

## LB1732

### Particulate matter modulates inflammation cytokine production and lipid formation in *Cutibacterium acnes*-stimulated SZ95 cell

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The skin is one of the main organs exposed to airborne particulate matter (PM). Recent some studies have demonstrated that PM is associated with inflammatory response and various skin diseases. Acne is a chronic inflammatory disease of the pilosebaceous unit caused by increased sebum production. Little is known concerning the potential effects of PMs on acne inflammation. Therefore, we conducted experiments to analyze the effects of PMs on *C. acnes* treated SZ95 cells. To investigate the effect of PM on *C. acnes*-treated SZ95, we injected Heat-killed *C. acnes* in SZ95 cell to induce acne-like status. PM, which mainly comprised polycyclic aromatic hydrocarbons was used as reference PMs. After SZ95 cells was treated with PM. Cell viability was measured by CCK-8 assay. The gene expression was investigated by real-time PCR and protein expression was assessed by Western blot. Intracellular lipid droplet formation was investigated by Nile Red O staining. Results showed that PM-induced pro-inflammatory cytokine and lipid synthesis factor mRNA expression and protein secretion in *C. acnes*-treated SZ95. In addition, when *C. acnes* stimulated SZ95 was treated with PM, lipid synthesis droplets were increased. In conclusion, the results of this study indicate that PM induces lipid synthesis and inflammatory response in *C. acnes*-stimulated SZ95 *in vitro*. Further experiments are needed to determine which signal transduction pathways are mediated by the activation of these reactions in subsequent experiments.



## LB1733

### Skin microbiota disruption is associated with disease severity in alopecia areata

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Background: The skin microbiome is a complex microbiological system with multiple interactions that can lead to both local and distant changes in homeostasis. Objectives: To determine the relationship between skin microbiota and severity in alopecia areata patients. Methods: We interrogated 16S rRNA gene on 20 naïve patients with severe (n=8) or mild (n=12) AA and healthy controls (n=4) using Swab samples collected from lesional scalp areas and analyzed on an Ion Torrent S5 sequencer. Disease status was classified as mild/moderate (< 50% involvement and <1yr duration) and severe (≥50% SALT, AT or AU). Linear discriminant analysis (LDA) effect size (LefSe) scores were used to identify the taxa that best discriminated between groups. Results: Alpha diversity did not differ among samples. However, beta-diversity (Jaccard) was different in mild (p=0.002) and severe AA (p=0.005) vs controls. Our results indicated that taxa of healthy scalp was dominated by four bacterial phyla: *Actinobacteriota* 56.1%, *Firmicutes* 27.2%, *Bacteroidota* 8.5%, and *Proteobacteria* 7%, representing a total of 18 different groups. There were apparent shifts in dominance associated with AA severity from *Actinobacteriota* (56.1% control; 48.4% mild; 37.9% severe) to *Proteobacteria* (7% control; 6.1% mild; 17.4% severe) groups. Species which showed LDA>2 between control and AA were *Brevibacillus*, *Anaeroplasm*, *Jeotgalicoccus*, *Asteroleplasma*, *Helicobacter* (all p<0.05). Conclusions: Amount of scalp microbiota does not vary between healthy subjects and patients with AA. However, there are changes in relative bacteria population with a positive correlation between proinflammatory (*Proteobacteria*)/anti-inflammatory (*Actinobacteriota*)/bacteria ratio and disease severity. Whether these changes are either the cause or consequence of AA needs to be explored further.



## LB1734

### GNPMB suppresses the antimicrobial activity of macrophages through targeting VAMP8

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Leprosy, caused by the intracellular microbe *Mycobacterium leprae*, provides a perfect model to study the mechanism of host immune response against intracellular bacterial pathogen. As a key part of innate immunity and the major host of leprosy pathogens, macrophage dysfunction is an important reason for *M. leprae* accumulation in lepromatous leprosy, while the mechanism remains unclear. In this study, we performed single cell RNA sequencing on leprosy biopsy specimens, and analyzed the gene expression profiles in macrophages. We found GNPMB, a kind of type I glycoprotein, was overexpressed in lepromatous leprosy, compared to the self-limited tuberculoid leprosy. Deficiency of MB21D2 in THP-1 cells showed less proinflammatory cytokine production upon *M. leprae* infection, such as IL-6, TNF- $\alpha$  and IL-1 $\beta$ . Consistently, we found addition of GNPMB led to significantly deficiency of antimicrobial activity in macrophages, which indicated that GNPMB plays a key role in promoting *M. leprae* to escape host immune response. Mechanistically, we demonstrated that GNPMB inhibits cellular autophagic flux by blocking the maturation of autolysosome. Upon IL-4 stimulation, the lysosome-localized GNPMB directly interacts with the lysosomal SNARE protein VAMP8, blocking the assembly of STX17-SNAP29-VAMP8 SNARE complex, which mediates the fusion of autophagosome with lysosome. Thus, our finding unveils new mechanisms by which *M. leprae* escapes innate host defense in the progressive form of leprosy and provides insights for developing new strategies to treat intracellular microbes.



