Skin Sciences Institute

Division Photo

First Row: *R. Boissy, M. Visscher, V. Narendran;* ***Second Row:*** *G. Kasting, S. Hoath, R. Wickett*

Division Data Summary

Research and Training Details

Number of Faculty	3
Number of Joint Appointment Faculty	9
Number of Research Students	5
Number of Support Personnel	19
Peer Reviewed Publications	17

Clinical Activities and Training

Number of Clinical Students	2

Faculty Members

Steven B. Hoath, MD, Professor ; *Medical Director, The Skin Sciences Institute*

Marty O. Visscher, PhD, Research Professor ; *Director, The Skin Sciences Institute*

Vivek Narendran, MD, Associate Professor ; *Director Newborn Nursery Christ Hospital, Director NICU University Hospital, Director Perinatal Outreach Program*

Raymond E. Boissy, PhD, Professor ; *Director, National Vitiligo Foundation*

W. John Kitzmiller, MD, Associate Professor ; *Head, Division of Plastic and Reconstructive Surgery; Director of Wound Care Drake Hospital*

Steven T. Boyce, PhD, Professor ; *Director, Department of Tissue Engineering, Shriners Burns Institute*

R. Randall Wickett, PhD, Professor ; *Director, Skin Pharmaceutics and Cosmetic Science Graduate Program*

Gerald B. Kasting, PhD, Associate Professor ; *Past Co-chairman, Gordon Research Conference*

Diya F. Mutasim, MD, Professor ; *Chairman, Department of Dermatology*

Charles L. Heaton, MD, Professor ; *Director, Sexually Transmitted Diseases Clinic and Training Program, Department of Health*

Zalfa Abdel-Malek, PhD, Associate Professor

Neville G. Pinto, PhD, Professor ; *Assistant Dean of Graduate Studies*

Trainees

- **Jodi Eubanks, PhD,**
- **Marisa Robinson, PhD,**
- **Jennifer Davis, PhD,**
- **Santosh Yadav, PhD,**
- **Allison Protain, MD,**
- **Shannon O'Brien, MD,**
- **Maria Victoria deCastro, MSN,**
- **Lesa Combs, ,**
- **Nancy Schwegman, ,**
- **Jill Winer, ,**
- **Lori Perkins, ,**
- **Claire Burkhart, ,**
- **Teresa Taylor, ,**
- **Theresa Flower, ,**
- **Tamina White, ,**
- **Linda Sluder, ,**
- **Shelly Sargent, ,**
- **Louise Smith, ,**
- **Maureen Rider, ,**
- **Amy Heubner, ,**
- **Elizabeth Mason, ,**

Significant Accomplishments in FY08

Host Defense Proteins on Neonatal Skin: Implications for Innate Immunity

Reduction in infant mortality is a global health goal. Simple skin care practices, e.g., oil massage, reduce infection rates among premature infants in developing countries. The skin provides innate immunity by functioning as a physical barrier and deterrent for pathogenic microbial invasion. During gestation, the fetis is encased in vernix caseosa and amniotic fluid, both of which contain host defense proteins (HDPs). However, birth eliminates this protection and exposes the skin to environmental microbes. We tested the hypothesis that the stratum corneum of the newborn contains a variety of active HDPs consistent with an important aspect of neonatal innate host defense beyond the physical barrier properties. We compared the spectrum, concentration, and activity of HDPs on the skin of neonates and adults and examined the effect of skin care practices, i.e., bathing. Lysozyme and lactoferrin were found in all samples. Lysozyme concentrations and muramidase activity were 5-fold higher in newborns and were not altered with bathing. These factors are likely to contribute importantly to the newborn infants' defense against bacterial infections.

Skin Care in the High Risk Neonate

NICU patients are at risk for skin breakdown due to prematurity, irritant exposure, and stress. There is a need to minimize damage, facilitate skin development and reduce infection risk infection, but the literature on the effects of skin care practices in high risk infants is limited. In collaboration with clinical staff from the CCHMC Regional Center for Newborn Intensive Care (RCNIC), we tested the hypothesis that diaper wipes with emollient cleansers and a soft cloth would minimize skin breakdown relative to the current RCNIC practice standard of a cloth and sterile water. In 130 infants (GA 23-41 weeks, at enrollment 30-51 wks), erythema, rash, transepidermal water loss (TEWL) and pH were determined daily for 5-14 days using standardized methods of skin integrity within the diaper and at diaper and chest control sites. Treatments were Wipe A, Wipe B, and cloth and water. Perineal erythema and TEWL were significantly

lower for wipes than cloth and water. Wipe B produced a significantly lower skin pH than Wipe A and cloth and water. The starting skin condition, stool total, age and time on current standard impacted the outcomes. Both wipes are appropriate for use on high risk premature and full term neonates and provide more normalized skin condition and barrier function versus the cloth and water standard. Wipe B may facilitate acid mantle development and assist in colonization, infection control and barrier repair. Neonatal skin continues to change for up to eight weeks postnatally, presumably as it adapts to the dry terrestrial environment. Provision of an acidic skin pH, as demonstrated for Wipe B, may offer an additional approach to facilitate barrier repair and acid mantle development in neonates at risk for skin compromise.

