Institution: University of Dundee
Unit of Assessment: UoA1 Clinical Medicine
Title of case study: Filaggrin - the major predisposing gene for atopic disease and a target for stratified therapeutic intervention
1. Summary of the impact

Atopic eczema and associated conditions – asthma, food allergy and hay fever – affect ~40% of the population in developed nations. They cause significant morbidity and create a multibillion-pound global healthcare burden. The discovery that loss-of-function mutations in the gene encoding filaggrin represent a strong risk factor for eczema, asthma and peanut allergy has defined a key pathological mechanism in atopic disease. This breakthrough in understanding has brought new focus on the skin barrier. It has shown impact in treatment approaches to maintain barrier function, translational research targeting epithelial dysfunction and improved public and professional awareness of the role of skin in atopic disease.

2. Underpinning research

Atopic eczema is a complex trait, in which multiple genetic risk factors interact with environmental factors in disease pathogenesis. Historically, research in atopic disease has focused on the immune response in the investigation of disease pathogenesis and in therapy development.

The research underpinning our breakthrough in understanding atopy pathogenesis dates back to 2006, when the group of Prof Irwin **McLean** (Professor of Human Genetics and Head of the Division of Molecular Medicine, University of Dundee) reported that the common monogenic skin disease, ichthyosis vulgaris (characterised by dry, scaly skin), is caused by loss-of-function mutations in the gene encoding filaggrin (*FLG*) [i]. The gene product profilaggrin is cleaved to produce monomeric filaggrin, playing a role in keratin filament aggregation, skin barrier formation and cutaneous hydration.

The **McLean** group made a seminal discovery, demonstrating that up to 50% of severe childhood eczema cases carry *FLG* loss-of-function mutations [ii,iii]. This created a paradigm shift in the eczema/allergy field by showing that one of the primary driving forces underlying common atopic disorders is impaired skin barrier function. These findings support a model for eczema aetiology whereby skin barrier dysfunction allows entry of allergens and irritants resulting in skin and systemic inflammation. Additional compelling evidence in support of this hypothesis was provided in 2009 when the **McLean** group published the first mouse model of filaggrin-related eczema and demonstrated that impaired skin barrier function leads to skin and systemic inflammation triggered by percutaneous allergen stimulation [iv].

The gene *FLG* is difficult to analyse due to its large size and highly repetitive sequence [i-iii]. Techniques developed by Prof **McLean** and colleagues to analyse other epidermal structural genes led to ground-breaking discoveries in rare genodermatoses throughout the 1990s and subsequently enabled the sequencing of *FLG* in advance of international competitors. A complex pattern of prevalent and rare *FLG* mutations in different population groups has now been identified. Longitudinal population-based genetic studies have confirmed that *FLG* is a major genetic factor in eczema, asthma and allergic rhinitis and that *FLG* haploinsufficiency is particularly associated with severe, early onset and persistent disease. Five recent genome-wide association studies and one meta-analysis have confirmed that *FLG* is the strongest genetic risk in atopic eczema, with an odds ratio >3. Furthermore *FLG* remains the only locus in which a relationship has been unequivocally demonstrated between gene and disease pathomechanism.

In 2011 Prof **McLean** and Dr Sara **Brown** (Wellcome Trust Intermediate Clinical Fellow, Clinical Senior Lecturer and Honorary Consultant Dermatologist, University of Dundee) led an international collaboration to investigate the role of *FLG* mutations in IgE-mediated peanut allergy. They reported a strong association with replication in independent population groups [v], representing

the first established genetic risk factor in this severe food allergy. Dr **Brown** also demonstrated in 2012 that copy number variation within *FLG* contributes to eczema risk with a dose-dependent effect [vi], illustrating the potential clinical utility of therapies aimed to increase filaggrin expression.

The University of Dundee has filed two patents in developing *FLG* genotyping as part of a personalised medicine approach for the treatment/prevention of atopic disease and for enhancement of filaggrin expression as a novel therapy. These patents underpin grant income to the **McLean** group from MRC and MRC Developmental Pathway Funding Scheme (totalling ~£1.6million) in collaboration with the Drug Discovery Unit, University of Dundee.

