National Atopic dermatitis Program

BACKGROUND AND SIGNIFICANCE

Eczema, or Atopic Dermatitis (AD), is the most common skin disorder in Singapore and represents a large clinical burden. National Skin Centre alone sees around 15,000 cases annually, of which 3,000 are new cases. AD usually begins in childhood and affects one-fifth of all Singaporean schoolchildren (Tay, 2002). It is characterised by dry, itchy and inflamed skin that causes pain and distress, loss of sleep, and school and work absences. It is highly detrimental to the quality of life of both sufferers and their care takers (Drucker 2017). A study on adult AD cohort also highlighted a positive correlation between disease severity and depression (Lim, 2016).

Globally, AD appears to have a similar prevalence in developed countries, and is projected to increase at an annual growth rate of 0.17% (GlobalData, 2014). Researchers have also reported higher financial burden on a familiy for AD compared to Type 1 diabetes mellitus (Williams 2008). Nothwithstanding the high prevalence, high personal and social cost of AD, there has been a dearth of innovation in AD therapeutics, especially for the severe recalcitrant population and research in this area has been underfunded globally.

AIM OF THE NATIONAL ATOPIC DERMATITIS RESEARCH PROGRAM.

The overall objective of this comprehensive translational research program is make Singapore one of the global leaders in AD through a better understanding of the epidemiology and pathogenesis of AD in the Asian context, and to develop cutting-edge diagnostic, therapeutics and patient engagement tools to improve the health outcomes of this chronic disease. Using a systematic and multidisciplinary approach, we aim to reduce the numbers affected by severe AD by 30% and to reduce the prevalence of AD by 10% by 2023, while attracting industry interest and creating economic value for Singapore. (The combined markets of US, France, Germany, Italy, Spain, UK and Japan alone were US$3,6B in 2014 and are projected to grow to US$7.3B by 2024 (GlobalData, 2014)). To achieve these targets, we will bring together dermatologists from all major healthcare institutions in Singapore together with leading skin biologists, biophysicists, imaging experts, biomedical engineers and material scientists, in 4 interconnected and complementary theme approaches: -

Theme 1. Epidemiology, disease burden and Systems Biology of Atopic Dermatitis. The aim of Theme 1 is to collect comprehensive local data on the epidemiology and disease burden of AD in Singapore, and to build an AD patient cohort for an integrated systems biology approach to the disease. The specific objectives of this theme for the duration of the grant are:

1. To build a good understanding of the epidemiology and health burden of AD as it occurs in Singapore.
2. To design and build a cohort of at least 2000 Asian AD patients and 2000 controls, which will provide study material for Themes 1- 4.
3. To build a better undertsanding of the pathogenesis of Asian AD by a systems biology approach based on the cohort study, to map out the complex interactions between identified novel genetic, epigenetic biomarkers, microbiome, immunological and imaging profiles with clinical phenotype and severity. Identified novel biomarkers that can be potential targets for interventions will be explored in Theme 2 and Theme 4.

Hypothesis. We hypothesize that the multifactorial pathogenesis of AD arises from the complex interaction of genetics, epigenomics, environment and microbiome of patient. As such, a systems biology approach, using a combination of clinical, genomic, transcriptomic and imaging data from our Asian AD cohort will allow us better to understand Asian AD pathogenesis. We further hypothesize that novel biomarkers, drug targets and intervention strategies can be identified from this systems biology approach and can be validated in Theme 2 for subsequent trials in Theme 4.

Theme 2. Precision interventions through discovery of novel targets in barrier, immunology and itch pathways and establishment of relevant AD models. The overarching aim of this theme is to investigate pathogenetic mechanisms that characterise Asian AD, by in depth study of the skin barrier, immunology and

itch in Asian AD patients. We will also develop pathophysiologically relevant *in vitro* AD models and specific animal models to validate findings arising from Themes 1 and 2. The specific aims of this theme are:

1. Skin barrier and microbiome:
    a. Genetic stratification of AD cohort, built in Theme 1: Filaggrin gene mutations are the major genetic risk factor associated with AD (Palmer, 2006) but approxiately 70% of patients in Singapore do not appear to have known defects in filaggrin. We will therefore stratify the cohort built in Theme 1 according to filaggrin mutation status coupled with further in depth analysis to identify novel genetic and epigenetic factors important for Asian AD.
    b. Characterisation of the AD microbiome: The skin microbiome is known to be dysbiotic during AD flares and in between AD flares (Chng, 2016). Analysis of microbiome composition in the AD cohort will contribute another important layer of information to help understand the complex multi-factorial interplay between host genetics, immune profile and the microbiome metagenome. Strain and functional capacity analysis will provide insights for intervention.
2. Immunological profiling: It is now well known that the immunological status of the skin is an important part of AD. In this subtheme, we will use immunomonitoring and immune analytics combining cell sorting, genomics and proteomics of tissue samples to develop an integrated picture of the activity of the whole immune system in AD skin. The specific aims are:
    a. To conduct high dimensional analysis of the immune cell composition to map the full immune signature in atopic dermatitis.
    b. To identify unique subsets of immune populations, with specific function that reflect distinct underlying immuno-pathogenic mechanisms and pathways in AD.
3. Itch is the most common symptom and the main source of morbidity in AD. Despite this, its pathophysiology is still very poorly understood. Our specific aims are:
    a. To identify the itch mediators and receptors which are altered in lesional skin compared to non-lesional skin of patients with AD, and to determine how their expression and distribution is correlated with cutaneous nerves.
    b. To identify novel targets of intervention for itch and explore the use of novel peptides to down-regulate such itch mediators and/or their receptors.
4. Pathophysiologically relevant AD models.
    a. Use pathophysiologically relevant animal models of AD, including the *Flaky tail* mouse/filaggrin deficient mouse, to study pathomechanisms and investigate novel therapeutics.
    b. Complimentary to well-characterized mouse models above, we will develop new disease models of AD to investigate novel predicted genetic targets and pathways as indentified in this theme, such as for example the conditional knockout of N-WASP in keratinocytes using K14-Cre, which results in a mouse with an AD phenotype.

