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Fungal spores are ubiquitous; however, the species vary, depending upon the environment and usage. This study investigates the culturable fungi found in the built environment of a cheese cave. Using three different media, viable fungi were captured from a cheese cave in the Eastern United States. Captured fungi were isolated and putative species identified by sequencing the ITS region of rRNA (DNA barcode). In addition, two control fungi were identified by DNA barcode: a commercial strain used for cheese-making and a strain commonly used in the teaching lab where this study was conducted. Due to insufficient sequence variability within the ITS region, the 31 isolated fungal strains plus two control strains were identified to genus or species complex, but could not be resolved to species. Three genera were identified: *Cladosporium* (4 isolates), *Scopulariopsis* (1 isolate), and *Penicillium* (26 isolates, plus both control strains). Most isolates, including the commercial cheese-making strain, belong to a group of *Penicillium* species that cannot be distinguished by ITS sequence (*Penicillium fuscoglaucum*, *Penicillium palitans*, *Penicillium commune*, *Penicillium caseifulvum*). The growth of a representative sample of the cheese-cave fungal isolates was assessed on milk agar, salted milk agar, and malt extract agar. The commercial cheese-making strain exhibits more robust growth on milk-based media (similar to cheese) compared to malt extract agar, correlating with the use in cheese production and indicating characteristic traits of domestication.