Activity of sarecycline in murine models of infection and inflammation
C Brunck1, J Del Rosso2, S Tyting3, M Draper4, JL Johnson5 and A Cracle1, 1 Dermatology, Yale University, New Haven, Connecticut, United States, 2 JDR Dermatology Research Foundation, Las Vegas, Nevada, United States, 3 The University of Texas Health Science Center at Houston School of Public Health, Houston, Texas, United States, 4 Paratek Pharmaceuticals, Inc, Boston, Massachusetts, United States, 5 Pathology and Dermatology, Northwestern University Feinberg School of Medicine, Chicago, Illinois, United States and 6 Boston University School of Medicine, Boston, Massachusetts, United States
Sarecycline (SAR) is a tetracycline-class antibiotic FDA-approved for treatment of moderate-to-severe acne. In vitro studies demonstrated a narrow-spectrum antibacterial activity, targeting clinically relevant Gram-positive bacteria with reduced activity against Gram-negative bacteria commonly found in the human gastrointestinal tract. A murine systemic (intraperitoneal) infection model was used to assess the in vivo efficacies of SAR, doxycycline (DOX), and minocycline (MIN) against S. aureus and E. coli. At 48 hours after systemic infection with S. aureus, SAR, DOX, and MIN had a protective dose to achieve 50% survival (ED50) of 0.25, 0.3, and 0.03 mg/kg, respectively. In contrast, SAR did not demonstrate in vivo efficacy against E. coli at the tested doses of >40 mg/kg, while DOX and MIN had an ED50 of 5.72 and 6.95 mg/kg, respectively. A murine neutropenic thigh wound infection model was used to model tissue-based infection to assess efficacies of SAR and DOX against S. aureus. At 24 hours post infection, SAR achieved a 2.0 log10 reduction of thigh bacterial burden comparable to DOX, with 50% effective dose (ED50) values of 8.23 and 8.32 mg/kg, respectively. The anti-inflammatory effects of SAR, DOX, or MIN, were tested in male, Sprague-Dawley rats using a cartagenen-induced footpad edema model. Mean percent infiltration at a dose of 100 mg/kg was 53.1, 36.0, and 20.5, respectively. SAR demonstrated in vivo efficacy against S. aureus but not E. coli in animals model of infection, confirming the narrow-spectrum activity observed in vitro. SAR also showed an anti-inflammatory effect comparable to DOX and MIN.

A basophil-neutrophil axis promotes itch
F Wang1, AM Trier1, F Li1, S Kim1, Z Chen1, J Chai2, M Mack3, S Morrison2, JB Hamilton1, J Baek1, TB Yang1, A M Ver Heul1, AZ Xu2, Z Xie1, X Dong4, M Kubo5, HH Wu2, C Hsieh2, X Dong4, Q Liu2, D Margolis6, M Ardeleanu3, MJ Miller2 and BS Kim1, 1 Dermatology, Washington University in St Louis, Missouri, United States, 2 Washington University in St Louis, Missouri, United States, 3 Regeneron Pharmaceuticals Inc, Tarrytown, New York, United States, 4 Johns Hopkins University, Baltimore, Maryland, United States, 5 Rikagaku Kenkyusho Yokohama Campus, Yokohama, Kanagawa, Japan and 6 University of Pennsylvania Perelman School of Medicine, Philadelphia, Pennsylvania, United States
Atopic dermatitis (AD) is a chronic and relapsing inflammatory skin disorder associated with severe, chronic itch. In addition to chronic itch, patients with AD often suffer flares of acute itch. Although recent seminal discoveries have unearthed the neuroimmune circuitry of itch, mechanistic understanding of the acute phase remains limited. Notably, we previously demonstrated that a large proportion of patients with AD harbor allergen-specific IgE and exhibit a propensity for acute itch flares, provoking the hypothesis that acute itch flares in AD may be driven by allergen-specific IgE. To definitively address this, we developed novel murine models of IgE-mediated itch and found that, although in the steady-state, IgE indeed stimulates acute, mast cell-dependent, histaminergic itch, in the context of AD-like disease, both mast cells and histamine become entirely dispensable for acute itch flares that happen on top of chronic itch. Rather, we strikingly found that this form of itch was critically dependent on the presence of basophils. Inflammatory basophils were prone to release cellular contents and intravital imaging revealed basophil-sensory neuron interactions within the skin that was provoked by cutaneous allergen exposure. Functionally, allergen-stimulated basophils exhibited enhanced production of leukotriene C4 (LTC4). The presence of CysLT2 on neurons, a receptor for LTC4, was critically required for acute itch flares in AD-associated inflammation. Collectively, our study demonstrates a previously unrecognized form of acute itch flare that emerges in the context of chronic skin inflammation to activate a non-canonical basophil-neutrophil circuit.

