergenic region
10F7	CN808129	1a	Aminoacid metabolism	NP_593668	Schizosaccharomyces pombe	serine hydroxymethyltransferase
10F9	CN808141	9	Hyp/Unknown Prot	EAA77491	Gibberella zeae PH-1	hypothetical protein FG07474
10F11	CN808148	1b	Carbohydrate metabolism	CAE76499	Neurospora crassa	related to acetate kinase
6F2	AJ273708	7b	Detoxification	CAE81997	Neurospora crassa	RAD50 homolog uvs-6
6F4	AJ273711	6d	Cell polarity	NP_593899	Schizosaccharomyces pombe	possibly involved in cell polarity; by similarity to yeast rax2
6F6	AJ273713	1a	Aminoacid metabolism	AAG13306	Gillichthys mirabilis	cysteine dioxygenase
6F8	AJ273722	6g	Cell death	T37963	Schizosaccharomyces pombe	caffeine-induced death protein 1 Cid1
6F10	AJ273729	9	Hyp/Unknown Prot	EAA69638	Gibberella zeae PH-1	hypothetical protein FG00378
6F12	AJ273732	9	Hyp/Unknown Prot	EAA67284	Gibberella zeae PH-1	hypothetical protein FG01855
7F2	AJ273925	9	Hyp/Unknown Prot	EAA67175	Gibberella zeae PH-1	hypothetical protein FG00617
7F4	AJ273928	9	Hyp/Unknown Prot	EAA72147	Gibberella zeae PH-1	hypothetical protein FG08359
7F6	AJ273930	9	Hyp/Unknown Prot	EAA70448	Gibberella zeae PH-1	hypothetical protein FG00855
7F8	AJ273935	3a	Ribosomal proteins	CAB88562	Neurospora crassa	probable ribosomal protein l13a
7F10	AJ273943	9	Hyp/Unknown Prot			unknown
7F12	AJ273948	7a	Stress response	NP_006251	Homo sapiens	protein-kinase, interferon-inducible double stranded RNA dependent, DnaJ (Hsp40) homolog, subfamily C, member 3
8F2	AJ274130	9	Hyp/Unknown Prot			unknown
8F4	AJ274133	1c	Lipid metabilism	CAB63910	Metarhizium anisopliae var. anisopliae	Esterase; STE1
8F6	AJ274137	7b	Detoxification	CAB71850	Streptomyces coelicolor	possible arsenic resistance membrane transport protein
8F8	AJ274140	9	Hyp/Unknown Prot			unknown
8F10	AJ274144	3c	Proteolysis	CAC07219	Metarhizium anisopliae var. anisopliae	subtilisin-like protease Pr1K
8F12	AJ274148	9	Hyp/Unknown Prot			unknown
9F2	AJ274311	2	Energy Metabolism	P23704	Neurospora crassa	ATP synthase b chain, mitochondrial precursor
9F4	AJ274318	4b	RNA modification	T38841	Schizosaccharomyces pombe	probable pre-mRNA splicing factor
9F6	AJ274320	3c	Proteolysis	NP_014604	Saccharomyces cerevisiae	a-type of subunit of 20S proteasome
9F8	AJ274323	1c	Lipid metabilism	AAG44888	Neurospora crassa	YFL034W: with putative lipase activity
9F10	AJ274326	5d	Transport proteins	CAA80308	Trichoderma harzianum	amino-acid permease INDA1
9F12	AJ274328	9	Hyp/Unknown Prot			unknown

10F2	CN808112	7a	Stress response	NP_013061	Saccharomyces cerevisiae	UBI4 locus contains five-six tandem, in-frame copies of ubiquitin ; transcription is induced in response to several stress conditions; Ubi4p
10F4	CN808121	1b	Carbohydrate metabolism	NP_013458	Saccharomyces cerevisiae	Transaldolase, enzyme in the pentose phosphate pathway; Tal1p
10F6	CN808125	1a	Aminoacid metabolism	S50546	Saccharomyces cerevisiae	Sam1p: Adenosylhomocysteinase: S-adenosylhomocysteine catabolism and/or methionine degradation
10F8	CN808135	9	Hyp/Unknown Prot	EAA51320	Magnaporthe grisea	hypothetical protein MG09337.4
10F10	CN808142	6g	Cell death	BAB33421	Pisum sativum	putative senescence-associated protein
10F12	CN808151	8	Transposable Elements	CAA96388	Fusarium poae	gag-like polyprotein
6H1	AJ273771	3b	Translation	S43861	Podospora anserina	translation elongation factor eEF-1 a chain
6H3	AJ273773	1a	Aminoacid metabolism	NP_013752	Saccharomyces cerevisiae	Involved in lysine biosynthesis, oxidative stress protection; Lys7p
6H5	AJ273777	9	Hyp/Unknown Prot			unknown
6H7	AJ273780	3c	Proteolysis	S22387	Metarhizium anisopliae	cuticle-degrading proteinase Pr1A
6H9	AJ273785	9	Hyp/Unknown Prot	EAA34727	Neurospora crassa	hypothetical protein
6H11	AJ273788	9	Hyp/Unknown Prot	EAA32321	Neurospora crassa	predicted protein
7H1	AJ273974	4c	Transcription	AAG02568	Schizosaccharomyces pombe	single-stranded TG1-3 binding protein (enhancer binding)
7H3	AJ273977	5f	Signaling	NP_012806	Saccharomyces cerevisiae	Protein kinase Prr1p
7H5	AJ273985	9	Hyp/Unknown Prot	AAF27107	Arabidopsis thaliana	unknown protein
7H7	AJ273993	3b	Translation	AAD03465	Homo sapiens	translation initiation factor eIF3 p40 subunit
7H9	AJ273996	9	Hyp/Unknown Prot			unknown
7H11	AJ273998	5b	Cytosqueloton	CAA94304	Sordaria macrospora	a-tubulin
8H1	AJ274173	9	Hyp/Unknown Prot			unknown
8H3	AJ274178	9	Hyp/Unknown Prot			unknown
8H5	AJ274181	9	Hyp/Unknown Prot			unknown
8H7	AJ274185	5e	Cellular transportation	CAB52718	Schizosaccharomyces pombe	COPII coated vesicle component sec24 homolog
8H9	AJ274187	9	Hyp/Unknown Prot	EAA70086	Gibberella zeae PH-1	hypothetical protein FG10243
8H11	AJ274189	5e	Cellular transportation	CAB91264	Neurospora crassa	probable transport vesicle fusion protein; SEC17
9H1	AJ274356	9	Hyp/Unknown Prot	EAA32525	Neurospora crassa	hypothetical protein
9H3	AJ274361	9	Hyp/Unknown Prot	CAC28722	Neurospora crassa	conserved hypothetical protein
9H5	AJ274363	9	Hyp/Unknown Prot	EAA71415	Gibberella zeae PH-1	hypothetical protein FG08554
9H7	AJ274366	5a	Cell wall	CAD70866	Neurospora crassa	related to chitinase
9H9	AJ274371	5e	Cellular transportation	AAC18088	Emericella nidulans	coatomer a subunit
9H11	AJ274373	6a	Cell cycle	EAA33274	Neurospora crassa	septin B
10H1	CN808179	4c	Transcription	CAC88374	Hypocrea jecorina	transcription factor
10H3	CN808189	5a	Cell wall	AAF40139	Aspergillus fumigatus	beta (1-3) glucanosyltransferase Gel2p
10H5	CN808193	6b	Chromosome structure	CAE85526	Neurospora crassa	related to nucleosome remodeling complex subunit RSC8
10H7	CN808197	2	Energy Metabolism	BAA11408	Cylindrocarpon lichenicola	cytochrome P450nor1
10H9	CN808199	9	Hyp/Unknown Prot	EAA31122	Neurospora crassa	predicted protein

10H11	CN808201	9	Hyp/Unknown Prot			Unknown
6H2	AJ273772	5d	Transport proteins	JQ0116	Neurospora crassa	phosphate-repressible phosphate permease
6H4	AJ273776	9	Hyp/Unknown Prot	EAA74132	Gibberella zeae PH-1	hypothetical protein FG06022
6H6	AJ273779	7c	Secondary metabolites	S39842	Fusarium scirpi	enniatin synthetase (peptide synthase)
6H8	AJ273781	9	Hyp/Unknown Prot			unknown
6H10	AJ273787	9	Hyp/Unknown Prot	EAA73994	Gibberella zeae PH-1	hypothetical protein FG05029
6H12	AJ273790	9	Hyp/Unknown Prot			unknown
7H2	AJ273975	1c	Lipid metabilism	CAB88598	Neurospora crassa	lanosterol synthase related protein
7H4	AJ273983	9	Hyp/Unknown Prot			unknown
7H6	AJ273986	9	Hyp/Unknown Prot			unknown
7H8	AJ273995	5d	Transport proteins	T40958	Schizosaccharomyces pombe	high affinity copper transporte
7H10	AJ273997	9	Hyp/Unknown Prot			unknown
7H12	AJ273999	9	Hyp/Unknown Prot			unknown
8H2	AJ274175	9	Hyp/Unknown Prot	EAA69788	Gibberella zeae PH-1	hypothetical protein FG10516
8H4	AJ274180	9	Hyp/Unknown Prot			unknown
8H6	AJ274182	1b	Carbohydrate metabolism	AAC98795	Schizosaccharomyces pombe	dolichol monophosphate mannose synthase
8H8	AJ274186	7a	Stress response	AAB97626	Podospora anserina	Mod-E, heat shock protein of the Hsp90 family
8H10	AJ274188	5d	Transport proteins	AAP13095	Emericella nidulans	TmpA : conserved transmembrane protein
8H12	AJ274191	1b	Carbohydrate metabolism	AAF94498	Vibrio cholerae	prpE protein, similar to acetate-CoA ligase
9H2	AJ274360	1d	Nucleotide metabolism	NP_011944	Saccharomyces cerevisiae	Protein involved in decay of mRNA containing nonsense codons; Nmd2p
9H4	AJ274362	1a	Aminoacid metabolism	AAB28355	Neurospora crassa	NAD(+)-specific glutamate dehydrogenase
9H6	AJ274364	5e	Cellular transportation	CAB46767	Schizosaccharomyces pombe	probable coatomer b subunit
9H8	AJ274368	3c	Proteolysis	CAB63908	Metarhizium anisopliae var. anisopliae	subtilisin-like serine protease Pr1J
9H10	AJ274372	1d	Nucleotide metabolism	CAF21551	Corynebacterium glutamicum	putative nucleoside-diphosphate-sugar epimerase
9H12	AJ274374	1b	Carbohydrate metabolism	S74210	Aspergillus niger	glucokinase (Glk)
10H2	CN808187	5a	Cell wall	BAD01559	Aspergillus kawachii	cell wall protein
10H4	CN808192	5f	Signaling	NP_190863	Arabidopsis thaliana	CBS domain-containing protein / octicosapeptide/Phox/Bemp1 (PB1) domain-containing protein
10H6	CN808194	9	Hyp/Unknown Prot			Unknown
10H8	CN808198	1d	Nucleotide metabolism	XP_322797	Neurospora crassa	ribonucleoside-diphosphate reductase large chain (un-24gene)
10H10	CN808200	6f	Clock control/light response	Q01302	Neurospora crassa	Clock-controlled protein 6
10H12	CN808204	7b	Detoxification	AAS60234	Aspergillus fumigatus	squalene epoxidase 1: confering resistance to antifungal terbinafine
11A1	CN808208	9	Hyp/Unknown Prot	EAA70103	Gibberella zeae PH-1	hypothetical protein FG09877
11A3	CN808215	9	Hyp/Unknown Prot	EAA76507	Gibberella zeae PH-1	hypothetical protein FG09615
11A5	CN808218	1f	Cofactor and vitamins	NP_593508	Schizosaccharomyces pombe	putative GTP cyclohydrolase; riboflavin biosynthesis

11A7	CN808225	7a	Stress response	CAA38037	Petunia x hybrida	heat shock protein
11A9	CN808227	5f	Signaling	AAM46085	Candida albicans	putative regulatory protein to increase pseudohyphae formation
11A11	CN808230	9	Hyp/Unknown Prot			Unknown
12A1	CN808463	5c	Extracellular Matrix/Cell wall protein	NP_766665	Bradyrhizobium japonicum	bll0025: 67 kDa Myosin-crossreactive streptococcal antigen
12A3	CN808468	2	Energy Metabolism	CAE09055	Gibberella fujikuroi	cytochrome P450 oxidoreductase
12A5	CN808472	5a	Cell wall	AAO61685	Aspergillus fumigatus	chitinase
12A7	CN808475	9	Hyp/Unknown Prot	EAA67675	Gibberella zeae PH-1	hypothetical protein FG10014
12A9	CN808478	9	Hyp/Unknown Prot	EAA69789	Gibberella zeae PH-1	hypothetical protein FG10517
12A11	CN808484	5f	Signaling	AAA35246	Yarrowia lipolytica	Ras-like GTP-binding protein RYL2
13A1	CN808692	7a	Stress response	NP_011705	Saccharomyces cerevisiae	congo red hypersensitive; Crh1p
13A3	CN808694	1b	Carbohydrate metabolism	EAA62719	Aspergillus nidulans	Acetyl-coenzyme A synthetase (Acetate--CoA ligase)
13A5	CN808698	9	Hyp/Unknown Prot	AAO78637	Bacteroides thetaiotaomicron	conserved hypothetical protein
13A7	CN808703	7a	Stress response	NP_594209	Schizosaccharomyces pombe	putative metal homeostatis protein
13A9	CN808706	9	Hyp/Unknown Prot	EAA57606	Aspergillus nidulans	predicted protein
13A11	CN808711	3b	Translation	CAB90146	Schizosaccharomyces pombe	translation initiation factor tif33
14A1	CN808850	2	Energy Metabolism	NP_625082	Streptomyces coelicolor	putative zinc-binding oxidoreductase
14A3	CN808852	1e	N-, P-, S-metabolism	CAE75707	Neurospora crassa	related to dimethylaniline monooxygenase
14A5	CN808854	7b	Detoxification	NP_565369	Arabidopsis thaliana	putative secretory protein: defense response to pathogen
14A7	CN808858	9	Hyp/Unknown Prot	EAA77418	Gibberella zeae PH-1	hypothetical protein FG09426
14A9	CN808860	1b	Carbohydrate metabolism	AAF97598	Acremonium chrysogenum	cystathionine-gamma-lyase
14A11	CN808863	6a	Cell cycle	S22694	Emericella nidulans	G2/mitotic-specific cyclin B
15A1	CN809004	9	Hyp/Unknown Prot			Unknown
15A3	CN809007	4b	RNA modification	AAD22102	Neurospora crassa	Pad-1: putative RNA splicing factor
15A5	CN809014	7b	Detoxification	CAD70932	Neurospora crassa	related to protein conferring sensitivity to killer toxin
15A7	CN809016	9	Hyp/Unknown Prot	EAA76332	Gibberella zeae PH-1	hypothetical protein FG06599
15A9	CN809021	4c	Transcription	CAC86433	Neurospora crassa	PRO1A C6 Zink-finger protein
15A11	CN809024	7a	Stress response	EAA70431	Gibberella zeae PH-1	Heat shock 70 kDa protein (HSP70)
11A2	CN808213	5c	Extracellular Matrix/Cell wall protein	NP_594446	Schizosaccharomyces pombe	extensin-like; with SH3 Src homology domain
11A4	CN808217	9	Hyp/Unknown Prot			Unknown
11A6	CN808221	1c	Lipid metabilism	Q9UUH4	Schizosaccharomyces pombe	C-4 methyl sterol oxidase
11A8	CN808226	1b	Carbohydrate metabolism	EAA64805	Aspergillus nidulans	3-phytase B precursor (Myo-inositol-hexaphosphate 3-phosphohydrolase B) (3 phytase B) (Myo-inositol hexakisphosphate phosphohydrolase B)
11A10	CN808229	1c	Lipid metabilism	CAC18223	Neurospora crassa	probable sterol C-24 reductase
11A12	CN808235	9	Hyp/Unknown Prot			Unknown