Quantitation of Epidermal and Mucosal Tissue Injury as a Function of Skin Pigmentation Using Imaging Techniques

Epidermal skin injury is common and can be caused by mechanical trauma (friction), exposure to solvents and chemical irritants, heat and environmental conditions (e.g., humidity). Stratum corneum (SC) barrier compromise allows penetration of irritants into the dermis. Dryness (low moisture), scaling, fissuring, erythema (inflammation) and itching can occur as cellular repair mechanisms are upregulated. Visual scoring methods based on severity (e.g., slight, mild, moderate, severe) and area (percent) of involvement are typically used to measure damage. However, the accuracy of visual methods is significantly impacted by the inherent skin pigmentation, varying from very light (Caucasian) to very dark (African). Erythema is difficult to assess in the darker skin due to masking by the red, yellow and brown pigments. As a consequence, tissue damage may be underdetermined and/or treated less aggressively in dark skinned patients. Epidermal and mucosal scratches were created in 20 light (L^* 68.2 ±2.3) and 20 dark skinned (L^* 46.4 ± 5.2) females. Injured and uninjured sites were treated with toluidine blue (TB), fluorescein (FL) and a TB/FL mixture and imaged under conditions of white and fluorescent light. Injured sites with TB and TB/FL had higher areas than the control for both light and dark subjects (ANOVA, $p < 0.05$). The intensity of the injured TB site was higher than the control for light skin only. The areas of injured sites with FL and TB/FL were higher than the control for both groups as were the intensities of the injured sites with FL. Application of these contrasts under white and fluorescent light can be used to quantify tissue injuries for L^* values > 35 and is a promising approach for the quantitation across a range of skin pigmentation.

Significant Publications in FY08

Yoshida Y, Hachiya A, Sriwiriyanont P, Ohuchi A, Kitahara T, Takema Y, Visscher M, Boissy R. Functional analysis of keratinocytes in skin color using a human skin substitute model composed of cells derived from different skin pigmentation types. FASEB J. 21, 2829-2839 (2007).

Reports in the literature suggested that PAR-2 mediated transfer of melanosomes was involved in the regulation of skin pigmentation. By grafting mixtures of keratinocytes and melanocytes from a combination of donor types with light skin fibroblasts onto the severe combined immunodeficient mice, we demonstrated the role of the keratinocyte in regulation of melanogenisis and distribution of transferred melanosomes.

Gibbins S, Hoath SB, Coughlin M, Gibbins A, Frank L. The universe of developmental care: A new conceptual model for application in the neonatal intensive care unit. Foundations in Newborn Care. Advances in Neonatal Care 8(3): 141-147 (2008).

While developmental care for high-risk infants is practices in most neonatal units around the world, inconsistency in the definition and application of the principles has resulted in criticism regarding its scientific merit. We propose the universe of developmental care model as the first major reformulation since Als' synactive theory. Central is the concept of a shared surface, manifested by the skin that forms the link between the organism and environment and becomes the focal point for human interactions. The theoretical underpinnings and components serve to guide future research.

Division Highlights

Epidermal Barrier Development and Vernix Biology

The premature infant has a poor epidermal barrier with few cornified layers, experiences high fluid loss, and is at risk for increased permeability to exogenous agents (e.g., infectious agents). A poor stratum corneum (SC) barrier poses risk for additional skin compromise, e.g., from adhesives, fecal enzymes, that may cause inflammation and further delay maturation. During latter gestation, the infant is encased in vernix caseosa, a viscous white material that plays an essential role in the development of the stratum corneum and is lacking in the premature neonate. Vernix is composed of water (80%), lipids (10.3%) and protein (9.1%). The hydrophobic lipids are of sebaceous and stratum corneum and surround fetal cellular components. Vernix has been investigated as a defense against infection both pre- and postnatally and found to have beneficial physiochemical properties that provide barrier protection and facilitate acid mantle development. Despite the very high water content, vernix films are permeable to water vapor and exhibit slow