3. References to the research

i. Smith FJD, Irvine AD, Terron-Kwiatkowski A, Sandilands A, Campbell LE, Zhao Y, Liao H, Evans AT, Goudie DR, Lewis-Jones S, Arseculeratne G, Munro CS, Sergeant A, O'Regan G, Bale SJ, Compton JG, Digiovanna JJ, Presland RB, Fleckman P, **McLean** WHI (2006) Loss-of-function mutations in the gene encoding filaggrin cause ichthyosis vulgaris. *Nat. Genet.* **38**, 337-42 (DOI: 10.1038/ng1743).

ii. Palmer CNA, Irvine AD, Terron-Kwiatkowski A, Zhao Y, Liao H, Lee SP, Goudie DR, Sandilands A, Campbell LE, Smith FJD, O'Regan GM, Watson RM, Cecil JE, Bale SJ, Compton JG, DiGiovanna JJ, Fleckman P, Lewis-Jones S, Arseculeratne G, Sergeant A, Munro CS, El Houate B, McElreavey K, Halkjaer LB, Bisgaard H, Mukhopadhyay S and **McLean** WHI (2006) Common loss-of-function variants of the epidermal barrier protein filaggrin are a major predisposing factor for atopic dermatitis. *Nat. Genet.* **38**, 441-6 (DOI: 10.1038/ng1767).

iii. Sandilands A, Terron-Kwiatkowski A, Hull PR, O'Regan GM, Clayton TH, Watson RM, Carrick T, Evans AT, Liao H, Zhao Y, Campbell LE, Schmuth M, Gruber R, Janecke AR, Elias PM, van Steensel MAM, Nagtzaam I, van Geel M, Steijlen PM, Munro CS, Bradley DG, Palmer CNA, Smith FJD, **McLean** WHI* and Irvine AD* (*joint senior authorship). (2007) Comprehensive analysis of the gene encoding filaggrin uncovers prevalent and rare mutations in ichthyosis vulgaris and atopic eczema. *Nat. Genet.* **39**, 650-4 (DOI: 10.1038/ng2020).

iv. Fallon PG, Sasaki T, Sandilands A, Campbell LE, Saunders SP, Mangan NE, Callanan JJ, Kawasaki H, Shiohama A, Kubo A, Sundberg J, Presland RB, Fleckman P, Shimizu N, Kudoh J, Irvine AD, Amagai M and **McLean** WHI. A homozygous frameshift mutation in the mouse Flg gene facilitates enhanced percutaneous allergen priming. *Nat. Genet.* **41**, 602-8 (DOI: 10.1038/ng.358).

v. **Brown** SJ, Asai Y, Cordell HJ, Campbell LE, Zhao Y, Liao H, Northstone K, Henderson J, Alizadehfar R, Ben-Shoshan M, Morgan K, Roberts G, Masthoff LJ, Pasmans SG, van den Akker PC, Wijmenga C, Hourihane JO, Palmer CN, Lack G, Clarke A, Hull PR, Irvine AD, **McLean** WHI (2011) Loss-of-function variants in the filaggrin gene are a significant risk factor for peanut allergy. *J. Allergy Clin. Immunol.* **127**, 661-7 (DOI: 10.1016/j.jaci.2011.01.031).

vi. **Brown** SJ, Kroboth K, Sandilands A, Campbell LE, Pohler E, Kezic S, Cordell HJ, **McLean** WHI, Irvine AD (2012) Intragenic copy number variation within filaggrin contributes to the risk of atopic dermatitis with a dose-dependent effect. *J. Invest. Dermatol.* 132, 98-104 (DOI: 10.1038/jid.2011.342).