12A2	CN808466	9	Hyp/Unknown Prot	EAA70704	Gibberella zeae PH-1	hypothetical protein FG00758
12A4	CN808470	9	Hyp/Unknown Prot	EAA68009	Gibberella zeae PH-1	hypothetical protein FG01629
12A6	CN808474	5d	Transport proteins	P07921	Kluyveromyces lactis	Lactose permease
12A8	CN808476	5f	Signaling	CAF06101	Neurospora crassa	related to integral membrane protein PTH11: mediates appressorium differentiation in response to inductive substrate cues
12A10	CN808480	6g	Cell death	NP_599221	Rattus norvegicus	preconditioning-inducible gene 1 protein: involved in autophagocytosis during starvation
12A12	CN808485	1b	Carbohydrate metabolism	CAA21255	Schizosaccharomyces pombe	SPBC1709.16c: prodicted aromatic ring-opening dioxygenase
13A2	CN808693	9	Hyp/Unknown Prot			unknown
13A4	CN808696	1b	Carbohydrate metabolism	NP_827376	Streptomyces avermitilis	putative enoyl-ACP reductase II
13A6	CN808700	5d	Transport proteins	NP_077357	Rattus norvegicus	Glycerol kinase; ATP-stimulated glucocorticoid-receptor translocaton promoter (Carbohydrate transport and metabolism)
13A8	CN808704	1a	Aminoacid metabolism	ZP_00103614	Desulfitobacterium hafniense	Threonine aldolase
13A10	CN808708	8	Transposable Elements	CAB56797	Magnaporthe grisea	transposase
13A12	CN808715	5d	Transport proteins	AAN10149	Emericella nidulans	major facilitator MIRB
14A2	CN808851	8	Transposable Elements	AAP31248	Fusarium oxysporum	transposase
14A4	CN808853	1c	Lipid metabilism	XP_326612	Neurospora crassa	lanosterol synthase related protein
14A6	CN808857	5c	Extracellular Matrix/Cell wall protein	CAB91245	Neurospora crassa	related to lustrin A: elastomeric biomineralization protein
14A8	CN808859	5b	Cytosqueloton	AAK69618	Fusarium proliferatum	beta-tubulin
14A10	CN808861	5b	Cytosqueloton	NP_773887	Bradyrhizobium japonicum	hypothetical cytosolic protein
14A12	CN808864	9	Hyp/Unknown Prot	EAA74465	Gibberella zeae PH-1	predicted protein
15A2	CN809006	9	Hyp/Unknown Prot	CAC82536	Agaricus bisporus	putative sugar transporter for sugar uptake
15A4	CN809012	1b	Carbohydrate metabolism	S36838	Yarrowia lipolytica	acetyl-CoA C-acyltransferase (EC 2.3.1.16), peroxisomal
15A6	CN809015	9	Hyp/Unknown Prot			Unknown
15A8	CN809018	1b	Carbohydrate metabolism	NP_713352	Leptospira interrogans serovar lai str. 56601	aldehyde reductase
15A10	CN809022	1b	Carbohydrate metabolism	CAC18252	Neurospora crassa	formate dehydrogenase
15A12	CN809025	9	Hyp/Unknown Prot			Unknown
11C1	CN808255	9	Hyp/Unknown Prot	EAA70850	Gibberella zeae PH-1	hypothetical protein FG02791
11C3	CN808268	9	Hyp/Unknown Prot	EAA67157	Gibberella zeae PH-1	hypothetical protein FG00563
11C5	CN808270	9	Hyp/Unknown Prot	EAA72241	Gibberella zeae PH-1	hypothetical protein FG08651
11C7	CN808272	1c	Lipid metabilism	CAD21081	Neurospora crassa	related to fatty acid hydroxylase
11C9	CN808278	9	Hyp/Unknown Prot			Unknown
11C11	CN808291	1c	Lipid metabilism	P53045	Saccharomyces cerevisiae	C-4 methyl sterol oxidase
12C1	CN808518	5a	Cell wall	BAC82548	Penicillium chrysogenum	cell wall synthesis protein
12C3	CN808527	5a	Cell wall	CAB88654	Neurospora crassa	related to mixed-linked glucanase precursor MLG1

12C5	CN808529	9	Hyp/Unknown Prot			Unknown
12C7	CN808531	9	Hyp/Unknown Prot	EAA50438	Magnaporthe grisea	hypothetical protein MG04197.4
12C9	CN808533	9	Hyp/Unknown Prot			Unknown
12C11	CN808539	5d	Transport proteins	NP_009739	Saccharomyces cerevisiae	Multidrug resistance dityrosine transporter
13C1	CN808740	9	Hyp/Unknown Prot	EAA75228	Gibberella zeae PH-1	hypothetical protein FG05657
13C3	CN808744	9	Hyp/Unknown Prot			Unknown
13C5	CN808749	9	Hyp/Unknown Prot			Unknown
13C7	CN808752	1b	Carbohydrate metabolism	NP_596686	Schizosaccharomyces pombe	acetyl-coa acetyltransferase
13C9	CN808754	5f	Signaling	AAC05299	Homo sapiens	serine kinase SRPK2
13C11	CN808756	5d	Transport proteins	AAK74075	Candida albicans	Golgi GDP-mannose transporter
14C1	CN808882	5b	Cytosqueloton	NP_595574	Schizosaccharomyces pombe	possibly mitochondrial protein synthesis; DUF28 domain
14C3	CN808884	9	Hyp/Unknown Prot	EAA67250	Gibberella zeae PH-1	hypothetical protein FG01872
14C5	CN808886	6b	Chromosome structure	AAC39458	Emericella nidulans	spindle assembly checkpoint protein SLDB
14C7	CN808889	5d	Transport proteins	AAC15999	Arabidopsis thaliana	potassium channel beta subunit homolog
14C9	CN808893	9	Hyp/Unknown Prot	EAA47887	Magnaporthe grisea	predicted protein
14C11	CN808895	1e	N-, P-, S-metabolism	NP_015392	Saccharomyces cerevisiae	Iron Sulfur Assembly -- IscA/NifA homolog; Isa2p
15C1	CN809046	4c	Transcription	NP_594774	Schizosaccharomyces pombe	putative zinc finger protein
15C3	CN809048	1a	Aminoacid metabolism	EAA67392	Gibberella zeae PH-1	homocysteine synthase
15C5	CN809052	3c	Proteolysis	NP_594544	Schizosaccharomyces pombe	putative proteasome component precursor
15C7	CN809057	9	Hyp/Unknown Prot	EAA70789	Gibberella zeae PH-1	hypothetical protein FG04078
15C9	CN809059	4b	RNA modification	CAD21097	Neurospora crassa	related to BRR5 (component of pre-mRNA polyadenylation factor PF I)
15C11	CN809061	1b	Carbohydrate metabolism	CAA36753	Aspergillus niger	benzoate 4-monooxygenase
11C2	CN808266	3c	Proteolysis	AAD55983	Magnaporthe grisea	ubiquitin-conjugating protein
11C4	CN808269	3d	Prot modification/targeting	CAA10978	Hypocrea jecorina	protein disulphide isomerase
11C6	CN808271	1b	Carbohydrate metabolism	EAA73952	Gibberella zeae PH-1	Glyceraldehyde 3-phosphate dehydrogenase (GAPDH)
11C8	CN808274	4c	Transcription	AAB04132	Nectria haematococca	cutinase G-box binding protein: transcription factor
11C10	CN808281	5c	Extracellular Matrix/Cell wall protein	CAB91245	Neurospora crassa	related to lustrin A: matrix protein, antileukoproteinass
11C12	CN808297	1a	Aminoacid metabolism	EAA34408	Neurospora crassa	glutamate decarboxylase [MIPS]
12C2	CN808520	9	Hyp/Unknown Prot	EAA77399	Gibberella zeae PH-1	hypothetical protein FG09407
12C4	CN808528	9	Hyp/Unknown Prot			Unknown
12C6	CN808535	9	Hyp/Unknown Prot			Unknown
12C8	CN808532	3d	Prot modification/targeting	NP_595479	Schizosaccharomyces pombe	putative acid phosphatase
12C10	CN808536	3c	Proteolysis	NP_013493	Saccharomyces cerevisiae	involved in a-factor processing; Ste23p: metallopeptidase activity

12C12	CN808540	9	Hyp/Unknown Prot			Unknown
13C2	CN808741	4c	Transcription	CAC36427	Botryotinia fuckeliana	GATA transcription factor
13C4	CN808746	9	Hyp/Unknown Prot	EAA33203	Neurospora crassa	predicted protein
13C6	CN808750	7a	Stress response	CAA75863	Gibberella fujikuroi	nitrogen metabolite repression-(nmr)-responsible protein
13C8	CN808753	3d	Prot modification/targeting	ZP_00109643	Nostoc punctiforme	Glutathione S-transferase
13C10	CN808755	3a	Ribosomal proteins	T43177	Schizosaccharomyces pombe	ribosomal protein homolog
13C12	CN808757	9	Hyp/Unknown Prot	EAA76359	Gibberella zeae PH-1	hypothetical protein FG06837
14C2	CN808883	3d	Prot modification/targeting	NP_012040	Saccharomyces cerevisiae	putative Upf1p-interacting protein; Nmd3p: ribosomal large subunit assembly and maintenance
14C4	CN808885	9	Hyp/Unknown Prot	EAA69902	Gibberella zeae PH-1	hypothetical protein FG02623
14C6	CN808888	5a	Cell wall	CAC07216	Metarhizium anisopliae	putative endochitinase CHI2
14C8	CN808892	1b	Carbohydrate metabolism	AAL80840	Pyrococcus furiosus	3-hydroxyisobutyrate dehydrogenase
14C10	CN808894	1b	Carbohydrate metabolism	CAE76360	Neurospora crassa	related to ethanol aminephosphotransferase
14C12	CN808897	9	Hyp/Unknown Prot			Unknown
15C2	CN809047	9	Hyp/Unknown Prot	EAA32732	Neurospora crassa	hypothetical protein
15C4	CN809049	5e	Cellular transportation	NP_592960	Schizosaccharomyces pombe	predicted to be involved in vesicular transport and membrane fusion
15C6	CN809056	9	Hyp/Unknown Prot			Unknown
15C8	CN809058	1b	Carbohydrate metabolism	AAL09829	Coccidioides posadasii	beta-glucosidase 5
15C10	CN809060	1b	Carbohydrate metabolism	AAL89587	Aspergillus niger	mannitol-1-phosphate dehydrogenase
15C12	CN809063	5a	Cell wall	CAB98243	Neurospora crassa	related to chitinase 3 precursor protein
11K1	CN808337	9	Hyp/Unknown Prot	EAA69291	Gibberella zeae PH-1	hypothetical protein FG10389
11K3	CN808340	1f	Cofactor and vitamins	CAD71251	Neurospora crassa	spermidine synthase (spe-3)
11K5	CN808343	9	Hyp/Unknown Prot	EAA71881	Gibberella zeae PH-1	hypothetical protein FG08404
11K7	CN808348	7b	Detoxification	CAA04716	Claviceps purpurea	catalase
11K9	CN808352	5a	Cell wall	CAB98243	Neurospora crassa	related to chitinase 3 precursor protein
11K11	CN808354	9	Hyp/Unknown Prot			Unknown
12K1	CN808587	5d	Transport proteins	CAD71001	Neurospora crassa	related to folate transporter/carrier (mitochondrial)
12K3	CN808590	9	Hyp/Unknown Prot			Unknown
12K5	CN808598	1f	Cofactor and vitamins	EAA67183	Gibberella zeae PH-1	NMT1 protein homolog, N-myristoyl transferase
12K7	CN808601	1b	Carbohydrate metabolism	NP_011390	Saccharomyces cerevisiae	Isozyme of methylenetetrahydrofolate reductase
12K9	CN808603	5b	Cytosqueloton	NP_593175	Schizosaccharomyces pombe	coronin CRN1 homolog, cortical actin cytoskeletal component that associates with the Arp2p/Arp3p complex to regulate its activity
12K11	CN808610	9	Hyp/Unknown Prot			Unknown
13K1	CN808780	5f	Signaling	CAB71146	Homo sapiens	putative serine/threonine protein kinase MAK-V

13K3	CN808783	4b	RNA modification	NP_592962	Schizosaccharomyces pombe	putative cleavage and polyadenylation factor
13K5	CN808786	7a	Stress response	NP_594209	Schizosaccharomyces pombe	putative metal homeostatis protein
13K7	CN808789	9	Hyp/Unknown Prot	AAB69310	Metarhizium anisopliae	cystein rich protein
13K9	CN808791	5d	Transport proteins	CAC83067	Podospora anserina	low-affinity copper transporter
13K11	CN808793	9	Hyp/Unknown Prot			unknown
14K1	CN808923	5b	Cytosqueloton	P36147	Saccharomyces cerevisiae	Hypothetical 22.0 Kda protein in LAS1-CCP1 intergenic region
14K3	CN808927	9	Hyp/Unknown Prot	EAA70447	Gibberella zeae PH-1	hypothetical protein FG00854
14K5	CN808929	5d	Transport proteins	CAD37009	Neurospora crassa	related to NCE102 protein: Non-classical export membrance protein
14K7	CN808936	6g	Cell death	AAD53944	Pythium aphanidermatum	25 kDa cell death inducing protein elicitor
14K9	CN808938	9	Hyp/Unknown Prot	EAA55098	Magnaporthe grisea	hypothetical protein MG06755
14K11	CN808942	1a	Aminoacid metabolism	AAL40804	Talaromyces emersonii	prolyl aminopeptidase
15K1	CN809092	1b	Carbohydrate metabolism	O34714	Bacillus subtilis	Oxalate decarboxylase oxdC
15K3	CN809094	5d	Transport proteins	CAB55170	Schizosaccharomyces pombe	SPAP8A3.03: ZIP zinc transporter domain
15K5	CN809097	5d	Transport proteins	NP_592788	Schizosaccharomyces pombe	putative transmembrane channel protein
15K7	CN809101	9	Hyp/Unknown Prot	EAA67299	Gibberella zeae PH-1	hypothetical protein FG10356
15K9	CN809103	9	Hyp/Unknown Prot			Unknown
15K11	CN809105	5d	Transport proteins	NP_010849	Saccharomyces cerevisiae	Ferrioxamine B transporter: induced during iron deprivation and diauxic shift
11K2	CN808339	5f	Signaling	AAK69534	Blumeria graminis	CAP20-like protein: involved in virulence, penetration and appressorium formation
11K4	CN808341	3d	Prot modification/targeting	NP_500392	Caenorhabditis elegans	protein phosphatase 1 (33.3 kD) (4E410)
11K6	CN808346	1a	Aminoacid metabolism	AAB19394	Saccharomyces cerevisiae	aspartate aminotransferase
11K8	CN808349	1b	Carbohydrate metabolism	EAA26806	Neurospora crassa	related to 6-phosphofructo-2-kinase
11K10	CN808353	1b	Carbohydrate metabolism	AAF20155	Botryotinia fuckeliana	cystathionine beta-lyase
11K12	CN808355	9	Hyp/Unknown Prot	EAA73093	Gibberella zeae PH-1	hypothetical protein FG08238
12K2	CN808589	9	Hyp/Unknown Prot	EAA77443	Gibberella zeae PH-1	hypothetical protein FG07426
12K4	CN808593	4b	RNA modification	EAA27582	Neurospora crassa	ribonucleoside-diphosphate reductase large chain (un-24gene)
12K6	CN808599	9	Hyp/Unknown Prot	EAA50930	Magnaporthe grisea 70-15	hypothetical protein MG04689.4
12K8	CN808602	4c	Transcription	ZP_00056870	Thermobifida fusca	Ankyrin repeat, cell surface receptors as well as in intracellular transcription factors where it is involved in DNA binding
12K10	CN808609	4c	Transcription	CAB91681	Neurospora crassa	related to AP-1-like transcription factor
12K12	CN808611	4c	Transcription	NP_593712	Schizosaccharomyces pombe	hypothetical Myb-like DNA-binding protein: transcription factor

13K2	CN808781	9	Hyp/Unknown Prot	AAC59341	Simian virus 40	small tumor anitgen t-ag
13K4	CN808784	9	Hyp/Unknown Prot			Unknown
13K6	CN808788	1a	Aminoacid metabolism	CAD70860	Neurospora crassa	probable glutamate N-acetyltransferase precursor
13K8	CN808790	5b	Cytosqueloton	CAF05895	Neurospora crassa	related to tubulin folding cofactor C
13K10	CN808792	1a	Aminoacid metabolism	AAF33834	Cladosporium fulvum	methionine synthase
13K12	CN808795	3d	Prot modification/targeting	AAK38137	Leishmania major	L344.14 (involved in protein-protein interaction)
14K2	CN808924	5d	Transport proteins	CAB63552	Schizosaccharomyces pombe	SPAC922.05c: with 10 predicted transmembrane helices
14K4	CN808928	1a	Aminoacid metabolism	EAA69962	Gibberella zeae PH-1	Glutamine synthetase (Glutamate--ammonia ligase)
14K6	CN808930	3a	Ribosomal proteins	NP_588435	Schizosaccharomyces pombe	40s ribosomal protein S2
14K8	CN808937	2	Energy Metabolism	P51599	Neurospora crassa	GCH1_NEUCR GTP cyclohydrolase I (GTP-CH-I)
14K10	CN808940	1b	Carbohydrate metabolism	AAD34044	Homo sapiens	CGI-49 protein, Saccharopine dehydrogenase
14K12	CN808943	5e	Cellular transportation	NP_595274	Schizosaccharomyces pombe	beta adaptin-like protein
15K2	CN809093	9	Hyp/Unknown Prot	EAA75072	Gibberella zeae PH-1	hypothetical protein FG06130
15K4	CN809096	9	Hyp/Unknown Prot			Unknown
15K6	CN809098	5d	Transport proteins	CAC18131	Neurospora crassa	probable phosphate transport protein MIR1
15K8	CN809102	9	Hyp/Unknown Prot	EAA33391	Neurospora crassa	hypothetical protein
15K10	CN809104	5d	Transport proteins	NP_010849	Saccharomyces cerevisiae	Ferrioxamine B transporter: induced during iron deprivation and diauxic shift
15K12	CN809107	5d	Transport proteins	CAD71039	Neurospora crassa	related to L-fucose permease
11G1	CN808388	7c	Secondary metabolites	AAO34671	Gibberella zeae	methyltransferase
11G3	CN808395	9	Hyp/Unknown Prot	EAA68704	Gibberella zeae PH-1	hypothetical protein FG00314
11G5	CN808398	5f	Signaling	CAE82003	Neurospora crassa	probable GTP-binding and Ras-related protein ypt5
11G7	CN808411	6a	Cell cycle	EAA31952	Neurospora crassa	probable puff-specific nuclear protein Bx42 (cell cycle control)
11G9	CN808417	6a	Cell cycle	XP_322961	Neurospora crassa	probable cyclin-dependent ser/thr protein kinase KIN28
11G11	CN808421	4c	Transcription	NP_832604	Bacillus cereus	Transcriptional regulator, AraC family
12G1	CN808650	4c	Transcription	CAE85582	Neurospora crassa	probable Transcription initiation factor TFIID
12G3	CN808656	9	Hyp/Unknown Prot	EAA77313	Gibberella zeae PH-1	hypothetical protein FG07941
12G5	CN808662	1c	Lipid metabilism	AAF26280	Aspergillus parasiticus	cytochrome P450 monooxigenase
12G7	CN808665	9	Hyp/Unknown Prot	EAA67953	Gibberella zeae PH-1	hypothetical protein FG02705
12G9	CN808668	9	Hyp/Unknown Prot	NP_176542	Arabidopsis thaliana	hydroxyproline-rich glycoprotein family protein
12G11	CN808670	7b	Detoxification	CAE76158	Neurospora crassa	related to multidrug resistance protein fnx1
13G1	CN808815	6a	Cell cycle	NP_593032	Schizosaccharomyces pombe	cell division cycle related-protein res2/pct1

