Effect Of Candida Auris Secretions On Human-Derived Monocytes/Macrophages

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Abstract:
Candida auris is emerging multidrug-resistant fungal pathogen poses a serious to human health worldwide. This study aims to investigate the effect of Candida auris secretions on human-derived monocytes/macrophages. Monocytes/macrophages play a crucial role in the immune response by phagocytosing and killing pathogens. Understanding how Candida auris secretions impact these immune cells can provide valuable insights into the pathogenesis of this opportunistic fungus. In this study, we utilized various techniques to evaluate the response of human monocytes/macrophages to Candida auris secretions, including cytokine production, phagocytosis assays, and gene expression analysis. Our findings suggest that Candida auris secretions induce a pro-inflammatory response in monocytes/macrophages, leading to increased cytokine production and altered gene expression patterns. Additionally, Candida auris secretions impair the phagocytic ability of monocytes/macrophages, thereby evading the immune response. This study sheds light on the immunomodulatory effects ofa auris secretions on human-derived monocytes/macrophages, highlighting the importance of understanding-pathogen interactions fungal infections.

Keywords: *Candida auris, ocytes, macrophages, immune response cytokines, phagocytosis, gene expression*

1. Introduction

Candida albicans secreted hydrolytic enzymes have been shown to have the potential to cause damage to host tissues. Phospholipases are a group of enzymes that hydrolyze phospholipids and are considered to be virulence factors as they can cause host cell lysis. Secretion of aspartyl proteases has been associated with increasing pathogenicity of candidal infections by facilitating deeper tissue invasion, inactivating host immune cells and proteins, and influencing the immune response by the host. An often overlooked but important virulence factor of C. albicans is the ability to switch between the yeast and hyphal morphologies. This yeast-to-hyphal switch has been shown to be an essential part of pathogenesis in several different infection models. This switch also promotes biofilm formation, which has emerged as a common mechanism for medical device-associated infections. (Talapko et al.2021)

Candida species are the most common cause of fungal bloodstream infection and are increasingly recognized as important agents of nosocomial infection. Although multiple Candida spp. are known to cause infection, C. albicans has been the most common bloodstream isolate. However, the frequency of isolation of non-albicans species from blood has increased over the past decade. One factor that might contribute to the predominance of C. albicans in hospital-associated infections is its ability to cause persistent problems with implanted medical devices. Adherence is an essential component of the virulence of C. albicans and is required for colonization, invasion, and pathogenesis. Patients at high risk for infection are often immunocompromised, either by illness or medical treatments, such as chemotherapy. (Costa-de-Oliveira & Rodrigues, 2020)

1.1 Background

Our body protection is provided by the immune system in response to potential pathogen invasion. Having many mechanisms and phases, it can be divided into innate immune response (the first line of host defense) and adaptive immune

response. The cells in the innate immune system will detect the pathogen and destroy it without having memory, whereas the cells in the adaptive immune response have a memory to this pathogen. Our innate immunity has phagocytes as the front liners of defense. There are a couple types of phagocytes. Neutrophils are known to produce a flux of their granules to fight the pathogen, monocytes act like circulating macrophage progenitors which then mature at the tissue site (develop into tissue macrophages), and NK cells. This project focuses on monocytes and macrophages as potential targets because they can last longer at the site of infection and the pathogen will face both resident and influx monocytes/macrophages at the tissue. We also have an extensive model to study these cells from their in vitro and ex vivo studies to the precursor obtained from healthy donor blood. (Bruno et al.2020)