Patents

- **McLean** WHI and Smith FJD (2005). "Identification of loss-of-function mutations in filaggrin causing ichthyosis vulgaris and predisposing to other diseases" PCT/GB2006004707. Priority GB/15.2.05/ GBA 0525492. Date of filing 15.12.06. Date of publication 27.08.2008.
- **McLean** WHI and Smith FJD (2006). "Prevention/treatment of ichthyosis vulgaris, atopy and other disorders." PCT/GB2007000109. Priority GB/18.01.06/ GBA 0600948. Date of filing 17.01.07. Date of publication 25.12.2012.

4. Details of the impact

Change in the understanding of atopy pathogenesis has led to change in clinical practice

A significant minority (9%) of the UK population carries one or more *FLG* null alleles [1], therefore the increased risk of atopic disease affects an estimated 5.7 million people, of whom approximately 2.4 million (42%) [2] are likely to develop atopic eczema which is directly attributable to *FLG* haploinsufficiency [3,4]. An additional 10.6% of the population are homozygous for the lowest copy number variants of *FLG*, associated with an increased risk of eczema (odds ratio ~1.67) [vi]. *FLG* mutations increase the risk of disease at every stage of the so-called 'atopic march', from eczema early in life, to asthma, food allergy and later allergic rhinitis [1,2].

The paradigm shift in understanding atopy has changed the focus of clinical care to epidermal barrier function [5,6]. Since 2008, the filaggrin/atopy link has been the subject of 90 review articles [PubMed search: filaggrin AND atopic 31/10/2013]. The research has attracted six major national/international research prizes, including the American Skin Association Achievement Award 2009.

The research focus on barrier function has informed the development of therapeutic guidelines. Thus, the NICE guidelines state: "Atopic eczema often has a genetic component that leads to the breakdown of the skin barrier. This makes the skin susceptible to trigger factors, including irritants and allergens, which can make the eczema worse" (http://guidance.nice.org.uk/CG57/QuickRefGuide/pdf/English). The NICE guideline on treatment emphasises the importance of emollient use to improve skin barrier function, now a key quality statement: "Children with atopic eczema are prescribed sufficient quantities (250–500 g weekly) from a choice of unperfumed emollients for daily use." (http://publications.nice.org.uk/atopic-eczema-in-children-qs44/list-of-quality-statements). This new understanding has been conveyed in undergraduate and postgraduate teaching. Examples include the UK Advanced Paediatric Dermatology Course, a lecture at the British Association of Dermatologists' Annual meeting 2013 and the NHS educational web pages 'NHS inform' and 'NHS choices' [7].

National and international public interest has increased awareness of atopic disease

The eczema genetic discovery has led to improved public understanding of science: Irwin **McLean** and Sara **Brown** have spoken to capacity audiences at Café Science Dundee [8]; on a wider scale, they have achieved major worldwide publicity, including front-page coverage by every major newspaper in the UK, BBC News website [9] and Newsnight, ITV, Sky News and a total of >100 TV and radio interviews. Media exposure has continued with the intense interest in peanut allergy.

The message of skin barrier impairment in eczema has increased public understanding of atopic disease, which improves compliance with emollient therapy [10]. The National Eczema Society explains: "*If you have eczema ...the protective barrier is therefore not as good as it should be... skin with eczema is more liable to become red and inflamed on contact with substances that are known to irritate or cause an allergic reaction*." http://www.eczema.org/what-is-eczema. The British Association of Dermatologists' patient information leaflet on atopic eczema (http://www.bad.org.uk/site/792/default.aspx) explains the genetically-determined skin barrier defect and use of emollient.

FLG genotype is used to stratify patients for clinical care and clinical trials

The knowledge that *FLG*-null genotype is most strongly associated with persistent, severe eczema and multiple atopic co-morbidities has facilitated clinical sub-classification [2]. In patients with signs of filaggrin deficiency, specific attention may be paid to the possible development of asthma and food allergy. Asthma management in particular is optimised by early recognition in children (http://www.brit-thoracic.org.uk/guidelines/asthma-guidelines.aspx).