Hypothesis. We hypothesize that besides filaggrin mutations, there will be other genetic factors of barrier dysfunction, possibly for example affecting tight junctions or desmosomes, which contribute to Asian AD. Developing novel models will help uncover these new barrier defects. We further hypothesize that linking eczema, skin barrier, itch pathways and immune system will provide insights for the management of AD, especially in the use of drugs to repair skin barrier, control itch/scratch cycle and regulate the immune system.

Theme 3. Rapid diagnostics and automated outcome measures from emerging imaging technologies. The overarching aim of this theme is to develop rapid diagnostics to stratify disease and to prognosticate disease progression. The specific objectives of this theme are: -

1. To develop an accelerated diagnostic test for filaggrin defects and other barrier dysfunction mutations for routine use in the clinics.
2. To develop and validate use of confocal Raman imaging for molecular level quantification of natural moisturizing factors, ceramides and filaggrin fragments.
3. To develop specific noise reduction, image enhancement algorithms with novel machine learning and biomedical image informatics to automatically detect and score AD severity in digital photos. This will be deployed in mobile Apps to be developed in Theme 4 for remote monitoring of patients and for patient engagement tools.

Hypothesis. We hypothesize that the state-of-the-art imaging modalities like confocal Raman, multispectral optocoustic tomography and micro-OCT will enable in vivo measurement of barrier function in normal, pre-disease and disease patients by quantifying levels of ceramides, natural moisturizing factor and filaggrin in

patient's skin, thus providing new prognostic indicators of Asian AD and allowing for a tailored approach to treatment. We further hypothesize that novel machine learning techniques can be used to automatically quantify the burden of disease in patients, enabling clinicians and patients to easily and objectively score disease severity. Such algorithms, when deployed in mobile apps, will also empower patients to better track their skin conditions and institute preventive measures when necessary.

Theme 4. Novel therapeutics options and enhancing compliance through gamification.

The overarching aim of this theme is to translate novel peptides, new drug targets identified in Theme 1-3, and test them in AD patients. Novel strategies to engage patient through mobile apps and gaming will also be used to improve compliance and remotely monitor patients, and improve patient self-care to reduce admission to hospitals. The specific aims of this theme are: -

1. To develop new class of topical immunomodulators for AD resistant to topical steroids or calcineurin inhibitors. Topical Methotrexate would be the first to be developed by NTU School of Material Science and Engineering and tested in AD patients in Singapore. The next class of novel immunomodulators to be developed are Kv1.3 inhibitors developed by Prof Chandy, LKC Medicine, NTU.
2. To develop and standardise trial protocols for the different types of AD and establish the Skin Research Institute of Singapore as the centre of choice for testing of novel therapeutics, hopefully biologics for AD developed in Theme 1-3 or by Phama companies
3. To develop novel nanoparticles and/or microneedle patches for precise and sustained delivery of actives (discovered in Theme 1-3) into specific depth of skin for optimal efficacy and minimal adverse effects.
4. To develop mobile apps to enable patients to actively monitor their eczema status through self inputs as well as wearables. The apps will dynamically suggest changes to treatment regimen based on changes in environment parameters, eczema, itch and sleep scores. The apps will also use the theory of gamification to improve compliance to therapy.

Hypothesis. We hypothesize that research into novel delivery methods using nanoparticles and microneedle patches will enable us to improve delivery through the skin barrier and thus re-purpose old systemic drugs like methotrexate for use topically, improving efficacy and reducing systemic side effects. We further hypothesize that Kv1.3 inhibitors (whether given systemically or topically) would be the next paradigm shift in management of AD. Finally, successful long-term management of AD often requires patients to be well-educated on potential disease triggers and on applying treatments correctly. Thus, for the subtheme on patient education and empowerment, our central hypothesis is that empowering adult patients through active self-monitoring, reminders and gamification will enhance compliance to treatment regimen and thus improve treatment outcomes:-

The main PIs and composition of the 4 themes are shown in the table below:-

Theme no.	Theme Title	Theme PI(s) and Institution
Theme 1	Epidemiology, disease burden and Systems Biology of Atopic Dermatitis.	A/Prof Josip Car, LKC Medicine Dr Yew Yik Weng, NSC Dr Mark Koh, KKH Prof Olaf Rotzschke, SIgN A*STAR A/Prof Chew Fook Tim, NUS
Theme 2	Precision interventions through novel biomarkers, pathways and establishment of relevant animal models.	Prof Birgit Lane, IMB A*STAR Prof Maurice van Steensel, IMB A*STAR Dr John Common, IMB A*STAR A/Prof Florent Ginhoux, SIgN A*STAR

Theme 3	Automated diagnostics and outcome measures from emerging imaging modalities	A/Prof Steven Thng, NSC Prof Teoh Swee Hin, NTU Prof Malini Olivo, SBIC A*STAR Prof Birgit Lane, IMB A*STAR
Theme 4	Novel therapeutics options and enhancing compliance through gamification	A/Prof Steven Thng A/Prof Tan Suat Hoon, NSC Prof Subbu Venkatraman, MSE, NTU Prof Teoh Swee Hin, NTU Prof George Chandy, LKC Medicine, NTU Dr Yew Yik Weng, NSC Dr Mark Koh, KKH