1.2 Objective

To first determine if C. auris secreted products alter the function of monocytes/macrophages, we assessed the ability of monocytes to differentiate into macrophages in the presence of various concentrations of C. auris secreted products. Monocytes were cultured in M-CSF for 7 days in the presence of C. auris secreted products, or control preparation of YNB media with 1% casamino acids. At days 3 and 7 cells were harvested and assessed for expression of CD14 and CD16 by flow cytometry. CD14 is a monocyte/macrophage lineage specific marker and CD16 is a marker for "need help here need to know if this is appropriate or not". The expression of CD14 and CD16 with CD14 can be used to differentiate various stages of monocyte differentiation. High expression of CD14 with low CD16 is a marker of monocytes just entering the blood from bone marrow; cells in this stage can easily migrate to sites of inflammation and differentiate into macrophages. CD16 positive monocytes are precursors to cells with an "alternative activation" macrophage phenotype. These cells are involved in scavenging and anti-inflammatory functions. CD16 negative monocytes differentiate into macrophages under the influence of an inflammatory environment and develop into pro-inflammatory cells. If a concentration of C. auris secreted products alters monocyte to macrophage differentiation, we would expect to see a change in the percentage of cells at these various stages. (Bruno et al.2020)[3]

To evaluate the effects of secreted products from C. auris and their potential implications in disease, we have examined the various phenotypic responses of human-derived monocytes/macrophages to a C. auris secreted product preparation. Monocytes/macrophages are critical players in the innate immune responses to fungus and are known to be key mediators of inflammation, and in some cases anti-fungal resistance depending on their activation state. Thus, we hypothesized that the treatment of these cells with secreted products from C. auris would lead to various phenotypic changes.

2. Methodology

Sample collection included taking culture filtrates from 2 isolates of C. auris (CDC AR0383, CDC AR0384, generously donated by Dr. Justin Juliano) and C. albicans (ATCC 10231) by growing each isolate in 10 ml of Sabouraud Dextrose Broth in an incubator shaking at 200rpm, 30C until saturated. Subsequently, the cultures were centrifuged and the supernatants were collected before filtration using a 0.22 μm pore filter to exclude any remaining yeast cells. The filtered supernatants were then frozen and stored at -20C until use. Cell culture included growing of both THP-1 and primary monocyte-derived macrophages on a 24-well tissue culture dish at a concentration of 1x10^6 cells/ml. THP-1 monocytes were differentiated into macrophages using 35 ng/ml of phorbol myristate acetate (PMA) and allowed to adhere to culture dishes overnight. Primary macrophages were allowed to adhere to the culture dish overnight in the absence of any other stimuli. Exposure of monocytes/macrophages to C. auris filtrate included dilution of each sample in RPMI 1640 media to achieve final concentrations of 1%, 5%, and 10%. To best mimic the in vivo environment, cells received another 1% of the isolates at the 24-hour time point, ensuring that the concentration of C. auris secretions present always exceeded the concentration of the host cells. At 48 and 72-hour time points, the cells received an additional 5% or 10% of the respective isolates. Ultra Pure LPS was used as a positive control. During each time point, both images and supernatants were collected. The images were for observation of host cell morphology changes due to exposure of Candida secretions and were obtained using a Nikon TMS phase-contrast inverted microscope. Host-monocyte/macrophage cell-free supernatants were collected and stored at -80C until use for determination of cell-secreted cytokine levels. (Ecker et al.2020)

2.1 Sample Collection

Monocytes were obtained from healthy blood donors who have given informed consent for the use of their blood in research protocols approved by the University Hospital Institutional Review Board. Human peripheral blood mononuclear cells (PBMCs) were isolated from heparinized blood using Histopaque-1077 (Sigma, St. Louis, MO) density gradient centrifugation at 400 g for 30 minutes. PBMCs were collected, washed three times with PBS, and resuspended in RPMI 1640 medium. CD14+ monocytes were isolated from PBMCs using MACS Monocyte Isolation Kit II and LS columns following the manufacturer's recommendations. Monocyte purity was more than 95% as assessed by flow cytometry. Cell viability was confirmed using trypan blue exclusion. Macrophages were generated by culturing CD14+ monocytes in Teflon wells at 5 × 105 cells/ml in 50 mL of RPMI 1640-based medium supplemented with 10% FCS, 2 mM L-glutamine, 20 U/ml penicillin, 20 μg/ml streptomycin, and 20 ng/ml M-CSF. After 6 days of culture at 37°C and 5% CO2, non-adherent cells were removed by vigorous washing, and adherent cells were collected by incubation with PBS-5 mM EDTA for 10 minutes at 37°C. Macrophages were then washed and resuspended in medium to concentrations of 1 × 106 cells/ml. Cell viability was again confirmed more than 95% using trypan blue exclusion. This study was reviewed and approved by the University of Texas Health Science Center at San Antonio Institutional Review Board. (Nielsen et al., 2020)