13G3	CN808817	1a	Aminoacid metabolism	ZP_00031601	Burkholderia fungorum	Threonine dehydrogenase and related Zn-dependent dehydrogenases
13G5	CN808820	1a	Aminoacid metabolism	ZP_00221987	Burkholderia cepacia R1808	Adenosylmethionine-8-amino-7-oxononanoate aminotransferase
13G7	CN808822	9	Hyp/Unknown Prot	XP_330800	Neurospora crassa	hypothetical protein
13G9	CN808824	5f	Signaling	NP_792187	Pseudomonas syringae	ribokinase
13G11	CN808828	7a	Stress response	CAA60962	Saccharomyces cerevisiae	oxidative stress resistance
14G1	CN808962	9	Hyp/Unknown Prot	EAA58728	Aspergillus nidulans	hypothetical protein AN6344
14G3	CN808965	1b	Carbohydrate metabolism	NP_596735	Schizosaccharomyces pombe	adenylosuccinate lyase
14G5	CN808971	1b	Carbohydrate metabolism	CAD21159	Neurospora crassa	acetyl-CoA synthetase
14G7	CN808974	7b	Detoxification	AAF22254	Yarrowia lipolytica	integral peroxisomal membrane protein
14G9	CN808976	1b	Carbohydrate metabolism	JQ0197	Emericella nidulans	4-aminobutyrate transaminase
14G11	CN808978	9	Hyp/Unknown Prot	EAA69291	Gibberella zeae PH-1	hypothetical protein FG10389
15G1	CN809126	9	Hyp/Unknown Prot	AAN68415	Pseudomonas putida	conserved hypothetical protein
15G3	CN809128	3b	Translation	AAO49454	Leptosphaeria maculans	elongation factor 1 beta subunit
15G5	CN809133	2	Energy Metabolism	AAH24673	Mus musculus	NADH dehydrogenase (ubiquinone) 1 alpha subcomplex
15G7	CN809136	9	Hyp/Unknown Prot	EAA76214	Gibberella zeae PH-1	hypothetical protein FG06701
15G9	CN809139	4c	Transcription	CAF32162	Aspergillus fumigatus	transcriptional regulator, putative
15G11	CN809141	9	Hyp/Unknown Prot	EAA75053	Gibberella zeae PH-1	hypothetical protein FG06111
11G2	CN808392	9	Hyp/Unknown Prot	EAA49119	Magnaporthe grisea	hypothetical protein MG00777.4
11G4	CN808396	1b	Carbohydrate metabolism	NP_104264	Mesorhizobium loti	dehydrogenase, (succinatesemialdehyde dehydrogenase, aldehyde dehydrogenase, aldehyde dehydrogenase)
11G6	CN808408	5b	Cytosqueloton	EAA67414	Gibberella zeae PH-1	TBA2_emeni tubulin alpha-2 chain
11G8	CN808412	9	Hyp/Unknown Prot			Unknown
11G10	CN808418	5a	Cell wall	T48815	Neurospora crassa	mixed-linked glucanase precursor (MLG1) related protein
11G12	CN808422	2	Energy Metabolism	BAA11408	Cylindrocarpon lichenicola	cytochrome P450nor1
12G2	CN808652	5b	Cytosqueloton	AAP68979	Gibberella zeae PH-1	beta-tubulin
12G4	CN808657	4c	Transcription	CAB89829	Sordaria brevicollis	C6 zinc finger gene pro1
12G6	CN808663	1d	Nucleotide metabolism	AAA68956	Mastomys hildebrantii	adenine phosphoribosyltransferase
12G8	CN808666	9	Hyp/Unknown Prot	EAA73892	Gibberella zeae PH-1	hypothetical protein FG05179
12G10	CN808669	7b	Detoxification	NP_596231	Schizosaccharomyces pombe	nucleotide excision repair protein
12G12	CN808671	1d	Nucleotide metabolism	AAK62983	Coccidioides immitis	DNA/RNA non-specific nuclease
13G2	CN808816	1b	Carbohydrate metabolism	P77243	Escherichia coli K12	2-methylcitrate dehydratase
13G4	CN808819	9	Hyp/Unknown Prot	EAA48319	Magnaporthe grisea	hypothetical protein MG10578
13G6	CN808821	1d	Nucleotide metabolism	NP_593779	Schizosaccharomyces pombe	putative U1 small nuclear ribonucleoprotein
13G8	CN808823	9	Hyp/Unknown Prot	EAA76030	Gibberella zeae PH-1	hypothetical protein FG06729

13G10	CN808826	7b	Detoxification	BAC07274	Neurospora crassa	MUS42 : DNA repair related protein
13G12	CN808830	1a	Aminoacid metabolism	AAH45269	Xenopus laevis	Aminotransferase class I and II
14G2	CN808963	1e	N-, P-, S-metabolism	BAC55941	Aspergillus oryzae	nitrilase-like protein
14G4	CN808966	9	Hyp/Unknown Prot	EAA61583	Aspergillus nidulans	hypothetical protein AN7795.2
14G6	CN808973	4c	Transcription	CAE76449	Neurospora crassa	related to transcription factor TFIIA-L
14G8	CN808975	2	Energy Metabolism	P07142	Neurospora crassa	Cytochrome c1, heme protein, mitochondrial precursor
14G10	CN808977	4b	RNA modification	AAD22102	Neurospora crassa	Pad-1: putative RNA splicing factor
14G12	CN808979	9	Hyp/Unknown Prot	EAA70113	Gibberella zeae PH-1	hypothetical protein FG09887
15G2	CN809127	6e	Sexual cycle	AAM95965	Aspergillus nidulans	ESDC: required for sexual development
15G4	CN809129	9	Hyp/Unknown Prot	EAA74151	Gibberella zeae PH-1	hypothetical protein FG05089
15G6	CN809135	3c	Proteolysis	CAE76151	Neurospora crassa	matrix AAA protease MAP-1 (mitochondrial):respiratory chain complexes assembly protein
15G8	CN809138	1b	Carbohydrate metabolism	AAO76662	Bacteroides thetaiotaomicron	dioxygenase, related to 2-nitropropane dioxygenase
15G10	CN809140	9	Hyp/Unknown Prot	EAA72664	Gibberella zeae PH-1	hypothetical protein FG08636
15G12	CN809142	1b	Carbohydrate metabolism	AAD37345	Emericella nidulans	AtaAp : acyl transferase homolog
16A1	CN809160	1b	Carbohydrate metabolism	BAC75193	Streptomyces avermitilis	putative L-lactate 2-monooxygenase
16A3	CN809163	5e	Cellular transportation	BAB85760	Fusarium oxysporum	putative mitochondrial carrier protein
16A5	CN809166	9	Hyp/Unknown Prot	EAA51742	Magnaporthe grisea	hypothetical protein MG03337
16A7	CN809169	3d	Prot modification/targeting	NP_588298	Schizosaccharomyces pombe	Glutathione S-transferase I (GST-I)
16A9	CN809173	9	Hyp/Unknown Prot	EAA75528	Gibberella zeae PH-1	hypothetical protein FG05292
16A11	CN809175	6a	Cell cycle	NP_010800	Saccharomyces cerevisiae	Early Meiotic Induction; Emi1p
17A1	CN809322	9	Hyp/Unknown Prot	AAH68035	Xenopus tropicalis	Unknown (protein for MGC:76267)
17A3	CN809325	9	Hyp/Unknown Prot	EAA75574	Gibberella zeae PH-1	hypothetical protein FG05929
17A5	CN809329	4c	Transcription	NP_595082	Schizosaccharomyces pombe	transcription initiation factor iif, beta subunit
17A7	CN809332	9	Hyp/Unknown Prot	EAA77845	Gibberella zeae PH-1	hypothetical protein FG07247
17A9	CN809334	3d	Prot modification/targeting	Q8N448	Homo sapiens	Numb-binding protein 2 (Ligand of Numb-protein X 2), containing zinc finger domain
17A11	CN809336	9	Hyp/Unknown Prot	EAA33203	Neurospora crassa	hypothetical protein
18A1	CN809491	1b	Carbohydrate metabolism	AAM20896	Hypocrea jecorina	L-xylulose reductase
18A3	CN809496	9	Hyp/Unknown Prot	EAA73901	Gibberella zeae PH-1	hypothetical protein FG06042
18A5	CN809499	9	Hyp/Unknown Prot	EAA67627	Gibberella zeae PH-1	hypothetical protein FG01602
18A7	CN809504	9	Hyp/Unknown Prot	EAA74356	Gibberella zeae PH-1	hypothetical protein FG05861
18A9	CN809506	1c	Lipid metabilism	CAF05884	Neurospora crassa	probable phosphatidylinositol/phosphatidylcholine transfer protein
18A11	CN809512	6f	Clock control/light response	CAD70877	Neurospora crassa	clock-controlled gene-6 protein

19A1	AJ298019	1b	Carbohydrate metabolism		Metarhizium anisopliae 2575	neutral Trehalase
19A3	AJ416695	3c	Proteolysis		Metarhizium anisopliae 2575	Pr1A Subtilisin
19A5	AJ400706	3c	Proteolysis		Metarhizium anisopliae 2575	Pr1D Subtilisin
19A7	AJ251967	3c	Proteolysis		Metarhizium anisopliae 2575	Pr1F Subtilisin
EMP						
16A2	CN809162	6a	Cell cycle	CAD70524	Neurospora crassa	related to tol protein: mediator of mating-type-associated vegetative incompatibility
16A4	CN809165	5e	Cellular transportation	NP_594256	Schizosaccharomyces pombe	putative translocation protein
16A6	CN809167	5d	Transport proteins	CAD70319	Neurospora crassa	probable sterol carrier protein
16A8	CN809172	3a	Ribosomal proteins	Q90YV5	Ictalurus punctatus	60S ribosomal protein L13
16A10	CN809174	9	Hyp/Unknown Prot	EAA73700	Gibberella zeae PH-1	hypothetical protein FG06138
16A12	CN809178	5a	Cell wall	AAA20128	Magnaporthe grisea	hydrophobin-like protein
17A2	CN809323	4a	RNA metabolism	NP_596441	Schizosaccharomyces pombe	histidyl-tRNA synthetase, mitochondrial precursor
17A4	CN809328	4c	Transcription	AAF14045	Arabidopsis thaliana	putative MYB family transcription factor
17A6	CN809330	9	Hyp/Unknown Prot	EAA67355	Gibberella zeae PH-1	hypothetical protein FG01848
17A8	CN809333	9	Hyp/Unknown Prot	EAA31331	Neurospora crassa	predicted protein
17A10	CN809335	9	Hyp/Unknown Prot			Unknown
17A12	CN809338	7a	Stress response	P31540	Neurospora crassa	Heat shock protein HSP98
18A2	CN809493	9	Hyp/Unknown Prot	EAA70076	Gibberella zeae PH-1	hypothetical protein FG10233
18A4	CN809497	1b	Carbohydrate metabolism	AAA86932	Emericella nidulans	dihydroorotate dehydrogenase
18A6	CN809502	6a	Cell cycle	AAD39491	Sporothrix schenckii	cyclin-dependent protein kinase
18A8	CN809505	9	Hyp/Unknown Prot	EAA77327	Gibberella zeae PH-1	hypothetical protein FG08969
18A10	CN809507	1b	Carbohydrate metabolism	ZP_00207763	Rhodobacter sphaeroides	Predicted acetamidase/formamidase
18A12	CN809513	1e	N-, P-, S-metabolism	CAF32103	Aspergillus fumigatus	SUN family protein, putative, in nitrate assimilation gene cluster
19A2	AJ251971	5f	Signaling		Metarhizium anisopliae 2575	Adenylate cyclase
19A4	AJ289823	3c	Proteolysis		Metarhizium anisopliae 2575	Pr1B Subtilisin
19A6	AJ251967	3c	Proteolysis		Metarhizium anisopliae 2575	Pr1E Subtilisin
16C1	CN809205	5e	Cellular transportation	CAD21383	Neurospora crassa	related to signal sequence receptor alpha chain
16C3	CN809208	9	Hyp/Unknown Prot	EAA72037	Gibberella zeae PH-1	hypothetical protein FG08863
16C5	CN809210	1f	Cofactor and vitamins	NP_593904	Gibberella zeae PH-1	putative pyridoxal kinase
16C7	CN809212	5f	Signaling	AAN68403	Pseudomonas putida	AMP-binding domain protein
16C9	CN809214	3c	Proteolysis	AAF76202	Sclerotinia sclerotiorum	aspartyl protease
16C11	CN809217	7c	Secondary metabolites	AAK33073	Fusarium sporotrichioides	cytochrome P450 (within trichothecene toxin gene cluster
17C1	CN809368	5a	Cell wall	CAC28732	Neurospora crassa	related to CSI2 protein, involved in chitin synthesis
17C3	CN809371	9	Hyp/Unknown Prot	EAA33225	Neurospora crassa	hypothetical protein

17C5	CN809373	4c	Transcription	CAC80052	Emericella nidulans	PENR2 protein: transcriptional DNA-binding protein
17C7	CN809376	9	Hyp/Unknown Prot	EAA75277	Gibberella zeae PH-1	hypothetical protein FG05460
17C9	CN809385	7b	Detoxification	AAO35087	Clostridium tetani E88	putative sialidase
17C11	CN809387	5f	Signaling	NP_060058	Homo sapiens	WD repeat domain 5 protein; WD-repeat protein 5: G-protein coupled receptor protein signaling pathway
18C1	CN809537	7b	Detoxification	NP_009742	Saccharomyces cerevisiae	Alkaline ceramidase that also has reverse (CoA-independent) ceramide synthase activity, catalyzes both breakdown and synthesis of phytoceramide; overexpression confers fumonisin B1 resistance; Ypc1p
18C3	CN809542	5d	Transport proteins	NP_506393	Caenorhabditis elegans	zinc transporter ZIP
18C5	CN809545	3a	Ribosomal proteins	Q01291	Neurospora crassa	40S ribosomal protein S0
18C7	CN809549	9	Hyp/Unknown Prot	EAA32838	Neurospora crassa	predicted protein
18C9	CN809551	5f	Signaling	EAA67754	Gibberella zeae PH-1	Guanine nucleotide-binding protein beta subunit-like protein (Cross-pathway control WD-repeat protein cpc-2)
18C11	CN809556	9	Hyp/Unknown Prot	CAB65565	Streptomyces coelicolor	putative membrane protein
19C1		7c	Secondary metabolites		Metarhizium anisopliae 2575	putative non-ribosomal peptide synthase 1
19C3		7c	Secondary metabolites		Metarhizium anisopliae 2575	putative non-ribosomal peptide synthase 2B6
19C5		5f	Signaling		Metarhizium anisopliae 2575	putative Egh16 homologue
19C7	AF218207	3a	Ribosomal proteins		Metarhizium anisopliae 2575	18S ribosomal RNA
16C2	CN809207	9	Hyp/Unknown Prot	EAA69513	Gibberella zeae PH-1	hypothetical protein FG00962
16C4	CN809209	9	Hyp/Unknown Prot	EAA75257	Gibberella zeae PH-1	hypothetical protein FG05440
16C6	CN809211	1b	Carbohydrate metabolism	NP_012878	Saccharomyces cerevisiae	Mannosidase, essential glycosylphosphatidylinositol (GPI)-anchored membrane protein required for cell wall biosynthesis, homologous to Dfg5p; Dcw1p
16C8	CN809213	9	Hyp/Unknown Prot			Unknown
16C10	CN809216	5c	Extracellular Matrix/Cell wall protein	AAL47843	Fusarium oxysporum	extracellular matrix protein precursor
16C12	CN809219	5e	Cellular transportation	P35848	Neurospora crassa	Mitochondrial import receptor translocase subunit TOM20
17C2	CN809369	2	Energy Metabolism	CAF32154	Aspergillus fumigatus	NAD-dependant D-isomer specific 2-hydroxyacid dehydrogenase
17C4	CN809372	4c	Transcription	NP_008909	Homo sapiens	zinc finger protein 183 (RING finger, C3HC4 type)
17C6	CN809374	4c	Transcription	BAA95154	Tursiops truncatus	flavocytochrome b558 (NADPH oxidase gp91phox)
17C8	CN809380	7c	Secondary metabolites	CAA41640	Streptomyces coelicolor	actVA 4: actinorhodin biosynthetic gene
17C10	CN809386	6b	Chromosome structure	AAR19291	Neurospora crassa	heterochromatin protein one
17C12	CN809389	9	Hyp/Unknown Prot	EAA51141	Magnaporthe grisea	predicted protein