water release. Vernix corneocytes without the lipid matrix had increased equilibrium water binding at water activities greater than 0.62 compared to native vernix. During resorption, both vernix and the isolated corneocytes showed full recovery of water content, thereby supporting the presence of a structured internal domain. Additionally, we examined the effect of vernix in vitro on the penetration of □-chemotropism, a potentially injurious proteolytic enzyme present in the developing epidermis, meconium and feces. Vernix films significantly impeded enzyme penetration. No □-chymotryptic activity was detected in vernix nor did it inhibit activity. The results are consistent with the hypothesis that vernix films retain endogenous (epidermal) chymotrypsin while preventing exposure to exogenous (e.g., pancreatic) chymotrypsin and further support the protective effects of vernix.

Based on these investigations and our previous findings of the multifunctionality of native vernix, we have developed a synthetic barrier cream analog. The water release profiles are similar to those of the native benchmark. The synthetic formulation significantly impeded a-chymotrypsin penetration and function as a protective barrier. In collaboration with the Center for Technology Commercialization, efforts to license the CCHMC patented vernix technology are making significant progress.

Mechanisms of Human Skin Pigmentation

Inflammatory conditions such as wounds, acne, burns, and skin grafting can result in localized disorders of pigmentation (hyper, hypo) and associated patient disfigurement. An understanding of the molecular mechanisms underlying the observed pigmentary changes across the diverse spectrum of inherent skin coloration is essential for the development of effective treatments. Recently, we used an in vivo xenograft system to determine the role of the keratinocytes in skin coloration by using cultured keratinocytes and melanocytes from light and dark skin donors in various combinations. The epidermal melanin content and the maturation stage of melanosomes in basal keratinocytes were significantly higher is cultures of dark skin derived keratinocytes. The ratio of individual/clustered melanosomes in recipient keratinocytes was increased in dark skin donor tissue. The genetic expression of endothelin-1, proopiomelanocortin, microphthalmia-associated transcription factor, tyrosinase, CP100 and MARTI were higher for tissue from dark skin donors. The findings demonstrate that the expression of melanogenic cytokines, maturation of melanosomes, melanin synthesis and melanosome distribution in human skin substitutes are influenced by the racial origin of the keratinocytes.

Clinically, modulation of melanogenesis in the melanocytes can currently be achieved using structural homologies of the substrate tyrosine to competitively inhibit the catalytic function of tyrosinase. Deoxyarbutin is our tyrosinase inhibitor based on this premise. In the pigmented guinea pig model, it demonstrated rapid and sustained skin lightening, in contrast to the currently marketed agent hydroquinone. In human clinical trials, topical treatment of pigmented areas resulted in significant skin lightening relative to placebo. To date, the safety profile indicates deoxyarbutin to be actionable in various geographies. CCHMC has licensed the technology.

Division Collaboration

Collaboration with Critical Care; Patient Services

Collaborating Faculty: Pattie Bondurant; M. Victoria deCastro, Lisa Combs, Lori Perkins, Nancy Schwegman, Jill Winer, Claire Burkhart, Teresa Taylor, Tamina White, Shelly Sargent, Louise Smith, Theresa Flower, Maureen Rider, Amy Heubner, Elizabeth Mason, Linda Sluder

Conducted two clinical and translational research projects among patients in the Regional Center for Newborn Intensive Care: (1) Wipes Versus Current Standard for Diaper Skin Care and (2) Effect of Chlorhexidine Gluconate on the Skin Integrity at PICC Line Sites.

Collaboration with Critical Care; Physical Medicine & Rehabilitation; OPD Base Treatment Center; Center for Professional Excellence; Patient Services; ;

Collaborating Faculty: Pattie Bondurant, Terry Palmisan; David Pruitt; Ann Marie Nie; Pat Schafferm, Diana Bailey; Sandy Conn, Clare Duane, Mary Jo Giaccone, Gayle Hertenstein, Debbie Hershberger, Veronica Jackson, Lisa Knapp, Patricia McLain, Laura Miller, Mary Porter, Lori Puthoff, Elixabeth Ritzi, Elisa Schaffer, Laura Schroer, Mary Stange, Teresa Taylor; ;

Patient Safety Breakthrough Collaborative, Pressure Ulcers Team. Improvement science based initiative to reduce the incidence of pressure uclers among the pediatric population by developing a skin care bundle. The team identified goals and key drivers, developed interventions and formulated education for spread throughout CCHMC.