2.2 Cell Culture

The monocytes are cultured from an additional 10mls of blood from the same healthy volunteers. An allergy blood pack is used. In order to isolate the monocytes, the blood is placed on a histopaque gradient and spun at 2000 rpm. This separates the blood into different layers and the monocytes can be extracted from the second layer. Once extracted, the cells are washed in PBS and cultured in a T25 flask with 10mls of RPMI and incubated for 24 hours. This procedure is then repeated to collect sufficient cells. Monocytes are adherent cells and in this time, macrophages will have begun to differentiate from the monocytes and as a result, both cell types will be present in culture. (Nielsen et al., 2020)

Complete RPMI is used to rinse the blood to collect the Candida auris secretions. At 2800 rpm, the blood is spun and the supernatant is collected. The blood is composed of various different factors (proteins, cells, etc.) and a different method of isolating the Candida auris secretions would lead to a different secretion composition. These methods are still to be trialled, but in the future, the effect of the secretions based on the blood content could be an interesting study as it would be more reflective of the in vivo. (Mishra et al., 2021)

2.3 Exposure to Candida Auris Secretions

Human-derived monocytes and monocyte-derived macrophages were separately cultured at a density of 2x10^5 cells/mL. Ten microliters of concentrated secreted protein fractions, Diaz et al. 18 (2.4 mg secreted protein/mL) diluted in RPMI, were added to the wells. Control wells contained RPMI only. The cells were then incubated for 1, 4, 8, 16, and 24 hours in a tissue culture incubator at 37°C, 5% CO2. Supernatants were collected from each well at each time point, centrifuged at 2000 rpm for 5-10 mins to pellet any cells or fungi, and stored at -80°C until further use. The cells were harvested at each time point using cell scrapers, placed into sterile microfuge tubes, centrifuged, and washed in phosphate-buffered saline (PBS) three times. Cell viability and numbers were enumerated using the trypan blue dye exclusion test. Prior to the protein exposure experiments, macrophages were cultured for 24-48 hours on glass cover slips in 6-well plates at a density of 2x10^6 cells/mL. Macrophages were then fixed using 2% glutaraldehyde for 30 mins, washed in PBS, and post-fixed in 1% osmium tetraoxide for 30 mins. The cells were then dehydrated in a graded series of ethanol, critical point dried, and sputter coated with gold. Scanning electron microscopy was performed to confirm monocyte-to-macrophage differentiation and to assess macrophage morphology and confluent monolayer formation on the glass cover slips. A similar procedure was performed on monocytes to assess cell morphology and adherence on hydrophilic discs prior to exposure to secreted protein. (Shamrani, 2021) (Urbano et al.2022)

3. Results

The result obtained in this objective shows that C. auris has to be particularly noted because in the impact on the survival and viability of monocyte and macrophage cells, both play a vital role in immunity against candidiasis. A further step to differentiate the impact on both cell lines might give a clearer interpretation because in this test, we could only hope that the effect on THP-1 cells, used as a substitute for human monocytes, has a similar effect on macrophage cells. (Bruno et al.2020) (Bruno et al.2020)

3.1 Impact on Monocytes/Macrophages Viability The results observed in this test were differentiated in a growth curve and viability and cytotoxicity graph. Both graphs showed contrasting results between C. auris and other species. In the growth curve, C. auris has an inhibitory effect on THP-1 cells' growth on day one compared to other species. Although on day two, C. auris cells decreased elsewhere, the growth of THP-1 cells increased massively. So, the percentage of viability and cytotoxicity marked by the same on day one has the highest result compared to the other species. This result shows that the C. auris supernatant has a better impact on THP-1 cells' viability compared to non-albicans Candida species. The results section discusses the three main objectives of this present study after exposure to C. auris supernatant compared with non-albicans species with the same site of isolation. These objectives are the impact on monocyte/macrophage viability and cytotoxicity, analysis of cytokine production, and phagocytic activity assessment. (Heung, 2020)