					70-15	
18C2	CN809540	7c	Secondary metabolites	CAD70509	Neurospora crassa	related to AM-toxin synthetase
18C4	CN809544	9	Hyp/Unknown Prot	EAA59244	Aspergillus nidulans	hypothetical protein AN3935.2
18C6	CN809546	8	Transposable Elements	AAB71689	Magnaporthe grisea	reverse transcriptase (polyA retrotransposons)
18C8	CN809550	1d	Nucleotide metabolism	BAB98936	Corynebacterium glutamicum	Predicted nucleoside-diphosphate-sugar epimerases
18C10	CN809552	6g	Cell death	CAD70452	Neurospora crassa	related to protein involved in authophagy (APG17)
18C12	CN809559	1a	Aminoacid metabolism	CAD10746	Aspergillus niger	aminopeptidase B
19C2		7c	Secondary metabolites		Metarhizium anisopliae 2575	putative non-ribosomal peptide synthase 2B5
19C4		7c	Secondary metabolites		Metarhizium anisopliae 2575	putative non-ribosomal peptide synthase 3D10
19C6		del	del			water
16K1	CN809248	1b	Carbohydrate metabolism	NP_594548	Schizosaccharomyces pombe	putative dehydrogenase
16K3	CN809250	5b	Cytosqueloton	CAD79678	Neurospora crassa	related to p24 protein, involved in membrane trafficking
16K5	CN809253	5a	Cell wall	NP_013314	Saccharomyces cerevisiae	Protein with similarity to Crh1p, which is a putative glycosidase of the cell wall; Crr1p
16K7	CN809258	1d	Nucleotide metabolism	XP_372959	Homo sapiens	similar to rRNA intron-encoded homing endonuclease
16K9	CN809260	9	Hyp/Unknown Prot	AAB69315	Metarhizium anisopliae	proline rich protein 5MeD
16K11	CN809263	6a	Cell cycle	NP_593680	Schizosaccharomyces pombe	DNA binding protein homolog: involved in mating pheromone recognition pathway
17K1	CN809412	5f	Signaling	P34208	Schizosaccharomyces pombe	Serine/threonine-protein kinase chk1
17K3	CN809414	5b	Cytosqueloton	CAB92017	Neurospora crassa	related to actin-interacting protein AIP3
17K5	CN809417	9	Hyp/Unknown Prot	EAA72774	Gibberella zeae PH-1	hypothetical protein FG04393
17K7	CN809419	2	Energy Metabolism	CAB91687	Neurospora crassa	probable cytochrome b5
17K9	CN809422	9	Hyp/Unknown Prot			unknown
17K11	CN809424	5c	Extracellular Matrix/Cell wall protein	AAC48526	Sus scrofa	gastric mucin
18K1	CN809582	9	Hyp/Unknown Prot	EAA75490	Gibberella zeae PH-1	hypothetical protein FG05254
18K3	CN809585	1b	Carbohydrate metabolism	AAC69851	Arabidopsis thaliana	branched-chain alpha keto-acid dehydrogenase E1 alpha subunit
18K5	CN809589	9	Hyp/Unknown Prot	EAA75949	Gibberella zeae PH-1	hypothetical protein FG09763
18K7	CN809591	1b	Carbohydrate metabolism	CAE20400	Prochlorococcus marinus	possible cystathionine gamma-synthase
18K9	CN809595	1b	Carbohydrate metabolism	NP_437878	Sinorhizobium meliloti	putative dihydrodipicolinate synthetase protein
18K11	CN809604	3c	Proteolysis	AAR87746	Botryotinia fuckeliana	aspartic protease precursor
16K2	CN809249	9	Hyp/Unknown Prot	EAA75924	Gibberella zeae PH-1	hypothetical protein FG09033
16K4	CN809252	1b	Carbohydrate metabolism	CAD21296	Neurospora crassa	related to cel1 protein precursor: Cellulase 1 precursor (Endoglucanase)
16K6	CN809257	9	Hyp/Unknown Prot	EAA48873	Magnaporthe grisea	hypothetical protein MG00531

16K8	CN809259	1a	Aminoacid metabolism	NP_012612	Saccharomyces cerevisiae	Tryptophan 2,3-dioxygenase, required for biosynthesis of nicotinic acid from tryptophan via kynurenine pathway
16K10	CN809262	9	Hyp/Unknown Prot	EAA75449	Gibberella zeae PH-1	hypothetical protein FG05213
16K12	CN809264	4c	Transcription	Q96NS1	Homo sapiens	Yippee-like zinc binding protein 5
17K2	CN809413	3c	Proteolysis	ZP_00218124	Burkholderia fungorum	Putative intracellular protease/amidase
17K4	CN809416	3b	Translation	NP_594285	Schizosaccharomyces pombe	translation initiation factor eif-2b
17K6	CN809418	9	Hyp/Unknown Prot	EAA69963	Gibberella zeae PH-1	hypothetical protein FG10265
17K8	CN809421	9	Hyp/Unknown Prot			unknown
17K10	CN809423	1a	Aminoacid metabolism	NP_010961	Saccharomyces cerevisiae	Putative S-adenosyl-L-homocysteine hydrolase with a probable role in S-adenosylhomocysteine catabolism and/or methionine degradation; Sah1p
17K12	CN809427	9	Hyp/Unknown Prot	EAA73352	Gibberella zeae PH-1	hypothetical protein FG03884
18K2	CN809584	9	Hyp/Unknown Prot	EAA69214	Gibberella zeae PH-1	hypothetical protein FG01068
18K4	CN809588	1f	Cofactor and vitamins	T40624	Schizosaccharomyces pombe	probable class v pyridoxal phosphate dependent aminotransferase
18K6	CN809590	1b	Carbohydrate metabolism	ZP_00119023	Cytophaga hutchinsonii	Carbonic anhydrase
18K8	CN809594	5d	Transport proteins	CAE76159	Neurospora crassa	related to monocarboxylate transporter 4
18K10	CN809600	9	Hyp/Unknown Prot	EAA77407	Gibberella zeae PH-1	hypothetical protein FG09415
18K12	CN809605	3b	Translation	EAA29185	Neurospora crassa	Eukaryotic translation initiation factor 4A (EIF-4A) (EIF4A)
16G1	CN809285	9	Hyp/Unknown Prot			Unknown
16G3	CN809288	6c	DNA synthesis	CAB71844	Schizosaccharomyces pombe	SPAC694.06c: involved in DNA replication checkpoint; invovled in the activation of Cds1p during replication stress (required)
16G5	CN809292	1b	Carbohydrate metabolism	ZP_00027461	Burkholderia fungorum	Predicted metal-dependent hydrolase of the TIM-barrel fold
16G7	CN809294	3a	Ribosomal proteins	CAD71058	Neurospora crassa	60S ribosomal proteins
16G9	CN809297	1b	Carbohydrate metabolism	CAD21495	Neurospora crassa	probable delta3, 5-delta2, 4-dienoyl-CoA isomerase precursor
16G11	CN809302	3d	Prot modification/targeting	CAD21421	Neurospora crassa	probable cyclophilin: catalyzes the cis-trans isomerization of peptide bonds N-terminal to proline residues
17G1	CN809450	3c	Proteolysis	XP_330028	Neurospora crassa	probable 26S proteasome regulatory particle chain RPT1 [MIPS]
17G3	CN809452	9	Hyp/Unknown Prot			unknown
17G5	CN809455	9	Hyp/Unknown Prot	EAA74964	Gibberella zeae PH-1	hypothetical protein FG06347
17G7	CN809457	2	Energy Metabolism	NP_595868	Schizosaccharomyces pombe	putative nadh-dependent flavin oxidoreductase
17G9	CN809462	5c	Extracellular Matrix/Cell wall protein	AAH36187	Homo sapiens	Ser/Arg-related nuclear matrix protein
17G11	CN809465	9	Hyp/Unknown Prot	EAA50099	Magnaporthe grisea 70-15	hypothetical protein MG03858.4
18G1	CN809633	9	Hyp/Unknown Prot			unknown

18G3	CN809636	6d	Cell polarity	NP_014106	Saccharomyces cerevisiae	Multicopy Suppressor of Bud Emergence; Msb3p
18G5	CN809639	1b	Carbohydrate metabolism	AAF34579	Hypocrea jecorina	1,2-a-D-mannosidase
18G7	CN809648	9	Hyp/Unknown Prot			unknown
18G9	CN809654	9	Hyp/Unknown Prot	EAA32547	Neurospora crassa	hypothetical protein
18G11	CN809658	6c	DNA synthesis	O94329	Schizosaccharomyces pombe	Probable DNA replication complex GINS protein PSF2
16G2	CN809287	9	Hyp/Unknown Prot	EAA32519	Neurospora crassa	predicted protein
16G4	CN809290	9	Hyp/Unknown Prot	EAA75645	Gibberella zeae PH-1	hypothetical protein FG06000
16G6	CN809293	1c	Lipid metabilism	AAO73955	Candida tropicalis	CYP52A15: Important for the Conversion of Fatty Acids and Alkanes to alpha, omega-Dicarboxylic Acids
16G8	CN809296	9	Hyp/Unknown Prot	EAA78022	Gibberella zeae PH-1	predicted protein
16G10	CN809301	1b	Carbohydrate metabolism	CAD71220	Neurospora crassa	related to chloroperoxidase
16G12	CN809303	1b	Carbohydrate metabolism	BAD09200	Oryza sativa	short-chain dehydrogenase/reductase family protein-like
17G2	CN809451	3d	Prot modification/targeting	EAA34512	Neurospora crassa	protein phosphatase 2A regulatory B subunit
17G4	CN809454	9	Hyp/Unknown Prot			unknown
17G6	CN809456	5f	Signaling	NP_588431	Schizosaccharomyces pombe	putative receptor-associated protein
17G8	CN809461	5d	Transport proteins	EAA74131	Gibberella zeae PH-1	Adenine nucleotide translocator
17G10	CN809464	7a	Stress response	AAQ14297	Gibberella acuminata	ice nucleation protein
17G12	CN809466	9	Hyp/Unknown Prot	EAA76041	Gibberella zeae PH-1	hypothetical protein FG07068
18G2	CN809635	5d	Transport proteins	T10477	Pichia pastoris	sec13 transport protein
18G4	CN809637	9	Hyp/Unknown Prot	EAA78282	Gibberella zeae PH-1	hypothetical protein FG06497
18G6	CN809647	9	Hyp/Unknown Prot	EAA71415	Gibberella zeae PH-1	hypothetical protein FG08554
18G8	CN809651	1b	Carbohydrate metabolism	CAA51721	Claviceps purpurea	glyceraldehyde-3-phosphate dehydrogenase
18G10	CN809655	5e	Cellular transportation	NP_596263	Schizosaccharomyces pombe	hypothetical coiled-coil protein
18G12	CN809659	7c	Secondary metabolites	ZP_00227715	Kineococcus radiotolerans	Protein involved in biosynthesis of mitomycin antibiotics/polyketide fumonisin
11B1	CN808236	1c	Lipid metabilism	CAE76615	Neurospora crassa	related to oxysterol-binding protein
11B3	CN808238	9	Hyp/Unknown Prot	EAA73899	Gibberella zeae PH-1	hypothetical protein FG05186
11B5	CN808245	6d	Cell polarity	AAN85204	Cryptococcus neoformans	calcineurin temperature suppressor Cts1
11B7	CN808247	6a	Cell cycle	S42719	Saccharomyces exiguus	actin-binding protein ABP1: involved in the spatial organisation of cell surface growth
11B9	CN808249	7b	Detoxification	AAF66098	Leptosphaeria maculans	putative cyanide hydratase
11B11	CN808251	1b	Carbohydrate metabolism	AAL09829	Coccidioides posadasii	beta-glucosidase 5
12B1	CN808487	3a	Ribosomal proteins	CAE75674	Neurospora crassa	probable 40s ribosomal protein S6.e, cytosolic
12B3	CN808490	1b	Carbohydrate metabolism	AAQ66044	Porphyromonas gingivalis	alpha-1,2-mannosidase family protein

12B5	CN808493	1b	Carbohydrate metabolism	XP_328167	Neurospora crassa	related to 6-phosphofructo-2-kinase
12B7	CN808498	5f	Signaling	T39655	Schizosaccharomyces pombe	VHS domain containing, signal transducing adaptor STAM-like protein
12B9	CN808506	6c	DNA synthesis	CAE76259	Neurospora crassa	related to DNA replication origin recognition protein Orc1p
12B11	CN808514	9	Hyp/Unknown Prot	EAA73854	Gibberella zeae PH-1	hypothetical protein FG06232
13B1	CN808718	9	Hyp/Unknown Prot			Unknown
13B3	CN808725	9	Hyp/Unknown Prot			Unknown
13B5	CN808726	3b	Translation	AAP35865	Homo sapiens	eukaryotic translation initiation factor 1A, Y chromosome
13B7	CN808729	1f	Cofactor and vitamins	AAK07850	Neurospora crassa	Snz-type pyridoxine vitamin B6 biosynthetic protein
13B9	CN808733	5a	Cell wall	XP_323602	Neurospora crassa	probable DFG5 protein [MIPS], required for cell wall biogenesis
13B11	CN808737	9	Hyp/Unknown Prot			Unknown
14B1	CN808865	2	Energy Metabolism	NP_011358	Saccharomyces cerevisiae	Oxidoreductase
14B3	CN808868	7c	Secondary metabolites	AAN74810	Gibberella moniliformis	Png1p: polyketide synthase gene required for biosynthesis of fumonisin
14B5	CN808872	9	Hyp/Unknown Prot	EAA29414	Neurospora crassa	predicted protein
14B7	CN808874	1b	Carbohydrate metabolism	NP_010618	Saccharomyces cerevisiae	ER membrane glycoprotein subunit of the glycosylphosphatidylinositol transamidase complex
14B9	CN808876	1c	Lipid metabilism	AAB62807	Nicotiana tabacum	S-adenosyl-methionine-sterol-C-methyltransferase homolog
14B11	CN808879	5a	Cell wall	BAD01559	Aspergillus kawachii	cell wall protein
15B1	CN809026	9	Hyp/Unknown Prot	EAA53523	Magnaporthe grisea	predicted protein
15B3	CN809028	9	Hyp/Unknown Prot			Unknown
15B5	CN809037	5b	Cytosqueloton	AAM66707	Aspergillus fumigatus	fucose-specific lectin
15B7	CN809039	9	Hyp/Unknown Prot	EAA60090	Aspergillus nidulans	hypothetical protein AN4855
15B9	CN809042	9	Hyp/Unknown Prot	EAA61011	Aspergillus nidulans	hypothetical protein AN4933
15B11	CN809044	9	Hyp/Unknown Prot	EAA71596	Gibberella zeae PH-1	hypothetical protein FG08290
11B2	CN808237	1b	Carbohydrate metabolism	AAN32715	Fusarium oxysporum	protein kinase SNF1 (sucrose non-fermenting 1)
11B4	CN808241	1a	Aminoacid metabolism	NP_013190	Saccharomyces cerevisiae	putative alanine transaminase (glutamyc pyruvic transaminase); Ylr089cp
11B6	CN808246	9	Hyp/Unknown Prot	EAA70991	Gibberella zeae PH-1	hypothetical protein FG04053
11B8	CN808248	5f	Signaling	NP_011043	Saccharomyces cerevisiae	Transmembrane osmosensor
11B10	CN808250	9	Hyp/Unknown Prot	AAM38208	Xanthomonas axonopodis	conserved hypothetical protein
11B12	CN808254	5a	Cell wall	CAB88654	Neurospora crassa	related to mixed-linked glucanase precursor MLG1
12B2	CN808488	4c	Transcription	EAA34005	Neurospora crassa	deoxyhypusine synthase (EC 2.5.1.46) B7F21.30
12B4	CN808491	1b	Carbohydrate metabolism	NP_699578	Brucella suis	xylulose-5-phosphate/fructose-6-phosphate phosphoketolase
12B6	CN808494	7a	Stress response	NP_014581	Saccharomyces cerevisiae	putative hemolysin-like protein
12B8	CN808505	9	Hyp/Unknown Prot			Unknown
12B10	CN808510	9	Hyp/Unknown Prot	EAA70132	Gibberella zeae PH-1	hypothetical protein FG09906

