Automated image analysis of the host-pathogen interaction between phagocytes and *Aspergillus fumigatus*.

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Projects

Molecular mechanisms of the interaction between *Aspergillus fumigatus* and alveolar macrophages
Details

Abstract

Aspergillus fumigatus is a ubiquitous airborne fungus and opportunistic human pathogen. In immunocompromised hosts, the fungus can cause life-threatening diseases like invasive pulmonary aspergillosis. Since the incidence of fungal systemic infections drastically increased over the last years, it is a major goal to investigate the pathobiology of A. fumigatus and in particular the interactions of A. fumigatus conidia with immune cells. Many of these studies include the activity of immune effector cells, in particular of macrophages, when they are confronted with conidia of A. fumigus wild-type and mutant strains. Here, we report the development of an automated analysis of confocal laser scanning microscopy images from macrophages coincubated with different A. fumigatus strains. At present, microscopy images are often analysed manually, including cell counting and determination of interrelations between cells, which is very time consuming and error-prone. Automation of this process overcomes these disadvantages and standardises the analysis, which is a prerequisite for further systems biological studies including mathematical modeling of the infection process. For this purpose, the cells in our experimental setup were differentially stained and monitored by confocal laser scanning microscopy. To perform the image analysis in an automatic fashion, we developed a ruleset that is generally applicable to phagocytosis assays and in the present case was processed by the software Definiens Developer XD. As a result of a complete image analysis we obtained features such as size, shape, number of cells and cell-cell contacts. The analysis reported here, reveals that different mutants of A. fumigatus have a major influence on the ability of macrophages to adhere and to phagocytose the respective conidia. In particular, we observe that the phagocytosis ratio and the aggregation behaviour of pksP mutant compared to wild-type conidia are both significantly increased.

Identifier

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