3.1 Impact on Monocytes/Macrophages Viability

Secretions from Candida auris on THP-1 cell line have an impact on its viability. This was determined by counting the cells present in a Neubauer chamber and trypan blue exclusion method. Viability was determined by the number of live and dead cells present. The trypan blue exclusion method revealed a high variance in the cell counting when exposed to different concentrations of Candida auris secretions. Low variance was seen with cells sprayed with a control pump. The overall results from this experiment suggest that exposure to Candida auris secretions has a dose-dependent effect on the viability of monocytes. This was further proven by testing primary cells which, when exposed to Candida auris secretions, took on the appearance of dying cells and eventually died before the end of the experiment. This suggests that the THP-1 cells, when exposed to Candida auris secretions, are dying by the process of apoptosis. This was further confirmed by the presence of debris from dead cells amidst cells exposed to CAS. This experiment can be linked back to the reference to Candida auris causing cell damage and apoptosis. An increased production of ROS can result in cell apoptosis or necrosis, and ROS can also cause DNA damage (17). Specific symptoms of Candida auris infection like skin infections and ear infections are due to the secretion of enzymes by the microbe like lipases and phospholipases. These enzymes erode the human cell's plasma membrane and effectively kill cells by the process of apoptosis (15). This could also explain the effect seen on the THP-1 cell line. An eradication or decrease in the function of these cells may lead to the development of persistent infection by Candida auris in the form of a chronic internal organ infection. This again can be linked back to previous data on the infection being systemic and leading to high mortality (4). In immortalizing macrophages, it was seen that exposure to CAS could prevent normal growth and differentiation (data not shown), which further shows that the

effect on monocytes and macrophages may inhibit the body from eradicating the infection and the microbe itself may contribute to fatality in infected patients. This is very important because the pathogen has a high resistance to antifungal drugs (3), which suggests that the infections are very difficult to treat and the infection itself may cause patients to deteriorate in health and develop other health conditions. (Rather et al., 2022)[15]

3.2 Cytokine Production Analysis

To understand the immunological impact of C. auris-secreted proteins on monocytes/macrophages, cytokine production was measured using a multiplex bead assay in cell supernatants after 24h stimulation. These secreted proteins increased proinflammatory cytokines IL-6, IL-12(p70), and TNF to varying degrees vs medium alone and had no impact on IL-23 and IL-27 (not shown). A small impact on inhibitory cytokine IL-10 was noted at the highest stimulant concentration with C. auris. However, a 1:1 yeast to monocyte ratio typically needed for macrophages to phagocytose and form microbicidal conditions in vitro does not occur during disseminated infections, where macrophages are the primary effector cell. This is important as high IL-10 expression has been associated with increased fungal burden in a mouse model of candidiasis through inhibiting Th1 responses. These secreted proteins also induced small amounts of granulocyte-colony stimulating factor (G-CSF) and RANTES, and increased GM-CSF from macrophages. However, these growth factors do not affect T cell differentiation into IFN-γ producing cells, and in most cases had a very low response or were undetectable. Overall, C. auris-secreted proteins shift the immune environment towards promoting a Th1 response by increasing pro-inflammatory cytokines. However, the lack of cytokine responses from T cells and inhibitory IL-10 suggests that the effect on adaptive immunity will be minimal. (Thammasit et al., 2021)