12B12	CN808516	5a	Cell wall	NP_010874	Saccharomyces cerevisiae	Utr2p: cell wall organization and biogenesis
13B2	CN808720	9	Hyp/Unknown Prot	NP_595005	Schizosaccharomyces pombe	hypothetical protein
13B4	CN808717	5d	Transport proteins	NP_062709	Mus musculus	Class II aldolase/adducin N-terminal domain protein [Carbohydrate transport and metabolism]
13B6	CN808727	7c	Secondary metabolites	Q00278	Aspergillus parasiticus	Aflatoxin biosynthesis ketoreductase NOR-1
13B8	CN808732	5d	Transport proteins	NP_594381	Schizosaccharomyces pombe	putative translocation protein
13B10	CN808734	3a	Ribosomal proteins	ZP_00126012	Pseudomonas syringae	Ribosomal protein S16
13B12	CN808739	1a	Aminoacid metabolism	NP_595239	Schizosaccharomyces pombe	putative fructosyl amino acid oxidase
14B2	CN808866	1b	Carbohydrate metabolism	CAE76329	Neurospora crassa	probable succinate dehydrogenase (ubiquinone) iron-sulfur protein precursor
14B4	CN808871	1b	Carbohydrate metabolism	EAA75808	Gibberella zeae PH-1	Isocitric dehydrogenase
14B6	CN808873	1b	Carbohydrate metabolism	CAA83503	Leishmania tarentolae	aldehyde dehydrogenase
14B8	CN808875	9	Hyp/Unknown Prot	EAA72097	Gibberella zeae PH-1	hypothetical protein FG08520
14B10	CN808878	3a	Ribosomal proteins	NP_566326	Arabidopsis thaliana	rhomboid protein-related
14B12	CN808880	9	Hyp/Unknown Prot			Unknown
15B2	CN809027	9	Hyp/Unknown Prot	EAA68759	Gibberella zeae PH-1	hypothetical protein FG00410
15B4	CN809032	9	Hyp/Unknown Prot	EAA76224	Gibberella zeae PH-1	hypothetical protein FG06711
15B6	CN809038	1b	Carbohydrate metabolism	AAF72527	Aspergillus nidulans	delta-1-pyrroline-5-carboxylate dehydrogenase
15B8	CN809040	1b	Carbohydrate metabolism	AAP57755	Hypocrea jecorina	Cel3b: putative secreted beta-glucosidase
15B10	CN809043	1a	Aminoacid metabolism	CAD71118	Neurospora crassa	related to ornithine decarboxylase antizyme
15B12	CN809045	3d	Prot modification/targeting	AAM48104	Emericella nidulans	theta class glutathione S-transferase
11D1	CN808298	7b	Detoxification	CAE76270	Neurospora crassa	related to phenol 2-monooxygenase
11D3	CN808309	9	Hyp/Unknown Prot	EAA29414	Neurospora crassa	predicted protein
11D5	CN808317	9	Hyp/Unknown Prot	EAA76214	Gibberella zeae PH-1	hypothetical protein FG06701
11D7	CN808323	9	Hyp/Unknown Prot	EAA70750	Gibberella zeae PH-1	hypothetical protein FG00804
11D9	CN808327	9	Hyp/Unknown Prot	EAA48457	Magnaporthe grisea	hypothetical protein MG00115.4
11D11	CN808330	1b	Carbohydrate metabolism	AAL84175	Magnaporthe grisea	3,4-dihydroxy-2-butanone 4-phosphate synthase
12D1	CN808541	5a	Cell wall	CAB44709	Metarhizium flavoviride	chitinase
12D3	CN808543	1a	Aminoacid metabolism	ZP_00096788	Novosphingobium aromaticivorans	Glycine/D-amino acid oxidases (deaminating)
12D5	CN808550	1b	Carbohydrate metabolism	AAC39498	Hypocrea jecorina	mannose-1-phosphate guanylyltransferase
12D7	CN808553	4a	RNA metabolism	CAE76417	Neurospora crassa	related to aspartate--tRNA ligase, mitochondrial
12D9	CN808558	4b	RNA modification	NP_595448	Schizosaccharomyces pombe	putative cleavage and polyadenylation specificity factor subunit
12D11	CN808565	9	Hyp/Unknown Prot	EAA70405	Gibberella zeae PH-1	hypothetical protein FG10089

13D1	CN808760	5b	Cytosqueloton	XP_328152	Neurospora crassa	Alp11 homolog of tubulin-folding cofactor B
13D3	CN808763	9	Hyp/Unknown Prot			Unknown
13D5	CN808765	9	Hyp/Unknown Prot			Unknown
13D7	CN808771	5d	Transport proteins	CAD71229	Neurospora crassa	related to transporter-like protein CTL2
13D9	CN808776	9	Hyp/Unknown Prot	EAA70019	Gibberella zeae PH-1	hypothetical protein FG10321
13D11	CN808778	9	Hyp/Unknown Prot			Unknown
14D1	CN808899	6c	DNA synthesis	Q8X082	Neurospora crassa	Probable activator 1 subunit 5 (Replication factor C subunit 5)
14D3	CN808902	9	Hyp/Unknown Prot	EAA68483	Gibberella zeae PH-1	hypothetical protein FG00470
14D5	CN808907	9	Hyp/Unknown Prot	EAA61746	Aspergillus nidulans	hypothetical protein AN7375
14D7	CN808910	1a	Aminoacid metabolism	JC4255	Neurospora crassa	met-10+ protein: involved in methionine biosynthesis, transport and utilization
14D9	CN808912	1b	Carbohydrate metabolism	XP_328193	Neurospora crassa	Alcohol dehydrogenase I
14D11	CN808914	6a	Cell cycle	CAE76569	Neurospora crassa	related to Het-c protein: heterokaryon incompatibility
15D1	CN809064	5f	Signaling	AAB64456	Saccharomyces cerevisiae	Ydr267cp: Beta-transducin-containing proteins, required for cell viability
15D3	CN809066	9	Hyp/Unknown Prot			Unknown
15D5	CN809070	9	Hyp/Unknown Prot	EAA29409	Neurospora crassa	hypothetical protein
15D7	CN809075	9	Hyp/Unknown Prot	EAA64687	Aspergillus nidulans	hypothetical protein AN2582
15D9	CN809081	1b	Carbohydrate metabolism	P78695	Neurospora crassa	78 kDa glucose-regulated protein homolog precursor (GRP 78) (Immunoglobulin heavy chain binding protein homolog) (BIP
15D11	CN809086	1c	Lipid metabilism	JN0553	Candida rugosa	triacylglycerol lipase (EC 3.1.1.3) 5 precursor
11D2	CN808304	7c	Secondary metabolites	AAO17168	Photorhabdus luminescens	Orf2: potative transposase orf within toxin gene complex
11D4	CN808313	1b	Carbohydrate metabolism	NP_948628	Rhodopseudomonas palustris	acyl-CoA dehydrogenase
11D6	CN808321	3d	Prot modification/targeting	NP_011600	Saccharomyces cerevisiae	Phosphorylation Inhibited by Long chain base; Pil1p
11D8	CN808326	1c	Lipid metabilism	AAN77731	Pichia ciferrii	sphinganine hydroxylase
11D10	CN808328	6d	Cell polarity	CAD21110	Neurospora crassa	related to p33ING1b (ING1) protein: nuclear growth inhibitor and candidate tumor suppressor
11D12	CN808333	1c	Lipid metabilism	AAO73449	Fusarium sporotrichioides	putative P450 monooxygenase
12D2	CN808542	5a	Cell wall	BAB62318	Aspergillus kawachii	endoglucanase B
12D4	CN808549	5f	Signaling	XP_327937	Neurospora crassa	related to ser/thr protein kinase IME2 [MIPS]
12D6	CN808551	9	Hyp/Unknown Prot	EAA68909	Gibberella zeae PH-1	hypothetical protein FG00187
12D8	CN808557	5d	Transport proteins	CAD37009	Neurospora crassa	related to NCE102 protein: Non-classical export protein 2
12D10	CN808562	9	Hyp/Unknown Prot	EAA77297	Gibberella zeae PH-1	hypothetical protein FG07925
12D12	CN808574	6a	Cell cycle	NP_011300	Saccharomyces cerevisiae	cyclin-like protein that interacts with Pho85p in affinity chromatography; Clg1p
13D2	CN808762	9	Hyp/Unknown Prot			Unknown

13D4	CN808764	9	Hyp/Unknown Prot	EAA76655	Gibberella zeae PH-1	hypothetical protein FG09539
13D6	CN808769	5d	Transport proteins	DAA01434	Mus musculus	adaptin-ear-binding coat-associated protein
13D8	CN808775	1b	Carbohydrate metabolism	Q12726	Yarrowia lipolytica	Homocitrate synthase, mitochondrial precursor
13D10	CN808777	2	Energy Metabolism	AAL58884	Aspergillus nidulans	putative oxidoreductase
13D12	CN808779	7c	Secondary metabolites	AAO27746	Fusarium sporotrichioides	putative methyltransferase
14D2	CN808900	5a	Cell wall	AAN77504	Lysobacter enzymogenes	beta-1,3-glucanase B
14D4	CN808905	3a	Ribosomal proteins	CAE75674	Neurospora crassa	probable 40s ribosomal protein S6.e, cytosolic
14D6	CN808909	9	Hyp/Unknown Prot			Unknown
14D8	CN808911	4c	Transcription	NP_595529	Schizosaccharomyces pombe	putative transcriptional regulatory protein with ARID DNA binding domain
14D10	CN808913	9	Hyp/Unknown Prot	EAA62742	Aspergillus nidulans	hypothetical protein AN5649
14D12	CN808918	9	Hyp/Unknown Prot			Unknown
15D2	CN809065	6e	Sexual cycle	CAD21376	Neurospora crassa	related to DNA binding protein NsdD protein: important role in sexual development
15D4	CN809069	9	Hyp/Unknown Prot	EAA71777	Gibberella zeae PH-1	hypothetical protein FG03088
15D6	CN809072	9	Hyp/Unknown Prot			Unknown
15D8	CN809078	5e	Cellular transportation	NP_011461	Saccharomyces cerevisiae	Protein localized to COPII-coated vesicles, involved in vesicle formation and incorporation of specific secretory cargo; required for the delivery of bud-site selection protein Axl2p to cell surface
15D10	CN809085	9	Hyp/Unknown Prot	EAA68463	Gibberella zeae PH-1	hypothetical protein FG00450
15D12	CN809091	9	Hyp/Unknown Prot			Unknown
11F1	CN808362	4b	RNA modification	NP_060853	Homo sapiens	WD repeat domain 33: pre-mRNA processing
11F3	CN808367	9	Hyp/Unknown Prot			Unknown
11F5	CN808372	9	Hyp/Unknown Prot	EAA67692	Gibberella zeae PH-1	hypothetical protein FG09954
11F7	CN808374	1a	Aminoacid metabolism	CAC40647	Aspergillus niger	prolyl aminopeptidase A
11F9	CN808381	9	Hyp/Unknown Prot	EAA55098	Magnaporthe grisea	hypothetical protein MG06755.4
11F11	CN808383	9	Hyp/Unknown Prot	EAA75648	Gibberella zeae PH-1	hypothetical protein FG06003
12F1	CN808616	9	Hyp/Unknown Prot	EAA75012	Gibberella zeae PH-1	hypothetical protein FG10755
12F3	CN808620	3c	Proteolysis	BAD16888	Oryza sativa	putative ubiquitin-associated (UBA) protein
12F5	CN808625	1b	Carbohydrate metabolism	EAA73718	Gibberella zeae PH-1	Probable succinyl-CoA ligase [GDP-forming] beta-chain, mitochondrial precursor (Succinyl-CoA synthetase, beta chain) (SCS-beta)
12F7	CN808629	7a	Stress response	AAP35603	Homo sapiens	macrophage erythroblast attacher
12F9	CN808634	9	Hyp/Unknown Prot	EAA73947	Gibberella zeae PH-1	predicted protein
12F11	CN808642	9	Hyp/Unknown Prot	EAA77783	Gibberella zeae PH-1	hypothetical protein FG09734
13F1	CN808796	5a	Cell wall	BAD01559	Aspergillus kawachii	cell wall protein
13F3	CN808798	9	Hyp/Unknown Prot	T49641	Neurospora crassa	nebula related protein
13F5	CN808805	9	Hyp/Unknown Prot	EAA52614	Magnaporthe grisea	hypothetical protein MG05306
13F7	CN808807	9	Hyp/Unknown Prot	EAA47842	Magnaporthe grisea 70-15	hypothetical protein MG03085.4
13F9	CN808810	3b	Translation	EAA77131	Gibberella zeae PH-1	Elongation factor

13F11	CN808812	5d	Transport proteins	BAC67162	Magnaporthe grisea	ATP-binding cassette transporter
14F1	CN808944	9	Hyp/Unknown Prot			Unknown
14F3	CN808947	2	Energy Metabolism	EAA34007	Neurospora crassa	chorismate synthase/flavin reductase, NADPH-dependent [MIPS]
14F5	CN808950	1b	Carbohydrate metabolism	CAE32303	Bordetella bronchiseptica	succinate-semialdehyde dehydrogenase [NADP+]
14F7	CN808952	5b	Cytosqueloton	NP_066267	Homo sapiens	ankyrin 3 isoform 1; ankyrin-3, node of Ranvier; link the integral membrane proteins to the underlying spectrin-actin cytoskeleton and play key roles in activities as cell motility, activation, proliferation, contact, and the maintenance of specialized me
14F9	CN808956	9	Hyp/Unknown Prot	EAA77223	Gibberella zeae PH-1	hypothetical protein FG07364
14F11	CN808960	3a	Ribosomal proteins	O59950	Yarrowia lipolytica	40S ribosomal protein S4
15F1	CN809108	7a	Stress response	CAD21287	Neurospora crassa	related to OPY2 protein: overproduction induces pheromone resistance
15F3	CN809110	3b	Translation	CAE85545	Neurospora crassa	related to translation initiation factor eIF3 subunit
15F5	CN809115	9	Hyp/Unknown Prot	EAA67249	Gibberella zeae PH-1	hypothetical protein FG00909
15F7	CN809117	9	Hyp/Unknown Prot	EAA73009	Gibberella zeae PH-1	hypothetical protein FG08048
15F9	CN809121	9	Hyp/Unknown Prot	NP_010390	Saccharomyces cerevisiae	Putative membrane protein, conserved in mammals; Tms1p, tumor differentially expressed
15F11	CN809123	1b	Carbohydrate metabolism	AAP08049	Bacillus cereus	3-oxoacyl-[acyl-carrier protein] reductase
11F2	CN808365	1b	Carbohydrate metabolism	NP_000427	Homo sapiens	3-oxoacid CoA transferase
11F4	CN808371	9	Hyp/Unknown Prot	EAA70737	Gibberella zeae PH-1	hypothetical protein FG00791
11F6	CN808373	6e	Sexual cycle	O94356	Schizosaccharomyces pombe	Meiotic expression up-regulated protein 6
11F8	CN808375	9	Hyp/Unknown Prot	EAA68393	Gibberella zeae PH-1	hypothetical protein FG00663
11F10	CN808382	7b	Detoxification	O94284	Schizosaccharomyces pombe	Sulfide: quinone oxidoreductase, mitochondrial precursor (Heavy metal tolerance protein 2) (Cadmium resistance protein)
11F12	CN808384	3d	Prot modification/targeting	NP_187168	Arabidopsis thaliana	phosphoglycerate/bisphosphoglycerate mutase family protein
12F2	CN808619	7c	Secondary metabolites	AAR90254	Botryotinia fuckeliana	polyketide synthase
12F4	CN808622	9	Hyp/Unknown Prot	EAA55404	Magnaporthe grisea 70-15	hypothetical protein MG09211.4
12F6	CN808628	9	Hyp/Unknown Prot	XP_357010	Mus musculus	similar to CG13990-PA
12F8	CN808633	1b	Carbohydrate metabolism	AAM65399	Arabidopsis thaliana	contains similarity to O-linked GlcNAc transferases
12F10	CN808635	9	Hyp/Unknown Prot	EAA76232	Gibberella zeae PH-1	hypothetical protein FG09610
12F12	CN808646	2	Energy Metabolism	NP_875054	Prochlorococcus marinus	NADPH-dependent reductase
13F2	CN808797	1b	Carbohydrate metabolism	ZP_00006666	Rhodobacter sphaeroides	Glucose/sorbosone dehydrogenases

13F4	CN808802	1a	Aminoacid metabolism	ZP_00096788	Novosphingobium aromaticivorans	Glycine/D-amino acid oxidases
13F6	CN808806	1b	Carbohydrate metabolism	AAG36950	Aspergillus nidulans	phe-inhibited DAHP synthase
13F8	CN808808	8	Transposable Elements	AAF97810	Cryphonectria parasitica	transposase
13F10	CN808811	9	Hyp/Unknown Prot	EAA72393	Gibberella zeae PH-1	hypothetical protein FG02893
13F12	CN808813	1b	Carbohydrate metabolism	BAC68337	Streptomyces avermitilis MA-4680	putative glycosyl hydrolase
14F2	CN808945	9	Hyp/Unknown Prot	EAA77767	Gibberella zeae PH-1	hypothetical protein FG09718
14F4	CN808948	3c	Proteolysis	NP_015428	Saccharomyces cerevisiae	20S proteasome beta-type subunit, responsible for the chymotryptic activity of the proteasome; Pre2p
14F6	CN808951	3d	Prot modification/targeting	EAA32582	Neurospora crassa	Serine/Threonine protein phosphatase PP2A catalytic subunit
14F8	CN808955	1b	Carbohydrate metabolism	CAC28692	Neurospora crassa	related to mevalonate kinase
14F10	CN808958	3c	Proteolysis	CAC95049	Metarhizium anisopliae	subtilisin-like serine protease PR1A
14F12	CN808961	9	Hyp/Unknown Prot			Unknown
15F2	CN809109	5b	Cytosqueloton	NP_014615	Saccharomyces cerevisiae	calcium-binding protein regulates mitochondrial distribution and morphology
15F4	CN809111	9	Hyp/Unknown Prot	CAB65565	Streptomyces coelicolor A3(2)	putative membrane protein
15F6	CN809116	1e	N-, P-, S-metabolism	AAM91942	Candida kefyr	Ure2p: nitrogen catabolite repression regulator
15F8	CN809118	3b	Translation	NP_001408	Homo sapiens	eukaryotic translation initiation factor 4B
15F10	CN809122	3b	Translation	AAR16425	Metarhizium anisopliae	translation elongation factor 1 alpha
15F12	CN809125	9	Hyp/Unknown Prot			Unknown
11H1	CN808426	9	Hyp/Unknown Prot	EAA34784	Neurospora crassa	predicted protein
11H3	CN808433	5d	Transport proteins	NP_503070	Caenorhabditis elegans	histidine-rich membrane transpoter ke4 (4S230)
11H5	CN808437	1c	Lipid metabilism	BAB93118	Kluyveromyces lactis	putative delta 8-sphingolipid desaturase
11H7	CN808447	3c	Proteolysis	CAD70384	Neurospora crassa	related to ubiquitin fusion degradation protein
11H9	CN808449	9	Hyp/Unknown Prot	EAA76224	Gibberella zeae PH-1	hypothetical protein FG06711
11H11	CN808452	9	Hyp/Unknown Prot	EAA69214	Gibberella zeae PH-1	hypothetical protein FG01068
12H1	CN808672	1b	Carbohydrate metabolism	NP_905145	Porphyromonas gingivalis	alpha-1,2-mannosidase family protein
12H3	CN808675	9	Hyp/Unknown Prot	EAA68061	Gibberella zeae PH-1	hypothetical protein FG01843
12H5	CN808678	5b	Cytosqueloton	NP_012194	Saccharomyces cerevisiae	mitochondrial acidic matrix protein
12H7	CN808682	4c	Transcription	AAK58576	Ustilago maydis	TPR(transcritpional repressor)-containing protein Mql1
12H9	CN808684	1b	Carbohydrate metabolism	BAA09319	Acremonium chrysogenum	beta-isopropylmalate dehydrogenase
12H11	CN808688	9	Hyp/Unknown Prot			unknown
13H1	CN808831	9	Hyp/Unknown Prot	EAA73945	Gibberella zeae PH-1	hypothetical protein FG05682
13H3	CN808833	9	Hyp/Unknown Prot	EAA50977	Magnaporthe grisea	hypothetical protein MG04736
13H5	CN808836	9	Hyp/Unknown Prot			Unknown