## LB1735

### Utilizing large-scale sequencing to depict changes in immune cells during cancer immunoediting in epidermal neoplasms

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Cancer immunoediting is a dynamic process whereby the immune system can both constrain and promote tumor development, which proceeds through three phases termed elimination, equilibrium, and escape. Although the cancer immunoediting process has set the foundation for understanding the immune system's dual roles on cancer cells and establishing the basis for revolutionary cancer immunotherapies, very few studies have captured cancer immunoediting in its early stages due to lack of proper mouse models. To address this deficiency, we developed a novel mouse model for fluorescent tracing of somatic, epithelial cell transformation to study the role of immunoediting in skin cancer development. For the first time, our model provides the direct visualization of epithelial cancer development *in vivo* and allows the observation of all three phases of cancer immunoediting. In addition, our model also allows tumor-infiltrating immune cells to be traced during this process. This model allowed us to utilize large-scale sequencing technologies and spatial transcriptomics to measure genome-wide gene expression changes in tumor cells and immune cells in the transition from equilibrium to escape phases. Our preliminary data shows that the transition from equilibrium lesions to escape tumors involves the upregulation of TGF $\beta$  signaling in escape tumors. We also observe changes in the immune cell composition in the equilibrium phase and escape phase. In the equilibrium phase, CD4+ and CD8+ T cells, NK cells, macrophages, Tregs, and Gamma delta T cells are observed. In the escape phase, CD8+ T cells, NK cells, Tregs, and macrophages expanded, while CD4+ T cells, dendritic cells, and Gamma delta T cells disappeared. Altogether, our work provides novel insights into the mechanisms driving phase transition during cancer immunoediting and changes in the immune cell composition during epithelial cancer progression.



## LB1736

### Changes and their significance of Th1, Th2, Th17 cells and SCCA levels in peripheral blood of elderly patients with atopic dermatitis

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Objective: To investigate the changes and significance of Th1, Th2, Th17 cells and squamous cell carcinoma antigen(SCCA) levels in peripheral blood of elderly patients with atopic dermatitis(AD). Methods: Twenty-two elderly AD patients (elderly AD group) and 16 healthy subjects(healthy control group) were enrolled in the Department of Dermatology, Sichuan Provincial People's Hospital from March 2022 to July 2022. Clinical data were collected from elderly AD patients, as well as laboratory parameters including peripheral blood EOS counts, total IgE values, and SCCA levels. Flow cytometry was used to detect the proportion of Th1, Th2 and Th17 cells in peripheral blood CD4+T cells in the two groups. Visual characteristics were compared between groups using the Mann-Whitney U test or one-way analysis of variance, and correlation analysis was performed using Spearman correlation analysis. Results: Elderly AD patients were predominantly male, with severe and widely distributed skin lesions, lichenoid eczema mainly on the extensor sides of the extremities and back, and some patients had one or more systemic diseases. The peripheral blood EOS count in the elderly AD group was significantly higher than that in the healthy control group ( $P<0.05$ ). The proportion of Th17 cells in the peripheral blood of the elderly AD group was significantly higher than that of the healthy control group ( $P<0.05$ ), and although the proportion of Th1 and Th2 cells was slightly increased compared with the healthy control group, the difference was not statistically significant. Peripheral blood EOS counts were negatively correlated with the proportion of Th2 cells ( $r=-0.441$ ,  $P<0.05$ ) and positively correlated with serum SCCA levels ( $r=0.612$ ,  $P<0.01$ ) in elderly AD patients. Serum SCCA levels were positively correlated with SCORAD scores in elderly AD patients ( $r=0.607$ ,  $P<0.01$ ). Conclusion: Elderly AD patients have unique clinical features, mainly Th17 type inflammation. Serum SCCA levels in elderly AD patients may reflect disease severity.



## LB1737

### Antivirulence therapy-a new paradigm in treating staphylococcal skin infections

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The human skin microbiota is comprising of mostly beneficial microbes with a few pathogens or opportunistic pathogens. Some pathogenic bacteria may cause chronic skin inflammation and induce undesirable skin conditions, from abscesses, cellulitis, to possible tumorigenesis. Antibiotics have been used to treat bacterial infections by aiming at eradicating all bacteria, bad and good altogether. But the emergence and dissemination of multidrug-resistant bacteria has become a global healthcare issue as a result. Using a well-studied community acquire methicillin-resistant *Staphylococcus aureus*(CA-MRSA) strain USA300, antibiotic-induced pathogenicity was assessed *in vitro* and *in vivo*. We have illustrated that  $\beta$ -lactam antibiotics and tetracyclines induced the expression of multiple surface-associated virulence factors as well as the secretion of toxins in *in vitro* studies, and in *in vivo* studies mice infected with MRSA and treated with ampicillin or tetracycline showed enhanced bacterial pathogenicity. Non-antibiotic antivirulence therapy suppressing the productions of virulence factors of *S. aureus* seems to be a viable alternative therapeutic option. Methods: We constructed using USA300 as the parental strain a series of deletants of know virulence regulators such as *agrA*, *saeQRS*, *saeR*, *saeS*, and *sigB*, to assess their pathogenicity in mice skin abscess model. The CFU on abscesses were counted and compared. We further used a potent broad-spectrum antivirulence compound Antenovus-J01 in a mice skin model to illustrate the efficacy of applying antivirulence agents in treating CA-MRSA infected wounds. Results: Deletion of major virulence regulators of USA300 rendered this strain of CA-MRSA lost its capability to form abscesses in mice skin and antivirulence compound Antenovus-J01 effectively reduced virulence expression in mice and helped wound healing. Conclusion: Our findings asserts that antibiotic-based therapy of staphylococcal infections may present risks for induced pathogenicity and antivirulence therapy is an effective alternative therapeutic option for staphylococcal skin infections.