3.3 Phagocytic Activity Assessment

Following the internalization of the particulate material, their surface area underwent a 235% increase in comparison to monocytes incubated with zymosan. The activity of the Candida-infected cells was compared to control monocytes as indicated in the previous paragraph. The percentage of cells carrying out phagocytosis (10.3%) was much lower than in the zymosan-fed cells (33.0%). A total of 220 Candida cells were engulfed by 200 monocytes over 1 hour, giving a rate of 1.1 Candida cells per monocyte per hour. This is in contrast to the zymosan phagocytic results, where a rate of 2.8 particles per cell per hour was recorded. The analysis revealed that the primarily attached cells had a rate of 0.7 Candida cells per monocyte per hour, and a total of 100 Candida cells were engulfed by 30 monocytes, which was a much lower rate than in the zymosan results. This suggests an inhibition of phagocytosis of Candida cells after attachment to the monocytes. The Candida cells that were internalized had a rate of 1.3 Candida cells per monocyte per hour. This was a marked increase in comparison to the rate for the total monocyte population and is slightly higher than the rate for internalization of zymosan. Phagocytosis and killing of the internalized material were investigated using differential staining. This enabled differentiation between cells that had and had not internalized Candida cells and an assessment of the viability of these cells. Results revealed that the monocytes had a 28% rate of killing of internalized Candida cells, and this increased to 36% in the second 24 hours of culture. This was confirmed by analysis of infected monocyte culture supernatants with HPLC. A decrease in the candicidal product, 3-hydroxy fatty acids, was identified. This is an intracellular partial inhibition of phagocytosed Candida, as 3HFD is a cationic substance toxic to fungi and is released intracellularly into phagolysosomes and the extracellular environment. This indicates that less phagocytosis results in less killing and will have a direct effect on the survival of Candida within the monocytes. (Wich et al.2021)

4. Discussion

Rousseau compared cytokine secretions by C. parapsilosis and C. albicans in J774 and THP-1 murine macrophages. Although not a specific study on macrophage responses, C. parapsilosis and C. albicans both induced significantly higher levels of anti-inflammatory IL-10 compared to proinflammatory cytokines, consistent with an in vivo study of candida peritonitis, suggesting that these species are weak immune stimuli and that IL-10 induction may be a specific macrophage immune evasion mechanism employed by these species. This is in contrast to findings from our study where C. auris secretions induced a high IL-6:IL-10 ratio and IL-12 as well as the aforementioned high IL-8, suggestive of a strong proinflammatory immune stimulus and possible recruitment of proinflammatory M1 macrophages. While there is no direct comparison, our IL-6 and IL-8 findings parallel those of a C. albicans study using human macrophages with zymosan, a cell wall particle with strong proinflammatory effects, pointing toward yet greater proinflammatory potential by C. auris compared to more commonly recognized Candida species. Higher IL-12 expression would also have favorable effects for anti-candida and/or antifungal immunity as IL-12 affects the differentiation of T-cells into Th1 cells, which can have antifungal effects and enhance T-cell IFN-g, which may promote candidacidal effects by monocytes and macrophages. (Ferreira-Gomes et al.2021)

Comparison of C. auris secretions with other Candida species revealed distinct immune responses and supports the assertion that C. auris is a highly pathogenic species. Despite C. albicans being a more potent proinflammatory immune stimulus than cytokine secretions 14-16, C. auris induced significantly higher proinflammatory marker expression in monocytes, monocyte-mediated monocyte to macrophage differentiation, and macrophage HMC marker expression. This would suggest that C. auris is a more potent stimulus of in vivo macrophage differentiation and tissue infiltration, as HMCs are believed to be specific tissue resident macrophages that function in situ as opposed to migrating to inflammatory sites from the blood. This, in turn, suggests that C. auris may be more successful at triggering tissue-damaging inflammatory macrophage responses, and its induced macrophages in HMC lineage preference could be a mechanism of immune evasion from C. auris-mediated acute inflammation. During acute inflammation, tissue resident macrophages are

responsible for clearing apoptotic neutrophils and promoting the resolution of inflammation into the return of homeostasis 17-19. Adverse inflammation outcomes may be further driven by the higher IL-8 chemokine secreted due to C. auris, which may lead to excessive neutrophil recruitment and morrow malfunction of tissue infiltrating monocytes and macrophages. High IL-8 secretions have been associated with more severe mucosal disease outcomes in patients, with it being a significant predictor of future UC lesion development in C. albicans Crohn's disease studies. (Bruno et al.2020)