13H7	CN808839	4b	RNA modification	NP_587684	Schizosaccharomyces pombe	RNA-binding protein; putative polyadenylation factor
13H9	CN808844	6a	Cell cycle	AAF18153	Neurospora crassa	heterokaryon incompatibility protein het-6
13H11	CN808848	1c	Lipid metabilism	XP_341697	Rattus norvegicus	similar to steroid dehydrogenase-like
14H1	CN808980	1b	Carbohydrate metabolism	CAD01124	Neurospora crassa	related to 7alpha-cephem-methoxylase P8 chain
14H3	CN808982	2	Energy Metabolism	NP_595306	Schizosaccharomyces pombe	thioredoxin-like protein
14H5	CN808987	1b	Carbohydrate metabolism	BAB84516	Monascus anka	acetamidase
14H7	CN808989	9	Hyp/Unknown Prot			Unknown
14H9	CN808992	1a	Aminoacid metabolism	NP_012612	Saccharomyces cerevisiae	Tryptophan 2,3-dioxygenase, required for biosynthesis of nicotinic acid from tryptophan via kynurenine pathway
14H11	CN809001	3d	Prot modification/targeting	CAF05891	Neurospora crassa	related to protein tyrosine phosphatase phi
15H1	CN809143	3a	Ribosomal proteins	EAA69730	Gibberella zeae PH-1	60S ribosomal protein L16
15H3	CN809146	9	Hyp/Unknown Prot	EAA77302	Gibberella zeae PH-1	hypothetical protein FG07930
15H5	CN809148	9	Hyp/Unknown Prot	EAA67323	Gibberella zeae PH-1	hypothetical protein FG10346
15H7	CN809151	1a	Aminoacid metabolism	BAA86934	Aspergillus oryzae	glutaminase A
15H9	CN809154	3d	Prot modification/targeting	CAD36971	Neurospora crassa	related to branch point bridging protein (MSL5)
15H11	CN809156	1d	Nucleotide metabolism	EAA67817	Gibberella zeae PH-1	ADP-ribosylation factor
11H2	CN808429	7a	Stress response	NP_013352	Saccharomyces cerevisiae	stress-induced yeast MPV17 homolog; Ylr251wp
11H4	CN808435	1b	Carbohydrate metabolism	AAO34674	Gibberella zeae	glucosidase
11H6	CN808445	9	Hyp/Unknown Prot	EAA67688	Gibberella zeae PH-1	hypothetical protein FG09950
11H8	CN808448	1c	Lipid metabilism	AAS66899	Chaetomium globosum	dienelactone hydrolase
11H10	CN808451	9	Hyp/Unknown Prot			Unknown
11H12	CN808455	1f	Cofactor and vitamins	CAD21231	Neurospora crassa	probable coproporphyrinogen oxidase precursor
12H2	CN808674	4c	Transcription	P40467	Saccharomyces cerevisiae	Putative 108.8 kDa transcriptional regulatory protein in FKH1-STH1 intergenic region
12H4	CN808677	7a	Stress response	NP_011835	Saccharomyces cerevisiae	cell wall integrity and stress response component 4; Wsc4p
12H6	CN808681	1c	Lipid metabilism	CAE76325	Neurospora crassa	related to steroid monooxygenase
12H8	CN808683	6d	Cell polarity	S47549	Saccharomyces cerevisiae	cell division control protein CDC31
12H10	CN808687	3c	Proteolysis	CAB63907	Metarhizium anisopliae	Subtilisin-like protease PR1H
12H12	CN808689	1b	Carbohydrate metabolism	ZP_00044625	Magnetococcus sp. MC-1	3-deoxy-D-arabino-heptulosonate 7-phosphate (DAHP) synthase
13H2	CN808832	6g	Cell death	AAA58585	Acanthamoeba polyphaga	hemolysin
13H4	CN808835	6g	Cell death	NP_588336	Schizosaccharomyces pombe	putative coatmer delta subunit
13H6	CN808838	5d	Transport proteins	NP_014522	Saccharomyces	monocarboxylate permease

					cerevisiae	
13H8	CN808841	5b	Cytosqueloton	NP_593695	Schizosaccharomyces pombe	putative vacuolar sorting protein
13H10	CN808846	5f	Signaling	AAF00024	Glomerella cingulata	putative seven transmembrane receptor
13H12	CN808849	5d	Transport proteins	CAB43936	Emericella nidulans	GABA permease
14H2	CN808981	5c	Extracellular Matrix/Cell wall protein	CAD36967	Neurospora crassa	related to integral membrane protein PTH11
14H4	CN808985	9	Hyp/Unknown Prot	EAA75104	Gibberella zeae PH-1	hypothetical protein FG05560
14H6	CN808988	4a	RNA metabolism	CAD21184	Neurospora crassa	related to RNA annealing protein
14H8	CN808991	4b	RNA modification	NP_011694	Saccharomyces cerevisiae	Poly(A)-binding protein binding protein
14H10	CN808997	9	Hyp/Unknown Prot	CAE25657	Rhodopseudomonas palustris	possible OmpA family member
14H12	CN809002	9	Hyp/Unknown Prot	EAA72610	Gibberella zeae PH-1	hypothetical protein FG08582
15H2	CN809144	9	Hyp/Unknown Prot			Unknown
15H4	CN809147	9	Hyp/Unknown Prot	EAA35275	Neurospora crassa	predicted protein
15H6	CN809150	9	Hyp/Unknown Prot	EAA77810	Gibberella zeae PH-1	hypothetical protein FG07212
15H8	CN809153	1b	Carbohydrate metabolism	CAD71225	Neurospora crassa	probable homoaconitase precursor
15H10	CN809155	2	Energy Metabolism	CAE76213	Neurospora crassa	probable GTP-binding protein
15H12	CN809159	1b	Carbohydrate metabolism	CAD70482	Neurospora crassa	related to triose phosphate/3-phosphoglycerate/phosphate translocator
16B1	CN809179	9	Hyp/Unknown Prot	EAA68810	Gibberella zeae PH-1	hypothetical protein FG02567
16B3	CN809186	1b	Carbohydrate metabolism	NP_588132	Schizosaccharomyces pombe	probable utp-glucose-1-phosphate uridylyltransferase
16B5	CN809189	9	Hyp/Unknown Prot			Unknown
16B7	CN809193	1c	Lipid metabilism	EAA35413	Neurospora crassa	related to lysophospholipase
16B9	CN809197	9	Hyp/Unknown Prot	EAA73262	Gibberella zeae PH-1	hypothetical protein FG04478
16B11	CN809201	5a	Cell wall	CAD70866	Neurospora crassa	related to chitinase
17B1	CN809340	9	Hyp/Unknown Prot	EAA71001	Gibberella zeae PH-1	hypothetical protein FG02946
17B3	CN809348	9	Hyp/Unknown Prot	EAA67497	Gibberella zeae PH-1	hypothetical protein FG00406
17B5	CN809356	9	Hyp/Unknown Prot	EAA55949	Magnaporthe grisea 70-15	hypothetical protein MG01600.4
17B7	CN809359	3a	Ribosomal proteins	AAK48942	Neurospora crassa	60S ribosomal protein P0
17B9	CN809362	6g	Cell death	NP_080678	Mus musculus	autophagy Apg3p/Aut1p-like
17B11	CN809366	1a	Aminoacid metabolism	NP_493686	Caenorhabditis elegans	Cystinosin/ERS1p repeat
18B1	CN809514	9	Hyp/Unknown Prot	EAA32201	Neurospora crassa	hypothetical protein
18B3	CN809517	4b	RNA modification	CAB91288	Neurospora crassa	related to PCF11 component of pre-mRNA 3'-end processing factor
18B5	CN809521	1a	Aminoacid metabolism	CAE76231	Neurospora crassa	related to O-methylsterigmatocystin oxidoreductase
18B7	CN809527	3a	Ribosomal proteins	CAE76105	Neurospora crassa	related to 40S ribosomal protein S9, mitochondrial precursor
18B9	CN809530	9	Hyp/Unknown Prot	EAA76155	Gibberella zeae PH-1	hypothetical protein FG09604
18B11	CN809534	1c	Lipid metabilism	CAB92911	Candida albicans	phosphatidylinositol phospholipase
19B1	AJ251964	3c	Proteolysis		Metarhizium anisopliae 2575	Pr1G Subtilisin
19B3	AJ251972	3c	Proteolysis		Metarhizium anisopliae 2575	Pr1I Subtilisin

19B5	AJ293220	3c	Proteolysis		Metarhizium anisopliae 2575	Pr1K Subtilisin
19B7		5b	Cytosqueloton		Metarhizium anisopliae 2575	beta-Tubulin 2
16B2	CN809183	1b	Carbohydrate metabolism	AAP08049	Bacillus cereus	3-oxoacyl-[acyl-carrier protein] reductase
16B4	CN809187	6b	Chromosome structure	AAL82734	Aspergillus fumigatus	structural maintenance of chromosome protein
16B6	CN809192	9	Hyp/Unknown Prot	EAA70750	Gibberella zeae PH-1	hypothetical protein FG00804
16B8	CN809194	3a	Ribosomal proteins	EAA26938	Neurospora crassa	40S ribosomal protein S3
16B10	CN809198	1b	Carbohydrate metabolism	EAA33030	Neurospora crassa	putative 3-hydroxyisobutyrate dehydrogenase G6G8.5
16B12	CN809203	2	Energy Metabolism	CAE82003	Neurospora crassa	probable GTP-binding protein ypt5
17B2	CN809341	1b	Carbohydrate metabolism	NP_588314	Schizosaccharomyces pombe	putative short-chain dehydrogenase
17B4	CN809353	1a	Aminoacid metabolism	Q92413	Emericella nidulans	Ornithine aminotransferase (Ornithine--oxo-acid aminotransferase)
17B6	CN809357	5d	Transport proteins	CAE47906	Aspergillus fumigatus	transporter, putative
17B8	CN809360	2	Energy Metabolism	T39660	Schizosaccharomyces pombe	ATP synthase gamma chain, mitochondrial precursor
17B10	CN809364	1b	Carbohydrate metabolism	CAE47972	Aspergillus fumigatus	isoflavone reductase, putative
17B12	CN809367	1b	Carbohydrate metabolism	NP_010368	Saccharomyces cerevisiae	Rrp8p: methyltransferase activity
18B2	CN809515	4a	RNA metabolism	NP_015112	Saccharomyces cerevisiae	Involved in tRNA biogenesis; Pus1p
18B4	CN809518	6e	Sexual cycle	P40900	Schizosaccharomyces pombe	Sexual differentiation process protein isp4
18B6	CN809525	4c	Transcription	T37601	Schizosaccharomyces pombe	probable trancription factor
18B8	CN809529	1b	Carbohydrate metabolism	ZP_00043011	Magnetococcus sp.	Hydrolases of the alpha/beta superfamily
18B10	CN809531	1b	Carbohydrate metabolism	AAA34745	Saccharomyces cerevisiae	lipoic acid synthase
18B12	CN809535	4b	RNA modification	NP_010196	Saccharomyces cerevisiae	Living Under Cap-binding complex expression, mRNA splice site selection
19B2	AJ251965	3c	Proteolysis		Metarhizium anisopliae 2575	Pr1H Subtilisin
19B4	AJ269535	3c	Proteolysis		Metarhizium anisopliae 2575	Pr1J Subtilisin
19B6		5b	Cytosqueloton		Metarhizium anisopliae 2575	beta-Tubulin 1
16D1	CN809221	9	Hyp/Unknown Prot	EAA77070	Gibberella zeae PH-1	hypothetical protein FG06760
16D3	CN809228	9	Hyp/Unknown Prot	EAA69658	Gibberella zeae PH-1	hypothetical protein FG00248
16D5	CN809231	6b	Chromosome structure	BAB20823	Schizosaccharomyces pombe	Dhp1p-interacting protein: 5'-to-3' exoribonuclease required for proper chromosome segregation
16D7	CN809235	1b	Carbohydrate metabolism	Q00667	Emericella nidulans	Homogentisate 1,2-dioxygenase (Homogentisicase)
16D9	CN809239	7b	Detoxification	CAA63158	Streptomyces anulatus	N-acetylpuromycin N-acetylhydrolase
16D11	CN809245	4b	RNA modification	AAH18376	Mus musculus	PRP31: required for pre-mRNA splicing

17D1	CN809392	9	Hyp/Unknown Prot			Unknown
17D3	CN809395	1c	Lipid metabilism	EAA36156	Neurospora crassa	probable delta(24)-sterol C-methyltransferase
17D5	CN809398	9	Hyp/Unknown Prot	EAA74535	Gibberella zeae PH-1	hypothetical protein FG10928
17D7	CN809120	1d	Nucleotide metabolism	CAB63906	Metarhizium anisopliae	DNase1 protein
17D9	CN809403	9	Hyp/Unknown Prot			unknown
17D11	CN809410	1b	Carbohydrate metabolism	AAP57757	Hypocrea jecorina	Axe2: acetyl xylanesterase
18D1	CN809560	9	Hyp/Unknown Prot	EAA71197	Gibberella zeae PH-1	hypothetical protein FG04176
18D3	CN809566	5f	Signaling	EAA76369	Gibberella zeae PH-1	14-3-3 protein homolog (TH1433)
18D5	CN809568	3d	Prot modification/targeting	NP_593015	Schizosaccharomyces pombe	putative golgi peripheral membrane protein (reassembly stacking)
18D7	CN809571	9	Hyp/Unknown Prot	EAA67802	Gibberella zeae PH-1	hypothetical protein FG00999
18D9	CN809573	9	Hyp/Unknown Prot	EAA76679	Gibberella zeae PH-1	hypothetical protein FG09360
18D11	CN809578	9	Hyp/Unknown Prot	EAA75923	Gibberella zeae PH-1	hypothetical protein FG06735
19D1		7c	Secondary metabolites		Metarhizium anisopliae 2575	putative non-ribosomal peptide synthase 2D
19D3		7c	Secondary metabolites		Metarhizium anisopliae 2575	putative non-ribosomal peptide synthase 2C
19D5		7c	Secondary metabolites		Metarhizium anisopliae 2575	putative non-ribosomal peptide synthase 3A
19D7		5a	Cell wall		Metarhizium anisopliae 2575	putative chitinase
16D2	CN809223	9	Hyp/Unknown Prot	EAA55970	Magnaporthe grisea	hypothetical protein MG01621
16D4	CN809229	9	Hyp/Unknown Prot			Unknown
16D6	CN809233	9	Hyp/Unknown Prot	EAA56445	Magnaporthe grisea	hypothetical protein MG06416
16D8	CN809238	1b	Carbohydrate metabolism	T41728	Schizosaccharomyces pombe	dolichyl-di-phosphooligosaccharide-protein glycotransferase
16D10	CN809240	9	Hyp/Unknown Prot	EAA76232	Gibberella zeae PH-1	hypothetical protein FG09610
16D12	CN809246	9	Hyp/Unknown Prot	EAA75365	Gibberella zeae PH-1	hypothetical protein FG11155
17D2	CN809393	1b	Carbohydrate metabolism	CAE76318	Neurospora crassa	related to glyoxal oxidase precursor
17D4	CN809397	7a	Stress response	CAE76143	Neurospora crassa	probable nuclear protein SNF4:involved in release from glucose repression
17D6	CN809399	3c	Proteolysis	CAD70938	Neurospora crassa	probable proteasome components
17D8	CN809402	4c	Transcription	NP_832604	Bacillus cereus	Transcriptional regulator, AraC family
17D10	CN809409	3d	Prot modification/targeting	CAD21258	Neurospora crassa	related to Glc8 protein (protein phosphatase)
17D12	CN809411	9	Hyp/Unknown Prot	EAA67904	Gibberella zeae PH-1	hypothetical protein FG01431
18D2	CN809563	9	Hyp/Unknown Prot	EAA29610	Neurospora crassa	hypothetical protein
18D4	CN809567	5e	Cellular transportation	NP_593257	Schizosaccharomyces pombe	vip1 protein, vasoactive intestinal peptide receptor 1; pituitary adenylate cyclase activating polypeptide receptor
18D6	CN809570	1c	Lipid metabilism	NP_822771	Streptomyces avermitilis	putative 3-ketosteroid-delta-1-dehydrogenase
18D8	CN809572	5b	Cytosqueloton	AAP68979	Gibberella zeae PH-1	beta-tubulin
18D10	CN809574	1d	Nucleotide metabolism	CAC41973	Colletotrichum lindemuthianum	putative Rab/GTPase
18D12	CN809580	9	Hyp/Unknown Prot	EAA75873	Gibberella zeae PH-1	hypothetical protein FG05798

