Isolation and Identification of Fungal Species and Scientific Preservation of World Heritage: Case of Fatehpur Sikri, Uttar Pradesh, India

Sanjay Prasad Gupta
National Research Laboratory for Conservation of Cultural Property, Lucknow, India

Sachin Kumar Agnihotri
Archaeological Survey of India, Science Branch, Agra, India

Abstract:
Fungi play a considerable role for the deterioration and widely recognized as major bio-deteriogens of cultural heritage depending on climatic conditions, humidity level and surface material for fungal colonization. The weathering of stone monuments is significantly increased by endolitic fungi. Fungi can deteriorate different substrates via various physical and chemical mechanisms. Hyphal growth and penetration into the substrate can cause symptoms like discoloration, bio-pitting, cricketing, exfoliation and patina formation. On the other hand, chemical mechanism includes acid secretion, release of extracellular enzymes, pigment production, and secondary mycogenic minerals formation. These processes can lead to serious, both esthetic and structural alterations which may be irreversible and could permanently impair artwork. Proper isolation and identification of fungi by different microscopic technique and in vitro biodegradation tests are pivotal in understanding complex bio-deterioration mechanism caused by fungal deteriogens. Bio-deterioration and bio-degradation studies require multidisciplinary approach and close collaboration of microbiologists, chemists and different personnel responsible for safeguarding of cultural property and artifacts, especially restorers and conservators. This article provides information on fungi infesting historical monument of Fatehpur Sikri and their management by biocidal compounds. Present investigation was conducted to evaluate the status of fungal decay of stone monuments of Fatehpur Sikri. A total of 06 fungal species were isolated from colored stains, patinas and biofilms produced on the surfaces of monuments of Fatehpur Sikri due to mechanism of bio-deterioration. The fungal species Alternaria alternata, Aspergillus fumigatus, Aspergillus flavus, Aspergillus niger, Cladosporium herbarum, and Rhizopus oryzae were prevalent.

Keywords: Biodegradation, biocidal treatment, cultural heritage, fungi, historical monuments, endolitic fungi, hyphal growth, discoloration and extracellular enzymes etc.

Introduction

History of Monument
World Heritage site and centrally protected monument of Fatehpur Sikri is an extension of the upper Vindhyan ranges located about 40 km south west of Agra on the bank of dried up natural lake. Due to abundant water, forest and raw material, the area was ideal for primitive man’s habitation. Besides, stone age tools, rock
shelters with paintings, Ochre colored Pottery (Circa 2nd millennium B.C.) and Painted Grey Ware (Circa 1200-800 B.C.) have also been found in this area. Sikri has been mentioned in the Mahabharata as ‘Saik’. Lexicons define ‘Saik’ as a region surrounded by water. An inscription found on the stone sculpture of Jaina Saraswati (dated 11th century mentions this place as ‘Sekriya’ which seems to be a similar derivative. All this shows that Sikri was continuously inhabitate since the prehistoric period. Babur visited here on the eve of the Khanwah battle in A.D.1527 and mentioned it as ‘Sikri’ in his memoirs. Akbar (1556-1605) grandson of Babur shifted his residence and court from Agra to Sikri, for a period of 13 years, from 1572 to 1585 to honour the sufi saint Sheikh Salim Chishti, who resided here in a cavern on the ridge. Akbar revered him very much as the Saint had blessed him with a son named Salim in 1569. He raised lofty buildings for his use, and houses for the public. Thus grew a great city with charming palaces and institutions. Akbar gave it the name of Fatehabad which in later days came to be known as “Fatehpur Sikri”.

Buland Darwaza, Fatehpur Sikri

The Buland Darwaza is made up of red and buff sandstone, decorated by white and black marble and is higher than the courtyard of the mosque. The Buland Darwaza is symmetrical and is topped by large free standing kiosks, which are then. It also has at the top centre, terrace edge gallery-kiosks on the roof, stylized buckler-battlements, small miner-spires, and inlay work with white and black marble. On the outside a long flight of steps sweeps down the hill giving the gateway additional height. It is 40 metres high and 50 metres above from the ground. The total height of the structure is about 54 metres from the ground level. It is a 15-storied high gateway acting as the southern entrance of the city of Fatehpur Sikri. The approach to the gate consists of 42 steps. It is semi octagonal in plan and two smaller triple-storeyed wings on either side. It has three kiosks on its top surrounded by thirteen smaller domed kiosks. There are smaller turrets surrounding the gateway.

Conservation Issue

The small chhatris and sand stone surface of the monument is covered with dust, dirt and other tarry matters. The exterior surface of the small chhatris also covered by deposition of algae, dried moss and bacterial slime. Due to deposition of micro-organism on lime plastered small chhatris/burjies, they become blackish in appearance and looking. This is extremely essential to clean and preserve the monument to retain its aesthetic value. Apart from dust, dirt and other tarry matters along with micro vegetation growth, there are number of bee hives at the ceiling of big arch, they stick to surface and urinate on the surface resulting formation of dark patches on stone surface.
There are also depositions of Pigeons excreta on stone surface, which formed a very thick layer of accretions and hiding the original texture of the monument. It has been noticed that at few places the red sand stone surface become porous and fragile. Also, due to continuous touching by visitors on sand stone surface become dirty and blackish in appearance. Human vandalism can also be seen at lower portion of Buland Darwaza.