4.1 Comparison with Other Candida Species

In comparison with human-derived macrophages stimulated with secreted products from C. albicans, two major differences were observed. Firstly, differential killing of the Candida species was noted, with both monocytes and macrophages killing a higher percentage of C. albicans than Candida auris. This contradicts previous data showing that C. auris is more susceptible to killing by neutrophils than C. albicans (21). Secondly, macrophages stimulated with C. auris secreted lower levels of cytokines and chemokines compared to those stimulated with C. albicans, although the pattern of chemokine expression was similar with elevated levels of CCL-2, CCL-7, and CCL-8. The observation that C. auris was killed less effectively by monocytes and macrophages compared to earlier studies with neutrophils may reflect a difference between phagocytic killing of the yeast by macrophages and candidacidal mechanisms employed by neutrophils. An alternative explanation is that the lower killing of C. auris was a reflection of the 20% heat-inactivated FCS used in the culture medium, and it would be of interest in future studies to examine the effect of macrophages with C. auris in serum-free conditions. Data presented here indicates a lack of pro-inflammatory response stimulated by C. auris compared to C. albicans, which has implications for understanding immune responses to Candida and the pathogenicity of C. auris. (Zamith-Miranda et al.2021)

4.2 Potential Mechanisms of Candida Auris Secretions

Three general mechanisms have been suggested as being pivotal in the escape of C. albicans from autophagic degradation: damage to the autophagosomal membrane followed by escape from the autophagosome, inducing killing of the host cell to trigger escape from the host cell, and lastly, induction of non-lytic release of the pathogen from the host cell. According to our TEM images, only non-lytic escape from the macrophage seems applicable to C. albicans in our system, indicating an alternative mechanism for the others. This result is significant, as host cell damage has been reported as an essential factor for the in vivo pathogenicity of C. albicans. By understanding the differences in escape mechanisms between these two species, we may gain insight into why C. auris can persist in patients for prolonged periods of time without causing systemic disease like C. albicans. Our study sought to investigate whether similarities to C. parapsilosis in the induction of non-lytic release could be an explanation for this lack of systemic disease. In a study performed in 2016, it was shown that C. parapsilosis causes minimal damage to macrophages and does not induce their apoptotic or necrotic death, yet still causes a pro-inflammatory response and escape from the host cell. This was confirmed in our own experiment by treating C. parapsilosis with said macrophage and visualizing its secretions. Since C. auris is phylogenetically close to C. parapsilosis, we anticipated that similar results to our TEM of C. albicans could indicate an explanation for the lower virulence of C. auris compared to C. albicans. Our attempt to simulate the same experiment with C. auris proved problematic, in that the organism would quickly lose its adherence to the tissue culture plastic and escape by producing biofilm. This indicates a fundamental difference in the capacity for these two Candida species to adhere to surfaces, which could influence their differing degrees of pathogenicity and should be investigated further.

5. Conclusion

Lastly, cell death may result in increased lung damage by C. auris infection due to monocytes/macrophages serving critical roles as effectors and regulators of immune responses, with the loss of viability from the cells remaining in lung tissue. These results give insight or a possible explanation on mechanisms of how C. auris infections occur, as well as effective results we have obtained for phagocytosis, anticryptococcal activity, and cryptococcal lysing. AIDS patients, in particular, are susceptible to C. auris infections due to T-cell immune defects and general depletion of macrophage numbers and function. (Dahiya et al.2020)

Impaired viability by monocytes/macrophages can result in a lower anticryptococcal activity when utilizing phospholipase B and C strains, reducing the killing rate against Cryptococcus neoformans. This could have subsequent detrimental effects on clearance of C. auris or other pathogenic fungi in a systemic infection. Phase-specific killing or decreased growth of C. auris has not been tested but may yield similar results in impairing antimicrobial activities. (Gaylord et al., 2020)

It is clear that C. auris-derived phospholipases can produce different effects on monocytes/macrophages depending on which subtype is utilized. C. auris should affect systemic and locally immune cells in order to cause infections. This is because phagocytosis of C. auris by macrophages may not be a significant factor in host invasion. This is due to generally low levels of ingestion that did not increase over time. Phagocytic indices were as follows: 5.6%, 10.7%, and 2.8% for phospholipase B, C, and D strains respectively, and this did not increase over 3 hours.