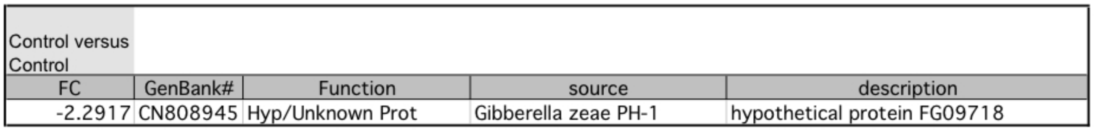
19D2		7c	Secondary metabolites		Metarhizium anisopliae 2575	putative non-ribosomal peptide synthase 2A
19D4		7c	Secondary metabolites		Metarhizium anisopliae 2575	putative non-ribosomal peptide synthase 3B
19D6		7c	Secondary metabolites		Metarhizium anisopliae 2575	putative non-ribosomal peptide synthase 3C
16F1	CN809265	5f	Signaling	AAR30131	Gibberella moniliformis	putative two-component signaling response regulator SKN7p (important in virulence)
16F3	CN809267	7a	Stress response	EAA59007	Aspergillus nidulans	heat shock protein 90 homolog (suppressor of vegetative incompatibility mod-E)
16F5	CN809269	1e	N-, P-, S-metabolism	BAB08697	Arabidopsis thaliana	polyamine oxidase
16F7	CN809271	5e	Cellular transportation	CAD71033	Neurospora crassa	Outer mitochondrial porin: required for the maintenance of mitochondrial osmotic stability and mitochondrial membrane permeability; Por1p
16F9	CN809276	9	Hyp/Unknown Prot	EAA77366	Gibberella zeae PH-1	hypothetical protein FG09008
16F11	CN809283	7b	Detoxification	NP_596231	Schizosaccharomyces pombe	nucleotide excision repair protein yeast rad23/ human HHR23A homolog
17F1	CN809430	9	Hyp/Unknown Prot			unknown
17F3	CN809434	5d	Transport proteins	CAD71083	Neurospora crassa	related to hexose transporter protein
17F5	CN809437	9	Hyp/Unknown Prot	CAF06066	Neurospora crassa	conserved hypothetical protein
17F7	CN809441	1b	Carbohydrate metabolism	CAB91740	Neurospora crassa	probable ATP citrate lyase subunit 1
17F9	CN809444	5d	Transport proteins	NP_010849	Saccharomyces cerevisiae	Ferrioxamine B transporter, induced during iron deprivation and diauxic shift
17F11	CN809448	9	Hyp/Unknown Prot	EAA28818	Neurospora crassa	predicted protein
18F1	CN809610	5e	Cellular transportation	CAB91382	Neurospora crassa	related to cytokinesis inhibitor byr4
18F3	CN809614	1b	Carbohydrate metabolism	CAE76317	Neurospora crassa	probable methylmalonate-semialdehyde dehydrogenase
18F5	CN809620	5f	Signaling	NP_012904	Saccharomyces cerevisiae	essential for cell growth and replication of M dsRNA virus; contains four beta-transducin repeats; Mak11p
18F7	CN809622	9	Hyp/Unknown Prot	EAA76739	Gibberella zeae PH-1	hypothetical protein FG06807
18F9	CN809626	7a	Stress response	NP_012284	Saccharomyces cerevisiae	Required for invasion and pseudohyphae formation in response to nitrogen starvation; Muc1p
18F11	CN809629	1d	Nucleotide metabolism	AAK08633	Aspergillus niger	uracil phosphoribosyltransferase
16F2	CN809266	9	Hyp/Unknown Prot	EAA67562	Gibberella zeae PH-1	hypothetical protein FG00910
16F4	CN809268	9	Hyp/Unknown Prot	EAA74243	Gibberella zeae PH-1	hypothetical protein FG10959
16F6	CN809270	3a	Ribosomal proteins	EAA67406	Gibberella zeae PH-1	60S ribosomal protein L17
16F8	CN809275	6b	Chromosome structure	AAR19291	Neurospora crassa	heterochromatin protein one
16F10	CN809278	9	Hyp/Unknown Prot	EAA68724	Gibberella zeae PH-1	hypothetical protein FG00492
16F12	CN809284	4b	RNA modification	CAA63557	Anabaena variabilis	RNA-binding protein

17F2	CN809433	7b	Detoxification	AAF22254	Yarrowia lipolytica	integral peroxisomal membrane protein involved in negative regulation of peroxisome number
17F4	CN809435	9	Hyp/Unknown Prot	EAA50265	Magnaporthe grisea 70-15	hypothetical protein MG04024.4
17F6	CN809440	2	Energy Metabolism	CAF32039	Aspergillus fumigatus	cytochrome p450 (E-class), putative
17F8	CN809442	9	Hyp/Unknown Prot	EAA16547	Plasmodium yoelii yoelii	hypothetical protein
17F10	CN809445	5d	Transport proteins	CAB43936	Emericella nidulans	GABA (gamma-amino-n-butyrate) permease
17F12	CN809449	1e	N-, P-, S-metabolism	CAF32031	Aspergillus fumigatus	smr family protein, putative, in nitrate assimilation gene cluster
18F2	CN809611	9	Hyp/Unknown Prot	EAA69510	Gibberella zeae PH-1	hypothetical protein FG00959
18F4	CN809615	9	Hyp/Unknown Prot	EAA58144	Aspergillus nidulans	hypothetical protein AN6615.2
18F6	CN809621	4c	Transcription	NP_012420	Saccharomyces cerevisiae	anti-silencing protein that causes depression of silent loci when overexpressed; Asf1p
18F8	CN809625	9	Hyp/Unknown Prot			unknown
18F10	CN809627	3a	Ribosomal proteins	AAO47090	Neurospora crassa	ribosomal L10 protein
18F12	CN809631	5d	Transport proteins	NP_595211	Schizosaccharomyces pombe	putative transmembrane transporter
16H1	CN809304	4c	Transcription	AAB69694	Magnaporthe grisea	putative transcriptional regulator
16H3	CN809306	9	Hyp/Unknown Prot			Unknown
16H5	CN809308	1c	Lipid metabilism	BAD01582	Aspergillus oryzae	secretory phospholipase A2
16H7	CN809311	9	Hyp/Unknown Prot	EAA47331	Magnaporthe grisea	hypothetical protein MG02574.4
16H9	CN809314	2	Energy Metabolism	P23704	Neurospora crassa	ATP synthase beta chain, mitochondrial precursor
16H11	CN809320	2	Energy Metabolism	O58669	Pyrococcus horikoshii	NAD-dependent deacetylase (Regulatory protein SIR2 homolog)
17H1	CN809467	9	Hyp/Unknown Prot	EAA72664	Gibberella zeae PH-1	hypothetical protein FG08636
17H3	CN809469	9	Hyp/Unknown Prot	EAA73470	Gibberella zeae PH-1	hypothetical protein FG04002
17H5	CN809474	5f	Signaling	CAB89868	Kluyveromyces lactis	putative membrane protein,putative high osmolarity sensor
17H7	CN809478	9	Hyp/Unknown Prot	EAA70677	Gibberella zeae PH-1	hypothetical protein FG00731
17H9	CN809486	1d	Nucleotide metabolism	EAA65920	Aspergillus nidulans	Positive regulator of purine utilization
17H11	CN809488	9	Hyp/Unknown Prot	NP_598212	Rattus norvegicus	preimplantation protein 3
18H1	CN809660	1b	Carbohydrate metabolism	O93806	Candida albicans	Glucosamine 6-phosphate N-acetyltransferase (Phosphoglucosamine transacetylase) (Phosphoglucosamine acetylase)
18H3	CN809665	9	Hyp/Unknown Prot	EAA74506	Gibberella zeae PH-1	hypothetical protein FG10899
18H5	CN809668	9	Hyp/Unknown Prot	EAA73525	Gibberella zeae PH-1	hypothetical protein FG04199
18H7	CN809672	3d	Prot modification/targeting	AAG09748	Penicillium chrysogenum	peroxin-1, involved in peroxisome biogenesis
18H9	CN809674	9	Hyp/Unknown Prot			unknown
18H11	Y10265	1b	Carbohydrate metabolism		Metarhizium anisopliae 2575	carbon responsse regulator
16H2	CN809305	9	Hyp/Unknown Prot	EAA78349	Gibberella zeae PH-1	hypothetical protein FG06564
16H4	CN809307	3d	Prot modification/targeting	NP_010769	Saccharomyces cerevisiae	repressible alkaline phosphatase; Pho8p

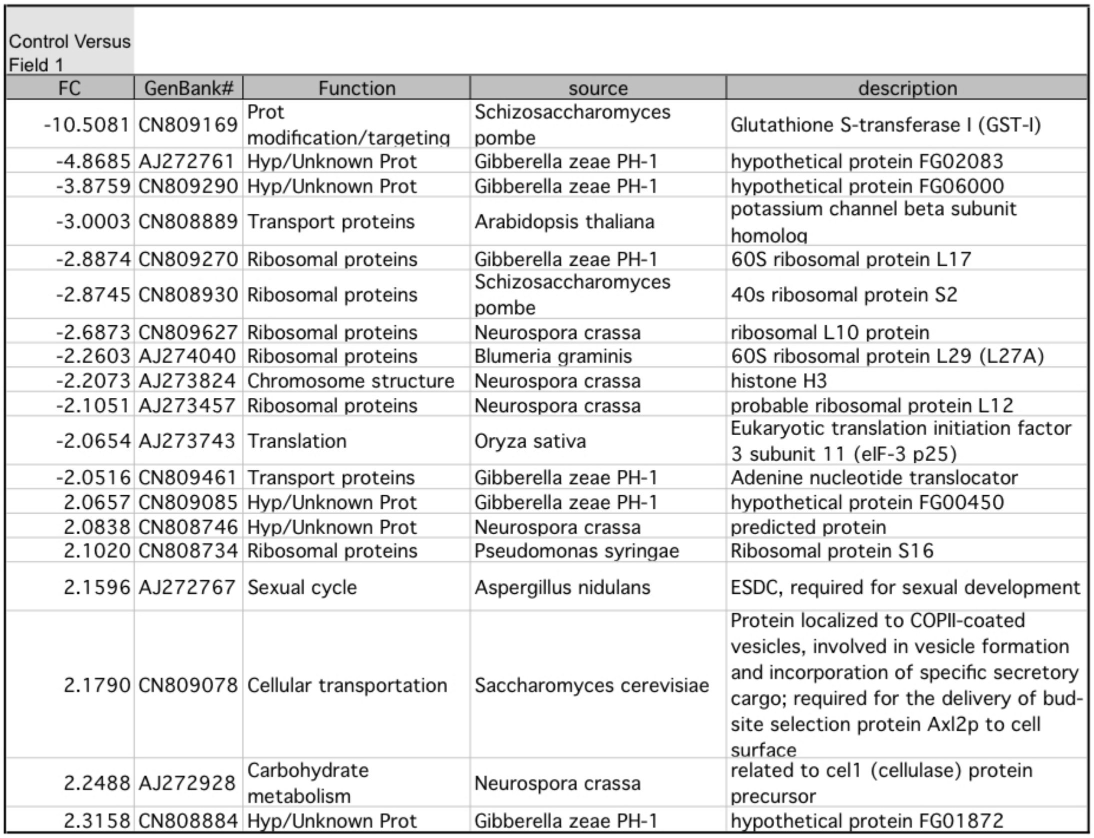
16H6	CN809309	9	Hyp/Unknown Prot	EAA71897	Gibberella zeae PH-1	hypothetical protein FG08420
16H8	CN809313	7a	Stress response	AAQ14297	Gibberella acuminata	ice nucleation protein
16H10	CN809316	1b	Carbohydrate metabolism	BAA31433	Yarrowia lipolytica	n-alkane-inducible cytochrome P450 gene (ALK1) essential for n-decane assimilation
16H12	CN809321	1c	Lipid metabilism	NP_495415.2	Caenorhabditis elegans	acid Sphingomyelinase, sphingomyelin phosphodiesterase (asm-1)
17H2	CN809468	3d	Prot modification/targeting	NP_588169	Schizosaccharomyces pombe	possible involvement in nuclear protein localisation
17H4	CN809472	3d	Prot modification/targeting	CAE75717	Neurospora crassa	related to EDE1 protein, Key endocytic protein involved in a network of interactions with other endocytic proteins, binds membranes in a ubiquitin-dependent manner
17H6	CN809475	9	Hyp/Unknown Prot	EAA75072	Gibberella zeae PH-1	hypothetical protein FG06130
17H8	CN809485	9	Hyp/Unknown Prot			unknown
17H10	CN809487	1b	Carbohydrate metabolism	CAD21128	Neurospora crassa	probable aldehyde dehydrogenase
17H12	CN809490	9	Hyp/Unknown Prot	EAA64348	Aspergillus nidulans	hypothetical protein AN9016.2
18H2	CN809664	3b	Translation	CAE76104	Neurospora crassa	probable translation elongation factor eEF-1, gamma chain
18H4	CN809666	9	Hyp/Unknown Prot	EAA71263	Gibberella zeae PH-1	hypothetical protein FG03359
18H6	CN809669	9	Hyp/Unknown Prot	EAA68153	Gibberella zeae PH-1	hypothetical protein FG01527
18H8	CN809673	2	Energy Metabolism	ZP_00183705	Exiguobacterium sp.	NAD-dependent aldehyde dehydrogenases
SSC7						
18H12	AJ006468	1e	N-, P-, S-metabolism		Metarhizium anisopliae 2575	nitrogen response regulator

Appendix B

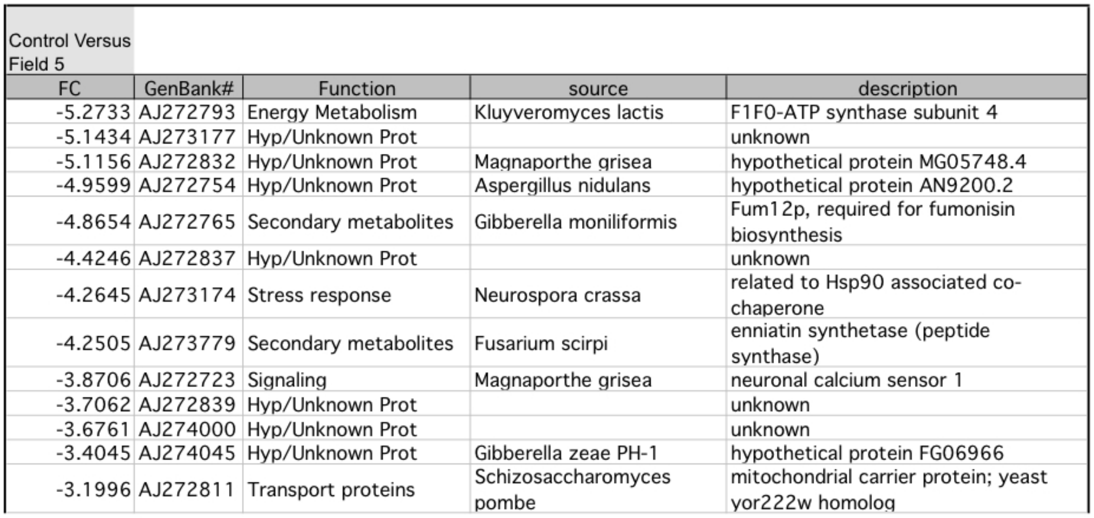
Tables with Genes listed that had non-significant fold changes in gene expression greater that 2.0



Control versus Control				
FC	GenBank#	Function	source	description
-2.2917	CN808945	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG09718