Heightened levels of microvesicle particles resulting from combination of ethanol and thermal burn injury
C Brower, AA Awoyemi, C Rapp, CE Borchers and JB Travers, Pharmacology & Toxicology, Wright State University, Dayton, Ohio, United States
Ethanol, in combination with thermal burn injury, is a clinically significant problem resulting in an increased morbidity and mortality due to acute multi-organ dysfunction from excess systemic cytokine release. Moreover, murine models of intoxicated burn injury replicate the acute toxic effects as well as a delayed systemic immunosuppression. Almost half of the admitted hospital patients with burn injuries were alcohol intoxicated at the time of admission. Our group has demonstrated that the lipid mediator Platelet-activating factor (PAF) plays an important role in the delayed immunosuppressive effects of intoxicated thermal burn injury. As PAF receptor signaling causes generation of subcellular microvesicle particles (MVP), we aimed at defining the role of MVP in the toxicity associated with EtOH + burn injury. Using HaCaT keratinocyte-derived cell line, we demonstrate that both thermal burn injury and EtOH alone generate increased release of MVP into the supernatant and that the two agents results in an increased generation of MVP in at least an additive fashion. These studies suggest that MVP might play a role in the augmented toxicity of intoxicated thermal burn injury.

Phenotypic heterogeneity of heterozygous carrier of cytosisis 8 (DOCK8) mutations
Joanne Paleologou, Medical School, Augusta University, Augusta, Georgia, United States
Background: Cytosisis 8 (DOCK8) deficiency leads to a combined immuno-deficiency characterized by cutaneous and systemic infections, atopy, and autoimmunity. Although there is limited information about simple or complex heterozygous mutations in the DOCK8 region or whether heterozygosity for DOCK8 is associated with cutaneous or systemic infections. Inclusion of heterozygous mutation analysis may improve the collective understanding of DOCK8 genetic variants. Objective: The purpose of this review is to provide useful insights into the clinical features, genetic analysis and genetic variants of DOCK8 deficiency, with particular emphasis on heterozygous DOCK8 mutations. Methods: PubMed, NCBI, and Medline were searched for scientific articles that provide information on DOCK8 deficiency. The keywords queried included “DOCK8 deficiency”, “mutations”, “heterozygous”, and “genetics”. Results: Due to the vital role that DOCK8 plays in the immune system as well as the actin cytoskeleton, the clinical hallmarks of DOCK8 deficiency are recurrent upper respiratory tract, cutaneous infections and eczema. Although most symptomatic individuals have homozygous autosomal recessive mutations, this review revealed similar clinical findings in patients with heterozygous DOCK8 mutations. Conclusion: Although individuals with heterozygous DOCK8 mutations are generally asymptomatic, recent literature highlights the vast phenotypic heterogeneity of these individuals.

Genotyping of 30 kinds of cutaneous human papillomaviruses by a multiplex microfluidic loop-mediated isothermal amplification and visual detection method
Y Wang, R Qi, X Gao and H Chen, The First Affiliated Hospital of China Medical University, Shenyang, China, Human papillomaviruses (HPVs), a group of non-enveloped small viruses with double-stranded circular DNA which lead to multiple skin diseases such as benign warts, are commonly seen in clinics. The current HPV detection systems aim mainly at mucosal HPVs, however, an efficient clinical approach for cutaneous HPVs detection is lacking. To establish a rapid detection system for cutaneous HPVs, we used a colorimetric loop-mediated isothermal amplification (LAMP) with hydroxynaphthol blue (HNB) dye in combination with microfluidic technology. The lower detection limit of the LAMP assay was 107 viral DNA copies/ml when tested on synthesized L1 DNA sequences, which was better than the conventional PCR. Compared to PCR sequencing, the sensitivity of HPV27, HPV2, HPV1, HPV57, HPV5, HPV14, HPV7 and HPV75 genotypes detections were 100%, whereas the detection specificity was 14.55%, 45.12%, 95.83%, 98.59% and 97.62% respectively, when tested on clinical samples. The new cutaneous type HPV detection system is characterized by both a good sensitivity and specificity compared to conventional methods.

Age and circadian regulation of cutaneous innate antimicrobial activity at homeostasis
V Lee1, SI Kirchner1,2, J Kwock1, X Ling1, DL Corcoran2 and AS MacLeod3, 1 Dermatology, Duke University School of Medicine, Durham, North Carolina, United States, 2 Molecular Genetics and Microbiology, Duke University School of Medicine, Durham, North Carolina, United States, 3 Center for Genomic and Computational Biology, Duke University School of Medicine, Durham, North Carolina, United States
Neonates and the elderly are at increased risk for skin viral and bacterial infections that have the potential to cause systemic malaise and death. Innate antimicrobial proteins (AMPs), a component of the natural antimicrobial defense program, are expressed in the skin and protect the host against potential viral infections. Interestingly, newborns and the elderly have immature diurnal circadian rhythms. Unknown to us is whether and how changes in circadian regulation throughout age alter homeostatic innate antimicrobial immune responses in the skin. We hypothesize that age-related circadian rhythmicity at homeostasis. Notably, skin from B6ma1+ mice showed reduced antimicrobial transcription compared to heterozygous littermates, and siRNA knockdown of CLOCK in human keratinocytes leads to reduced basal HPV mRNA expression (p < 0.05). Intriguingly, a subset of cutaneous AMPs mRNA are not expressed in neonatal mice, but peak AMP expression induction with increased transcript levels was observed in adult mice (p < 0.05). Moreover, elderly mice show reduced constitutive AMP expression levels (p < 0.05) and protein expression compared to adult mice. These findings suggest that variability in AMP expression across age may be due to changes in circadian regulation of keratinocyte innate immune programs. Future studies are directed to investigate whether age and associated changes in circadian regulation similarly impact anticipatory innate immune responses in the setting of wounds.