Automatic detection of pneumocystis jiroveci in microscopic images: adaeq learning-based approach

Ericd Reyens1,2, Juan Botera-Valencia1, William Grisado-Escobar2, Karen Aranguez2, Indra Beritil1, Tenny Naranjo Preciado1,2
1Department of Electronics and Telecommunications, Instituto Tecnológico Metropolitano, Medellin, Colombia
2Medical and Experimental Mycology Group, Corporación para Investigaciones Biológicas, Medellin, Colombia

Objective: Pneumocystis jiroveci Proventio is one of the diseases that most affect immunocompromised patients today, and under certain circumstances, it can be fatal. One of the most widely used techniques in diagnostic laboratories for the detection of its enzootial agent is optical microscopy. However, some of the disadvantages of this technique are its low sensitivity and low accuracy, and high dependence on an expert to make the diagnosis. Thus, this work aims to develop a computational tool based on a deep learning approach to automatically detect the presence of P. jiroveci Proventio fungus from optical images, and to increase the accuracy of this conventional technique.

Methods: The study involved 29 random patients, from whom respiratory samples (bronchoalveolar lavage, and bronchoscope lavage) were collected. Methanomycin silver staining was then used to prepare the samples. Subsequently, the slides of the analyzed patients were observed using the Leica DM500 microscope using a Leica ICC50 HD camera, and the optical images were taken in at least four random positions on the specimen holder. Thus, an image dataset of 29 different patients was created to detect whether a patient is positive or negative for P. jiroveci Proventio. Finally, a deep learning approach based on convolutional neural networks (CNN) was proposed and evaluated to improve the accuracy of the microscopy technique. The proposed CNN model incorporates global and local features for pixel-wise segmentation.

Results: First, the dataset was processed and segmented using the connected-components methodology. Later, the segmented images were labeled with the help of an expert to train the algorithm. To validate the response of the proposed deep learning approach the obtained results were compared with the obtained conventional image classification techniques like co-occurrence matrix and K NN. The obtained results reveal that the proposed methodology allows to increase the accuracy in the P. jiroveci Proventio identification up to 89%, while the co-occurrence matrix and K NN only achieve accuracies of 88% and 85% respectively.

Conclusion: It is possible to demonstrate that techniques based on digital image processing are a useful tool to support the processes of analysis and diagnosis of samples in medical patients with P. jiroveci Proventio. In addition, the obtained results demonstrate that methods based on deep learning allow us to develop more precise and accurate analysis methodologies for the analysis of patient samples with P. jiroveci Proventio. Our model can be improved by adding new layers, but this would introduce even more hyperparameters that should be adjusted. We intend to extend our model architecture in other areas of medical imaging with the usage of deep learning and computer vision techniques.

The relationship between violence of Candida albicans and environmental oxygen concentration

Masahiro Aba1, Sota Sado1, Takayuki Shitohara2, Aiko Nagamori3, Minoru Nogi3,4, Yoshidego Miyawaki3,4
1Department of Fungal Infections, National Institute of Infectious Diseases, Shinkajuku-ku, Japan
2Antimicrobial Resistance Research Center, National Institute of Infectious Diseases, Shinkajuku-ku, Japan

Objective: Candida albicans is known to colonize human skin and mucous membranes, and cause candidiasis under various immunompressive conditions. Among these Candida species, C. albicans is reported to be the most frequently isolated species, and could colonize on skin, vagina, gastrointestinal tract, and medical devices. Environmental factors including oxygen concentration is thought to affect the capability of colonization and virulence of Candida species. However, previous research was performed under aerobic condition, and few research focused on hypoxic conditions mimicking inside the human body. Therefore, in this study, we performed main experiments utilizing C. albicans under various oxygen conditions to evaluate the effect of environmental oxygen concentration on virulence. Through our study, we aimed to clarify the actual behavior of C. albicans in the human body.

Methods: In this study, fungi CCF434, D4, and D4 were used, and were treated with tetramethyl violet to cause C. albicans disinfection. Mice were divided into 4 groups according to the pre-culture conditions: aerobic, microaerobic (1% oxygen concentration), anaerobic. Under each oxygen condition, C. albicans was grown at room temperature for 2 days. Mice were injected with 105 colony-forming units (CFU) into each mouse at 2.5 × 105 colony-forming units. Post-infection, we performed main experiments utilizing C. albicans under various oxygen conditions to evaluate the effect of environmental oxygen concentration on virulence. Through our study, we aimed to clarify the actual behavior of C. albicans in the human body.

Conclusion: This study, finally CCF434 mice, 7-8 weeks old, were used and infected via laryngeal wash for cause C. albicans disinfection. Mice were divided into 4 groups according to the pretreatment conditions: aerobic, microaerobic (1% oxygen concentration), anaerobic. Under each oxygen condition, C. albicans was grown at room temperature for 2 days. Mice were injected with 105 colony-forming units (CFU) into each mouse at 2.5 × 105 colony-forming units. Post-infection, we performed main experiments utilizing C. albicans under various oxygen conditions to evaluate the effect of environmental oxygen concentration on virulence. Through our study, we aimed to clarify the actual behavior of C. albicans in the human body.
Table 1. Genotype, phagocytosis and proliferation index for 21 strains of Cryptococcus spp.

| Strain | ST* | Genotype | Clonal complex | Phagocytosis | Proliferation (PI)** |
|--------|-----|-----------|----------------|--------------|---------------------|
| A      | 23  | VNI       | 23             | Low          | PI < 1.0            |
| R      | 23  | VNI       | 23             | Intermediate | PI < 1.0            |
| B      | 63  | VNI       | 23             | Low          | PI < 1.0            |
| F      | 63  | VNI       | 23             | Low          | PI < 1.0            |
| J      | 311 | VNI       | 2              | High         | PI > 1.0            |
| E      | 608 | VNI       | 2              | Low          | PI < 1.0            |
| Q      | 612 | VNI       | 2              | Low          | PI < 1.0            |
| N      | 32  | VNI       | 32             | Low          | PI < 1.0            |
| P      | 32  | VNI       | 32             | Low          | PI < 1.0            |
| M      | 607 | VNI       | 32             | Low          | PI < 1.0            |
| H      | 338 | VNI       | 32             | Low          | PI < 1.0            |
| L      | 610 | VNI       | 32             | Intermediate | PI < 1.0            |
| I      | 606 | VNI       | 32             | Intermediate | PI < 1.0            |
| D      | 609 | VNI       | 32             | Intermediate | PI < 1.0            |
| K      | 20  | VGIi      | 3              | Low          | PI < 1.0            |
| T      | 132 | VGIi      | 3              | Low          | PI < 1.0            |
| S      | 157 | VGIi      | 3              | Low          | PI < 1.0            |

*ST: Sequence type, **PI: Proliferation index

Conclusions: Candida albicans was the common species isolated in our study. Candida Chrome Agar can be used as a routine media for rapid identification and Candida speciation. AST needs to be done routinely to know the susceptibility pattern of isolates and initiate proper treatment of patients. Since oral yeast colonization was associated with low CD4+ count (<200 cells/μl), oral lesions can serve as early markers for HIV infection.