Control Versus Field 1				
FC	GenBank#	Function	source	description
-10.5081	CN809169	Prot modification/targeting	Schizosaccharomyces pombe	Glutathione S-transferase I (GST-I)
-4.8685	AJ272761	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG02083
-3.8759	CN809290	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG06000
-3.0003	CN808889	Transport proteins	Arabidopsis thaliana	potassium channel beta subunit homolog
-2.8874	CN809270	Ribosomal proteins	Gibberella zeae PH-1	60S ribosomal protein L17
-2.8745	CN808930	Ribosomal proteins	Schizosaccharomyces pombe	40s ribosomal protein S2
-2.6873	CN809627	Ribosomal proteins	Neurospora crassa	ribosomal L10 protein
-2.2603	AJ274040	Ribosomal proteins	Blumeria graminis	60S ribosomal protein L29 (L27A)
-2.2073	AJ273824	Chromosome structure	Neurospora crassa	histone H3
-2.1051	AJ273457	Ribosomal proteins	Neurospora crassa	probable ribosomal protein L12
-2.0654	AJ273743	Translation	Oryza sativa	Eukaryotic translation initiation factor 3 subunit 11 (eIF-3 p25)
-2.0516	CN809461	Transport proteins	Gibberella zeae PH-1	Adenine nucleotide translocator
2.0657	CN809085	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG00450
2.0838	CN808746	Hyp/Unknown Prot	Neurospora crassa	predicted protein
2.1020	CN808734	Ribosomal proteins	Pseudomonas syringae	Ribosomal protein S16
2.1596	AJ272767	Sexual cycle	Aspergillus nidulans	ESDC, required for sexual development
2.1790	CN809078	Cellular transportation	Saccharomyces cerevisiae	Protein localized to COPII-coated vesicles, involved in vesicle formation and incorporation of specific secretory cargo; required for the delivery of bud-site selection protein Axl2p to cell surface
2.2488	AJ272928	Carbohydrate metabolism	Neurospora crassa	related to cel1 (cellulase) protein precursor
2.3158	CN808884	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG01872



Control Versus Field 5				
FC	GenBank#	Function	source	description
-5.2733	AJ272793	Energy Metabolism	Kluyveromyces lactis	F1F0-ATP synthase subunit 4
-5.1434	AJ273177	Hyp/Unknown Prot		unknown
-5.1156	AJ272832	Hyp/Unknown Prot	Magnaporthe grisea	hypothetical protein MG05748.4
-4.9599	AJ272754	Hyp/Unknown Prot	Aspergillus nidulans	hypothetical protein AN9200.2
-4.8654	AJ272765	Secondary metabolites	Gibberella moniliformis	Fum12p, required for fumonisin biosynthesis
-4.4246	AJ272837	Hyp/Unknown Prot		unknown
-4.2645	AJ273174	Stress response	Neurospora crassa	related to Hsp90 associated co-chaperone
-4.2505	AJ273779	Secondary metabolites	Fusarium scirpi	enniatin synthetase (peptide synthase)
-3.8706	AJ272723	Signaling	Magnaporthe grisea	neuronal calcium sensor 1
-3.7062	AJ272839	Hyp/Unknown Prot		unknown
-3.6761	AJ274000	Hyp/Unknown Prot		unknown
-3.4045	AJ274045	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG06966
-3.1996	AJ272811	Transport proteins	Schizosaccharomyces pombe	mitochondrial carrier protein; yeast yor222w homolog

Control versus Control

FC	GenBank#	Function	source	description
-2.2917	CN808945	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG09718

Control Versus Field 1

FC	GenBank#	Function	source	description
-10.5081	CN809169	Prot modification/targeting	Schizosaccharomyces pombe	Glutathione S-transferase I (GST-I)
-4.8685	AJ272761	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG02083
-3.8759	CN809290	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG06000
-3.0003	CN808889	Transport proteins	Arabidopsis thaliana	potassium channel beta subunit homolog
-2.8874	CN809270	Ribosomal proteins	Gibberella zeae PH-1	60S ribosomal protein L17
-2.8745	CN808930	Ribosomal proteins	Schizosaccharomyces pombe	40s ribosomal protein S2
-2.6873	CN809627	Ribosomal proteins	Neurospora crassa	ribosomal L10 protein
-2.2603	AJ274040	Ribosomal proteins	Blumeria graminis	60S ribosomal protein L29 (L27A)
-2.2073	AJ273824	Chromosome structure	Neurospora crassa	histone H3
-2.1051	AJ273457	Ribosomal proteins	Neurospora crassa	probable ribosomal protein L12
-2.0654	AJ273743	Translation	Oryza sativa	Eukaryotic translation initiation factor 3 subunit 11 (eIF-3 p25)
-2.0516	CN809461	Transport proteins	Gibberella zeae PH-1	Adenine nucleotide translocator
2.0657	CN809085	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG00450
2.0838	CN808746	Hyp/Unknown Prot	Neurospora crassa	predicted protein
2.1020	CN808734	Ribosomal proteins	Pseudomonas syringae	Ribosomal protein S16
2.1596	AJ272767	Sexual cycle	Aspergillus nidulans	ESDC, required for sexual development
2.1790	CN809078	Cellular transportation	Saccharomyces cerevisiae	Protein localized to COPII-coated vesicles, involved in vesicle formation and incorporation of specific secretory cargo; required for the delivery of bud-site selection protein Axl2p to cell surface
2.2488	AJ272928	Carbohydrate metabolism	Neurospora crassa	related to cel1 (cellulase) protein precursor
2.3158	CN808884	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG01872

Control Versus Field 5

FC	GenBank#	Function	source	description
-5.2733	AJ272793	Energy Metabolism	Kluyveromyces lactis	F1F0-ATP synthase subunit 4
-5.1434	AJ273177	Hyp/Unknown Prot		unknown
-5.1156	AJ272832	Hyp/Unknown Prot	Magnaporthe grisea	hypothetical protein MG05748.4
-4.9599	AJ272754	Hyp/Unknown Prot	Aspergillus nidulans	hypothetical protein AN9200.2
-4.8654	AJ272765	Secondary metabolites	Gibberella moniliformis	Fum12p, required for fumonisin biosynthesis
-4.4246	AJ272837	Hyp/Unknown Prot		unknown
-4.2645	AJ273174	Stress response	Neurospora crassa	related to Hsp90 associated co-chaperone
-4.2505	AJ273779	Secondary metabolites	Fusarium scirpi	enniatin synthetase (peptide synthase)
-3.8706	AJ272723	Signaling	Magnaporthe grisea	neuronal calcium sensor 1
-3.7062	AJ272839	Hyp/Unknown Prot		unknown
-3.6761	AJ274000	Hyp/Unknown Prot		unknown
-3.4045	AJ274045	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG06966
-3.1996	AJ272811	Transport proteins	Schizosaccharomyces pombe	mitochondrial carrier protein; yeast yor222w homolog

2.6864	AJ273529	Ribosomal proteins	Schizosaccharomyces pombe	60S ribosomal protein L19B
3.3571	CN808746	Hyp/Unknown Prot	Neurospora crassa	predicted protein

Filed 1 versus Field 2

FC	GenBank#	Function	source	description
-3.4597	AJ274215	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG00806
-3.3044	CN809627	Ribosomal proteins	Neurospora crassa	ribosomal L10 protein
-3.2169	AJ272778	Hyp/Unknown Prot	Aspergillus nidulans	hypothetical protein AN2582.2
-2.6097	AJ273011	Cell wall	Trichoderma harzianum	b-1,3 exoglucanase
-2.4420	AJ273680	Lipid metabilism	Streptomyces avermitilis	NADH-dependent, putative enoyl-ACP reductase II
-2.3503	AJ273928	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG08359
-2.2971	AJ273743	Translation	Oryza sativa	Eukaryotic translation initiation factor 3 subunit 11 (eIF-3 p25)
-2.2715	CN808255	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG02791
-2.2310	AJ272932	Hyp/Unknown Prot	Magnaporthe grisea	hypothetical protein MG06953.4
-2.1891	AJ273268	Signaling	Cochliobolus heterostrophus	GTPase activating protein homolog
-2.0451	AJ273824	Chromosome structure	Neurospora crassa	histone H3
-2.0108	CN808740	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG05657

Field 2 versus Field 3

FC	GenBank#	Function	source	description
-6.9799	AJ273435	Ribosomal proteins	Candida maltosa	40S ribosomal protein S13 (S15)
-5.3053	CN808241	Aminoacid metabolism	Saccharomyces cerevisiae	putative alanine transaminase (glutamyc pyruvic transaminase); Ylr089cp
-4.7651	AJ273011	Cell wall	Trichoderma harzianum	b-1,3 exoglucanase
-4.6995	CN809475	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG06130
-4.1722	AJ273824	Chromosome structure	Neurospora crassa	histone H3
-3.8129	AJ273680	Lipid metabilism	Streptomyces avermitilis	NADH-dependent, putative enoyl-ACP reductase II
-3.6544	CN808557	Transport proteins	Neurospora crassa	related to NCE102 protein: Non-classical export protein 2
-3.2079	AJ273125	Cell wall	Saccharomyces cerevisiae	Protein with similarity to Crh1p, which is a putative glycosidase of the cell wall; Crr1p (cell wall organization and biogenesis)
-2.7444	CN809371	Hyp/Unknown Prot	Neurospora crassa	hypothetical protein
-2.6903	CN808255	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG02791
-2.6364	AJ274215	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG00806
-2.5657	AJ273975	Lipid metabilism	Neurospora crassa	lanosterol synthase related protein
-2.5349	CN808445	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG09950
-2.4862	AJ272713	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG01872
-2.3729	CN808448	Lipid metabilism	Chaetomium globosum	dienelactone hydrolase
-2.3434	CN808930	Ribosomal proteins	Schizosaccharomyces pombe	40s ribosomal protein S2
-2.0890	CN808375	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG00663
-2.0856	CN808221	Lipid metabilism	Schizosaccharomyces pombe	C-4 methyl sterol oxidase
-2.0697	CN809627	Ribosomal proteins	Neurospora crassa	ribosomal L10 protein

Field 3 versus Field 4

FC	GenBank#	Function	source	description
-9.6262	AJ272793	Energy Metabolism	Kluyveromyces lactis	F1F0-ATP synthase subunit 4
-9.3533	AJ272799	Cell wall	Aspergillus fumigatus	GEL1 protein
-7.6468	AJ274045	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG06966

-6.4936	AJ274064	Cell cycle	Neurospora crassa	related to cell cycle regulation and aging protein
-6.4375	CN808905	Ribosomal proteins	Neurospora crassa	probable 40s ribosomal protein S6.e, cytosolic
-4.7999	AJ272778	Hyp/Unknown Prot	Aspergillus nidulans	hypothetical protein AN2582.2
-4.5880	CN808746	Hyp/Unknown Prot	Neurospora crassa	predicted protein
-4.5386	AJ272811	Transport proteins	Schizosaccharomyces pombe	mitochondrial carrier protein; yeast yor222w homolog
-4.1968	AJ273975	Lipid metabilism	Neurospora crassa	lanosterol synthase related protein
-4.1725	AJ272832	Hyp/Unknown Prot	Magnaporthe grisea	hypothetical protein MG05748.4
-4.0436	CN809302	Prot modification/targeting	Neurospora crassa	probable cyclophilin: catalyzes the cis-trans isomerization of peptide bonds N-terminal to proline residues
-3.9371	CN809615	Hyp/Unknown Prot	Aspergillus nidulans	hypothetical protein AN6615.2
-3.6152	AJ273072	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG06022
-3.5876	AJ273776	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG06022
-3.2973	AJ273680	Lipid metabilism	Streptomyces avermitilis	NADH-dependent, putative enoyl-ACP reductase II
-3.2057	AJ274000	Hyp/Unknown Prot		unknown
-3.1276	AJ273943	Hyp/Unknown Prot		unknown
-3.0888	AJ272765	Secondary metabolites	Gibberella moniliformis	Fum12p, required for fumonisin biosynthesis
-2.9439	CN808542	Cell wall	Aspergillus kawachii	endoglucanase B
-2.9336	AJ274215	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG00806
-2.9250	AJ273011	Cell wall	Trichoderma harzianum	b-1,3 exoglucanase
-2.8019	CN808789	Hyp/Unknown Prot	Metarhizium anisopliae	cystein rich protein
-2.7160	CN809258	Nucleotide metabolism	Homo sapiens	similar to rRNA intron-encoded homing endonuclease
-2.7103	AJ272809	Aminoacid metabolism	Neurospora crassa	putative ornithine carbamoyl transferase precursor
-2.6734	CN808732	Transport proteins	Schizosaccharomyces pombe	putative translocation protein
-2.6666	AJ272728	Cell cycle	Neurospora crassa	related to sporulation protein SPO72
-2.6369	AJ273248	Energy Metabolism	Neurospora crassa	H+-transporting ATP synthase a chain
-2.6079	AJ273870	Cell death	Pisum sativum	putative senescence-associated protein
-2.5980	AJ274041	Proteolysis	Homo sapiens	lysosomal pepstatin insensitive protease
-2.5762	AJ274191	Carbohydrate metabolism	Vibrio cholerae	prpE protein, similar to acetate-CoA ligase
-2.5637	AJ273114	Carbohydrate metabolism	Aspergillus tubingensis	ferulic acid esterase A
-2.3412	AJ274105	Hyp/Unknown Prot	Neurospora crassa	hypothetical protein
-2.3131	AJ273930	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG00855
-2.2756	CN809192	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG00804
-2.2751	AJ274333	Aminoacid metabolism	Rhodococcus erythropolis	putative peptidase
-2.2433	CN809178	Cell wall	Magnaporthe grisea	hydrophobin-like protein
-2.2288	AJ272839	Hyp/Unknown Prot		unknown
-2.2063	AJ273711	Cell polarity	Schizosaccharomyces pombe	possibly involved in cell polarity; by similarity to yeast rax2
-2.1992	AJ272761	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG02083
-2.1861	AJ272932	Hyp/Unknown Prot	Magnaporthe grisea	hypothetical protein MG06953.4
-2.1844	AJ273407	Hyp/Unknown Prot		Unknown
-2.1453	AJ272808	Hyp/Unknown Prot		unknown
-2.1344	CN808371	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG00791
-2.1334	AJ274102	Proteolysis	Metarhizium anisopliae	carboxypeptidase
-2.0940	CN809442	Hyp/Unknown Prot	Plasmodium yoelii yoelii	hypothetical protein
-2.0818	AJ272771	Hyp/Unknown Prot		unknown
-2.0759	AJ273411	Nucleotide metabolism	Neurospora crassa	probable adenylate kinase

-2.0366	AJ272786	Proteolysis	Schizosaccharomyces pombe	Highly conserved ring finger; similar to yeast apc11 SPC(ubiquitin-protein ligase activity)
-2.0336	AJ273781	Hyp/Unknown Prot		unknown
2.1145	CN808135	Hyp/Unknown Prot	Magnaporthe grisea	hypothetical protein MG09337.4
2.1260	CN808151	Transposable Elements	Fusarium poae	gag-like polyprotein
2.1547	CN807958	Hyp/Unknown Prot		Unknown
2.1947	AJ273303	Cell wall	Kluyveromyces marxianus var. lactis	WSC4 homolog, WSC4p is involved in cell wall integritz and stress response
2.3956	CN808204	Detoxification	Aspergillus fumigatus	squalene epoxidase 1: confering resistance to antifungal terbinafine

Field 4 versus Field 5

FC	GenBank#	Function	source	description
-3.6905	AJ272841	Hyp/Unknown Prot	Neurospora crassa	predicted protein
-3.5676	AJ272839	Hyp/Unknown Prot		unknown
-2.7737	CN808958	Proteolysis	Metarhizium anisopliae	subtilisin-like serine protease PR1A
-2.4644	AJ273943	Hyp/Unknown Prot		unknown
-2.4623	CN808878	Ribosomal proteins	Arabidopsis thaliana	rhomboid protein-related
-2.4421	AJ274002	Energy Metabolism	Rhizobium sp. NGR234	probable Oxidoreductase
-2.3730	AJ274093	Hyp/Unknown Prot		Unknown
-2.3320	CN808927	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG00854
-2.2798	AJ273623	Carbohydrate metabolism	Phaeosphaeria avenaria	b-glucosidase
-2.2685	AJ272728	Cell cycle	Neurospora crassa	related to sporulation protein SPO72
-2.2332	AJ272713	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG01872
-2.1585	AJ273529	Ribosomal proteins	Schizosaccharomyces pombe	60S ribosomal protein L19B
-2.0682	AJ272761	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG02083
2.0003	CN808472	Cell wall	Aspergillus fumigatus	chitinase
2.1101	CN809038	Carbohydrate metabolism	Aspergillus nidulans	delta-1-pyrroline-5-carboxylate dehydrogenase
2.1406	CN808816	Carbohydrate metabolism	Escherichia coli K12	2-methylcitrate dehydratase
2.1542	CN808112	Stress response	Saccharomyces cerevisiae	UBI4 locus contains five-six tandem, in-frame copies of ubiquitin ; transcription is induced in response to several stress conditions: Ubi4p
2.1719	AJ274045	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG06966
2.2132	AJ273651	Hyp/Unknown Prot	Neurospora crassa	probable endopolyphosphatase (EC 3.6.1.10) B24P7.140 precursor
2.2176	CN809647	Hyp/Unknown Prot	Gibberella zeae PH-1	hypothetical protein FG08554
2.3157	CN809302	Prot modification/targeting	Neurospora crassa	probable cyclophilin: catalyzes the cis-trans isomerization of peptide bonds N-terminal to proline residues
2.3318	CN809258	Nucleotide metabolism	Homo sapiens	similar to rRNA intron-encoded homing endonuclease
2.5151	CN809336	Hyp/Unknown Prot	Neurospora crassa	hypothetical protein
2.9345	CN809615	Hyp/Unknown Prot	Aspergillus nidulans	hypothetical protein AN6615.2
4.1870	AJ273797	Hyp/Unknown Prot	Schizosaccharomyces pombe	hypothetical protein SPBC2A9.07c
4.2436	AJ272974	Transcription	Neurospora crassa	related to prefoldin subunit 1: contain a DNA binding leucine zipper motif
4.5575	AJ272778	Hyp/Unknown Prot	Aspergillus nidulans	hypothetical protein AN2582.2
7.4313	CN809178	Cell wall	Magnaporthe grisea	hydrophobin-like protein

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