Collaboration with Neurology; Biostatistics and Epidemiology

Collaborating Faculty: Tanya Phillips; Jareen Meinzen-Derr

Examine and evaluate evidence supporting the use of amplitude-integrated EEG as a quantitative predictor of neurodevelopmental outcome in full-term infants with hypoxic ischemic encephalopathy.

Collaboration with Biosatistics and Epidemiology

Collaborating Faculty: Jareen Meinzen-Derr

Investigations of host defense proteins on the surface of neonatal skin and the implications for innate immunity

Division Publications

1. Westbroek W, Adams D, Huizing M, Koshoffer A, Dorward H, Tinloy B, Parkes J, Helip-Wooley A, Kleta R, Tsilou E, Duvernay P, Digre KB, Creel DJ, White JG, Boissy RE, Gahl WA. **Cellular defects in Chediak-Higashi syndrome correlate with the molecular genotype and clinical phenotype**. *J Invest Dermatol.* 2007; 127: 2674-7.
2. Gibbins S, Hoath SB, Coughlin M, Gibbins A, Franck L. **The universe of developmental care: a new conceptual model for application in the neonatal intensive care unit**. *Adv Neonatal Care.* 2008; 8: 141-7.
3. Spitzmiller RE, Phillips T, Meinzen-Derr J, Hoath SB. **Amplitude-integrated EEG is useful in predicting neurodevelopmental outcome in full-term infants with hypoxic-ischemic encephalopathy: a meta-analysis**. *J Child Neurol.* 2007; 22: 1069-78.
4. Tansirikongkol A, Hoath SB, Pickens WL, Visscher MO, Wickett RR. **Equilibrium water content in native vernix and its cellular component**. *J Pharm Sci.* 2008; 97: 985-94.
5. Gunt HB, Kasting GB. **Equilibrium water sorption characteristics of the human nail**. *Int J Cosmet Sci.* 2007; 29: 487.
6. Gunt HB, Kasting GB. **Effect of hydration on the permeation of ketoconazole through human nail plate in vitro**. *Eur J Pharm Sci.* 2007; 32: 254-60.
7. Gunt HB, Miller MA, Kasting GB. **Water diffusivity in human nail plate**. *J Pharm Sci.* 2007; 96: 3352-62.
8. Kasting GB, Bhatt VD, Speaker TJ. **Microencapsulation decreases the skin absorption of N,N-diethyl-m-toluamide (DEET)**. *Toxicol In Vitro.* 2008; 22: 548-52.
9. Kretsos K, Kasting GB. **A geometrical model of dermal capillary clearance**. *Math Biosci.* 2007; 208: 430-53.
10. Kretsos K, Miller MA, Zamora-Estrada G, Kasting GB. **Partitioning, diffusivity and clearance of skin permeants in mammalian dermis**. *Int J Pharm.* 2008; 346: 64-79.
11. Wang TF, Kasting GB, Nitsche JM. **A multiphase microscopic diffusion model for stratum corneum permeability. II. Estimation of physicochemical parameters, and application to a large permeability database**. *J Pharm Sci.* 2007; 96: 3024-51.
12. Walker VP, Akinbi HT, Meinzen-Derr J, Narendran V, Visscher M, Hoath SB. **Host defense proteins on the surface of neonatal skin: implications for innate immunity**. *J Pediatr.* 2008; 152: 777-81.
13. Tansirikongkol A, Visscher MO, Wickett RR. **Water-handling properties of vernix caseosa and a synthetic analogue**. *J Cosmet Sci.* 2007; 58: 651-62.
14. Yoshida Y, Hachiya A, Sriwiriyanont P, Ohuchi A, Kitahara T, Takema Y, Visscher MO, Boissy RE. **Functional analysis of keratinocytes in skin color using a human skin substitute model composed of cells derived from different skin pigmentation types**. *FASEB J.* 2007; 21: 2829-39.
15. Eppler AR, Kraeling ME, Wickett RR, Bronaugh RL. **Assessment of skin absorption and irritation potential of arachidonic acid and glyceryl arachidonate using in vitro diffusion cell techniques**. *Food Chem Toxicol.* 2007; 45: 2109-17.
16. Tansirikongkol A, Wickett RR, Visscher MO, Hoath SB. **Effect of vernix caseosa on the penetration of chymotryptic enzyme: potential role in epidermal barrier development**. *Pediatr Res.* 2007; 62: 49-53.
17. Wickett RR, Kossmann E, Barel A, Demeester N, Clarys P, Vanden Berghe D, Calomme M. **Effect of oral intake of choline-stabilized orthosilicic acid on hair tensile strength and morphology in women with fine hair**. *Arch Dermatol Res.* 2007; 299: 499-505.