Clinical trials of barrier enhancement interventions are underway. The Barrier Enhancement for Eczema Prevention study (2010-11) demonstrated a 50% reduction in eczema incidence in babies receiving daily emollients, interacting with *FLG* genotype (http://www.controlled-trials.com/ISRCTN84854178). A larger study is funded by the NIHR HTA Programme: 'A randomised controlled trial to determine whether skin barrier enhancement with emollients can

prevent eczema in high risk children' in which 1282 high-risk babies will be screened for *FLG* mutations and monitored for development of eczema, asthma and hay fever (http://www.nets.nihr.ac.uk/projects/hta/126712). Collectively, these three allergic diseases rank sixth for annual expenditures among chronic health conditions in the US, with a total estimated bill of ~$24billion (http://www.epa.gov/ORD/gems/scinews_aeroallergens.htm). A randomised controlled trial of silk therapeutic clothing for the long-term management of eczema in children (http://www.hta.ac.uk/project/2984.asp) also includes our *FLG* genotype-stratified analysis.

The finding that intragenic copy number variation determines eczema risk with a dose-dependent effect [vi] indicates that an increase in functional filaggrin of only 5-10% is sufficient to significantly reduce eczema risk. This gives added impetus to the search for filaggrin up-regulation therapies which would be applicable to 33% of the population carrying low copy number [vi].

5. Sources to corroborate the impact (indicative maximum of 10 references)

Reviews in medical and scientific journals:

1. McAleer MA and Irvine AD (2011) The multifunctional role of filaggrin in allergic skin disease. *J. Allergy Clin. Immunol.* **131**, 208-91 (DOI: 10.1016/j.jaci.2012.12.668).
2. Irvine AD, McLean WHI and Leung DY (2011) Filaggrin mutations associated with skin and allergic diseases. *New Engl. J. Med*. **365**, 1315-27 (DOI: 10.1056/NEJMra1011040).

Meta-analyses of *FLG* effect:

3. Rodríguez E, Baurecht H, Herberich E, Wagenpfeil S, Brown SJ, Cordell HJ, Irvine AD and Weidinger S (2009) Meta-analysis of filaggrin polymorphisms in eczema and asthma: robust risk factors in atopic disease. *J. Allergy Clin. Immunol*. **123**, 1361-70 (DOI: 10.1016/j.jaci.2009.03.036).
4. van den Oord RA and Sheikh A (2009) Filaggrin gene defects and risk of developing allergic sensitisation and allergic disorders: systematic review and meta-analysis. *Brit. Med. J.* **339**, b2433 (DOI: 10.1136/bmj.b2433).

Reviews in science media and the cosmetic industry:

5. Ainsworth C (2011) SKIN into the breach. A focus on skin barrier disorders has opened up new thinking about how allergies kick in. *Nature* **479**, S12-14 (DOI: 10.1038/479S12a).
6. Harding CR, Aho S and Bosko CA (Unilever) (2013) Filaggrin – revisited. *Int. J. Cosmetic Science* **35**, 412-423 (DOI: 10.1111/ics.12049).

NHS educational resources:

7. http://www.nhsinform.com/health-library/articles/e/eczema-atopic/causes and http://www.nhs.uk/news /2011/03March/Pages/peanut-allergy-faulty-gene-research.aspx.

Examples of public engagement:

8. Café Science Dundee http://www.cafesciencedundee.co.uk/?p=1235 and http://www.cafesciencedundee.co.uk/?p=1019
9. BBC news has given extensive coverage to the filaggrin story including skin barrier and eczema (http://news.bbc.co.uk/1/hi/scotland/tayside_and_central/4817512.stm) and genetic risk for peanut allergy (http://www.bbc.co.uk/news/uk-scotland-tayside-central-12698727) which reached number 7 most-read on the BBC news webpage.
10. Eczema Outreach Scotland (http://eczemaoutreachscotland.org.uk/.) a support group for patients and their families, established in 2011. Dr Sara **Brown** is a medical adviser and attends educational and outreach events.