**Fungal Deterioration of the Monument**

The weathering and decay of cultural heritage is a complex process, which is caused by the interaction of many physical, chemical and biological agents. The different biological deterioriogens such as bacteria, algae, cyanobacteria, bryophytes, mosses, fungi, insects, rodents, birds and human beings play a momentous role in the decay of historical monuments. The biological growth of microorganisms can cause staining, cracking, powdering, disfigurement and displacement of building material, which leads to the permanent loss of stone monuments. Such aesthetic damage is accompanied by the transformation of the chemical and mechanical properties of stone material and this causes the formation of surface patinas of different colors. Among different biological agents, fungi play more dangerous role in the bio-deterioration of stone monuments because of their complex metabolic activities on stone surface. The growth of the fungal genera on stone monuments was a cause of staining and structural decay of stone material of these monuments. Fungi produces many inorganic and organic acids during their metabolic activities on monuments. These acids cause mineral dissolution and change structural configuration of stone material. The organic acids such as oxalic, lactic and gluconic acids function as chelating agents and can demineralized a variety of stone substrates including calcium, silicon, iron, magnesium and manganese. The enzymes produce during metabolic activities of fungi are involved in transformation of complex and binding molecules of stone monuments into simple dissolvable molecules. Fungal metabolites can cause solubilization of cations and produce patinas of different mineralogical composition. The discoloration of monuments due to the formation of patinas is more havoc on light colored stone monuments. The interaction between fungal hyphae and stone substrate also causes the formation of bio-films with different colors and chemical compositions. The bio-deterioration of binding material of stone monuments starts by uptake of calcium and then this action leaves the monumental surface eroded and exposed it to water and frost attack. In an investigation it was found that black stain on stone monuments were due to Aspergillus (Burford, E.P. et.al.,2003, Gorbushina, A.A et.al. 1993 & 2004, Gaylarde, C.C.et.al.,2006.)

![Figure 2. World Heritage Site, Fatehpur Sikri: a) Before and b) showing after scientific preservation](image-url)
Material and Methods

Specific morphology and physiology of fungi enables them to colonize multifarious substrates, including cultural heritage artifacts. Due to their pronounced metabolic capacity, fungal deteriogens are able to significantly influence both aesthetic appearance and integrity of monuments, sculptures, murals, paintings, textile and documentary heritage. Now a day, conversance of fungal biology is becoming crucial in proper assessment of contamination and colonization of artworks but also in their adequate storage and protection. Since mycology as a science gains more and more application in the conservation and restoration procedures, the investigations in this scientific filed become essential in cultural heritage safeguard (Grover R et.al., 2007, K., Sharma,2010 and Clair, St. and Seaward, M.2004).

Results and Discussion

Isolation and Identification of Fungi

During the investigation period PDA media was used for the isolation of microorganisms. Samples were collected from the surface of monument. Few drops of sample pour in the petridishes and kept this petridishes at 28±1°C for 7 days for incubation (Grover et.al., 2007). At the end of incubation period fungal colonies were counted, isolated and identified with the help of available literature. The fungal species were Alternaria alternata, Aspergillus fumigatus, Aspergillus flavus, Aspergillus niger, Cladosporium herbarum, and Rhizopus species were observed.

Removal of Dust, Dirt and Micro-Organism

The scientific steps taken by Archaeological Survey of India for the removal of dust and dirt accretion upkeep of stones in neutral pH and preservation and stone strengthening strategies by soft brushing. The moss, fungi and lichen were removed by applying 2-3% solution of ammonia in water and scrubbing with nylon brush. Black patches appeared after removal of thick layer of moss, fungi and lichens which were washed out with the help of dilute solution of oxalic acid in water. A dilute solution of a non ionic detergent with liquid ammonia was applied on treated surface to remove dirt, dust and little amount of acid and ammonia if remained on the surface during the chemical treatment. Lime wash, red, ochre and iron oxide accretion were removed using dilute (10-15%) aqueous acetic acid and oxalic acid solution as per the suitability and afterwards neutralized using aqueous ammonium hydroxide solution. To Arrest further micro vegetation growth 2% aq. solution of sodium pentachlorophenate was applied on clean dried surface (W. H. Dukes, Conservation of Stone, August, 23, 1972). The brittleness and powdering of stones were arrested by the application (brushing and impregnation till saturation) of ethyl silicate based coating material which forms glass like silica gel binder (SiO2aq.) with release of ethanol (evaporates) as by products. Here, noteworthy point is that the intake of stone strengthener materials was comparatively more in the case of damaged and pulverized stones. This can be attributed to the availability of more pores for penetration. The coating of stone strengthener applied on the deteriorated and flaky stone surface by simple brushing and impregnation till saturation.

Selection of Preservative

It is essential that the preservative solution applied on the monuments is should be of good quality. It should be colourless and transparent and must not turn yellow or become coloured with age, but should be fairly stable for long period of time. It should also offer reasonable protection to the monument against moisture and its film should be hard and strong enough to protect the stone surface from injurious accretions. Therefore, for the preservation of Buland Darwaja at Fatehpur Sikri, silane-siloxane based compound (Wacker BS-290) have been chosen (Wacker, 2002), which was diluted with Mineral turpentine oil in the ratio of 1:16 and was applied on the monument by soft paint brush. This compound is water repellent and stops settling of water on stone surface.

Conclusion
To preserve these monuments scientific treatment is very essential. But it is more essential that the identification of problem and selection of chemical should be according the problem of stone surface and preservative solution applied on monuments is of good quality (Agrawal, 1993-94). It should be colorless and transparent and should not turn yellow or become colored with age but should be fairly stable for long parried of time. It should offer reasonable protection to monument and sculpture against moisture and its film should be hard and stone enough to protect the stone surface from injurious accretions.

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