THE CONTROL OF A FUNGAL OUTBREAK IN A SHOW CAVE

CESAREO SAIZ-JIMENEZ[1], VALME JURADO[1], ESTEFANIA PORCA[1], SOLEDAD CUEZVA[2], ANGEL FERNANDEZ-CORTES[2], and SERGIO SANCHEZ-MORAL[2]

[1]Instituto de Recursos Naturales y Agrobiologia, CSIC, Apartado 1052, 41080 Sevilla, Spain

[2]Museo Nacional de Ciencias Naturales, CSIC, 28006 Madrid, Spain

Castañar de Ibor Cave (Spain) is a low energy cave with very high microenvironmental stability throughout the annual cycle and minimum rates of energy exchange with the atmosphere. This show cave was discovered in 1967 and declared a Natural Monument in 1997. Later in 2003 the cave was opened to public visits. On 26 August 2008, the cave walls and soils appeared colonized by long, white fungal mycelia. This event was the result of an accidental input of organic matter on the afternoon of 24 August 2008. The outbreak was initiated by *Mucor circinelloides* and *Fusarium solani*.

1. Introduction

In recent years, European caves have been suffering episodes of biological contamination that threaten the conservation of these geological and cultural heritage sites. Until now, there was no direct evidence on the causes of fungal contamination in caves, such as those that occurred in 2001 in Lascaux Cave, France, after a heavy fungal outbreak (BASTIAN AND ALABOUVETTE, 2009). Opening of a cave to visitors represents a strong and often irreversible impact on the whole ecosystem. Bacteria, fungi, arthropods, etc. have constructed delicate and balanced trophic relationships in caves, which are frequently interrupted by major environmental changes as a consequence of visits. These changes include concrete stairs, air conditioning (BASTIAN et al., 2009) and elevators, etc., which are some of the examples of changes found in European caves.

Castañar de Ibor (Caceres, Spain) is defined as a 'low energy' cave showing very high micro-environmental stability throughout the annual cycle, under natural conditions (LARIO et al., 2006). This large microclimatic stability is a basic characteristic of karst systems and causes a high sensitivity by the ecosystem to changes in environmental conditions, such as temperature and anthropic activities. Therefore, in 2003, a strict control was established for visitors.For instance, during 2007 only 1,010 people accessed the cave with a maximum quota of 5 visitors per group and an average stay of 65 minutes.

2. Cave Microbiology

On the afternoon of August 24, 2008, accidental vomiting by a visitor led to the input of detritus into Castañar de Ibor Cave.The effects of this impact were not observed until August 26, 2008, when cave walls and soil demonstrated long, white fungal mycelia. On September 4, three samples were obtained from the disturbed area, with additional sampling on September 11 and October 28, 2008.

The fungal outbreak in the cave coincided with the period of greatest air exchange between the cave and the outer atmosphere. During summer (from June to October) the exterior air temperature is constantly above the cave air temperature.The external relative humidity falls below 60% and registered rainfall represents 33.4% of the annual total over a period of 130 days. These factors provoke underground air renewal and, therefore, the dispersion of fungi in the cavity.

From 20 samples collected from the cave, all of them showing fungal growth, we have retrieved a total of 26 isolates (Table 1). While 10-20 days after the event, only two fungi were identified: *Mucor circinelloides* and *Fusarium solani*, representing up to 75 and 25 % of the isolates, they decreased to 13 and 7 % two months after the outbreak. These two fungi behaved as "r" strategists (FRANKLAND, 1992), and were able to occupy the habitat rapidly with explosive rates of reproduction. *Mucor circinelloides* produces long hyphae, with a fluffy appearance, that can grow to a height of several cm. This fungus is common in caves, and colonizes animal feces and insects (NOVÁKOVÁ, 2009).

Spores disseminated by rodent feces and insects - also observed in this cave - germinated rapidly in response to available nutrients. *Fusarium solani* is also frequently found in caves (JURADO et al., 2008). Members of the *Fusarium solani* are ubiquitous plant pathogens and saprotrophs. This was also the fungus found invading Lascaux Cave in 2001 (BASTIAN AND ALABOUVETTE, 2009) and the Takamatsuzuka and Kitora Tumuli in Japan (KIYUNA, 2008).

Fungal successions on decomposing organic matter are

	September 4, 2008	September 11, 2008	October 28, 2008
Number of samples	3	7	10
Number of isolates	3	8	15
Mucor circinelloides	2	6	2
Fusarium solani	1	2	1
Mucor racemosus	-	-	1
Fusarium oxysporum	-	-	2
Chaetomium globosum	-	-	1
Mortierella alpina	-	-	1
Hypocrea lixii	-	-	1
Aspergillus ustus	-	-	2
Paecilomyces lilacinus	-	-	1
Cosmospora consors	-	-	1
Penicillium sp.	-	-	2

Table 1:.Fungi identified in the outbreak of Castañar de Ibor Cave.

rarely followed to an end-point. In this case, it was possible to follow the fungal succession in the cave and to identify secondary colonizers. In fact, other fungi such as *Fusarium oxysporum, Chaetomium globosum, Mortierella alpina, Hypocrea lixii, Aspergillus ustus, Paecilomyces lilacinus, Cosmospora consors* and a *Penicillium* sp. were isolated two months after the disturbance event from small white spots in the soil. Most of these fungi are necrotrophic, which are characterized by their ability to use dead plant or animal tissues as a source of nutrients. Some others have been reported to be entomopathogens (i.e. disease agents in insects), such as *M. circinelloides, F. solani, F. oxysporum,* and *Paecilomyces lilacinus* (HUMBER AND HANSEN, 2005).

Treatments to stop the fungal outbreak in Castañar de Ibor Cave included a careful mechanical removal of a few cm of the polluted top sediment and sterilization of the underlying materials with commercial hydrogen peroxide (FAIMON et al., 2003). The treatment was repeated when necessary, and after the initial outbreak the cave only presented scattered white colonizations that appeared from time to time along the visitor trail as well as some colonized rodent feces. These small spots were likely due to the detritus transported inside by former visitor footsteps. No colonizations were observed in the sediment of non-visited halls and galleries, other than in some rodent feces. Therefore, it appears that the strategy to control the outbreak was successful.

This study provided evidence of the fragility of show caves and demonstrated that visits may compromise conservation. This raises an intriguing question: Should rock art caves be subjected to the ravages of visits or should they be protected from signficant visitation and associated disturbance events, as in the case of some French caves that have never been open to public visits?

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