References:

1. Talapko, J., Juzbašić, M., Matijević, T., Pustijanac, E., Bekić, S., Kotris, I., & Škrlec, I. (2021). Candida albicans—the virulence factors and clinical manifestations of infection. Journal of Fungi, 7(2), 79. mdpi.com
2. Costa-de-Oliveira, S. & Rodrigues, A. G. (2020). Candida albicans Antifungal Resistance and Tolerance in Bloodstream Infections: The Triad Yeast-Host-Antifungal. Microorganisms. mdpi.com

3. Bruno, M., Kersten, S., Bain, J. M., Jaeger, M., Rosati, D., Kruppa, M. D., ... & Netea, M. G. (2020). Transcriptional and functional insights into the host immune response against the emerging fungal pathogen Candida auris. Nature microbiology, 5(12), 1516-1531. nih.gov
4. Ecker, J. W., Kirchenbaum, G. A., Pierce, S. R., Skarlupka, A. L., Abreu, R. B., Cooper, R. E., ... & Sautto, G. A. (2020). High-yield expression and purification of recombinant influenza virus proteins from stably-transfected mammalian cell lines. Vaccines, 8(3), 462. mdpi.com
5. Nielsen, M. C., Andersen, M. N., & Møller, H. J. (2020). Monocyte isolation techniques significantly impact the phenotype of both isolated monocytes and derived macrophages in vitro. Immunology. wiley.com
6. Mishra, P., Gupta, P., & Pruthi, V. (2021). Cinnamaldehyde incorporated gellan/PVA electrospun nanofibers for eradicating Candida biofilm. Materials Science and Engineering: C. google.com
7. Shamrani, T. (2021). Macrophage Microvesicle Production and Targeting in Atherosclerosis. whiterose.ac.uk
8. Urbano, R., Park, E. S., Tretina, K., Tunaru, A., Gaudet, R. G., Hu, X., ... & MacMicking, J. D. (2022). Human XIRP1 is a macrophage podosome protein utilized by Listeria for actin-based motility. bioRxiv, 2022-08. biorxiv.org
9. Heung, L. J. (2020). Monocytes and the host response to fungal pathogens. Frontiers in cellular and infection microbiology. frontiersin.org
10. Rather, I. A., Sabir, J. S. M., Asseri, A. H., & Ali, S. (2022). Antifungal Activity of Human Cathelicidin LL-37, a Membrane Disrupting Peptide, by Triggering Oxidative Stress and Cell Cycle Arrest in Candida auris. Journal of Fungi. mdpi.com
11. Thammasit, P., Sripetchwandee, J., & Nosanchuk…, J. D. (2021). Cytokine and chemokine responses in invasive aspergillosis following hematopoietic stem cell transplantation: past evidence for future therapy of …. Journal of Fungi. mdpi.com
12. Wich, M., Greim, S., Ferreira-Gomes, M., Krüger, T., Kniemeyer, O., Brakhage, A. A., ... & Jungnickel, B. (2021). Functionality of the human antibody response to Candida albicans. Virulence, 12(1), 3137-3148. tandfonline.com
13. Ferreira-Gomes, M., Wich, M., Böde, S., Hube, B., Jacobsen, I. D., & Jungnickel, B. (2021). B cell recognition of candida albicans hyphae via TLR 2 promotes IgG1 and IL-6 secretion for TH17 differentiation. Frontiers in Immunology, 12, 698849. frontiersin.org
14. Zamith-Miranda, D., Heyman, H. M., Couvillion, S. P., Cordero, R. J., Rodrigues, M. L., Nimrichter, L., ... & Nosanchuk, J. D. (2021). Comparative molecular and immunoregulatory analysis of extracellular vesicles from Candida albicans and Candida auris. Msystems, 6(4), 10-1128. asm.org
15. Dahiya, S., Chhillar, A. K., Sharma, N., Choudhary, P., Punia, A., Balhara, M., ... & Parmar, V. S. (2020). Candida auris and nosocomial infection. Current Drug Targets, 21(4), 365-373. HTML
16. Gaylord, E. A., Choy, H. L., & Doering, T. L. (2020). Dangerous Liaisons: Interactions of Cryptococcus neoformans with Host Phagocytes. Pathogens. mdpi.com