Vaccine against scabies: necessity and possibility

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SUMMARY

Scabies is an infectious disease that is endemic in poorly resourced communities, and also common in industrialized countries. Although the disease, which is caused by infestation of *Sarcoptes scabiei*, is generally mild, the need for a vaccine against *S. scabiei* is proposed. The immunological mechanisms that control *S. scabiei* infection are discussed and the current status of scabies vaccine development reviewed. Future directions for scabies vaccine development are also addressed.

Key words: Scabies, protective immune responses, vaccine.

INTRODUCTION

Scabies is an ectoparasite infection caused by the mite *Sarcoptes scabiei* variety *hominis*. The disease remains endemic in impoverished communities and in developing countries, where one report shows that up to 9% of the population and 19% of those attending a primary healthcare centre are infected (Hengge *et al*. 2006). In developed countries, outbreaks of scabies occur in hospitals, kindergartens and aged care centres (Heukelbach and Feldmeier, 2006; Mounsey and McCarthy, 2013; Mounsey *et al*. 2013; Rampton *et al*. 2013).

Ordinary scabies and crusted scabies (Norwegian scabies, or scabies crustosa) are the two main clinical manifestations of scabies. Ordinary scabies presents with an intensely pruritic skin rash, and in general mite numbers are self-limiting, in the range of 10–12 mites per patient. In a small minority of infected people however, the disease involves hyper-proliferation of mites, with thickening and depigmentation of skin; often accompanied by secondary infection with *Staphylococcus aureus* and *Streptococcus pyogenes*. In these latter situations, the disease is sometimes life threatening due to subsequent complications (Walton *et al*. 1999, 2004, 2008; Walton and Currie, 2007; Mounsey *et al*. 2010; Walton, 2010).

Ordinary scabies is generally treated with topical acaricides. For crusted scabies, crust and scale removal is necessary and an intensive treatment regimen including both oral and topical acaricides is recommended (Hengge *et al*. 2006; Mounsey and McCarthy, 2013; Mounsey *et al*. 2013).

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IS A VACCINE NEEDED FOR PREVENTING SCABIES INFECTION?

Vaccination is one of the greatest achievements against human infectious diseases. Deadly, or chronic infectious diseases that have significant impact on human society, such as those caused by polio virus or hepatitis B virus have been or are now being controlled after the introduction of vaccines (Nossal, 2003). However, comparatively, scabies only causes minor skin disease in most people (Walton *et al*. 1999, 2004, 2008; Walton and Currie, 2007; Mounsey *et al*. 2010; Walton, 2010). Is there a need for a vaccine against this disease?

Scabies is endemic among people living in low-resource communities with overcrowded housing, such as Aboriginal people living in Northern Australia and those in developing countries (Walton *et al*. 2004). Scabies is also often associated with secondary infection by *S. pyogenes*, which is a major precipitant of acute post-streptococcal glomerulonephritis, and possibly rheumatic fever (McCarthy *et al*. 2004). Although community education, disease reporting and improved drug supply can improve the control of scabies, these measures can be difficult to implement (La Vincente *et al*. 2009). An effective vaccine has the potential to protect these groups of people more efficiently, with lower economic costs. Moreover, the intensive use of acaricides leads to the development of drug resistance in scabies mites. Clinical and *in vitro* resistance of *S. scabiei* to ivermectin has been reported in human crusted scabies (Currie *et al*. 2004) and also in dogs infested with *S. scabiei* var. *canis* (Terada *et al*. 2010). Permethrin resistance has been observed in a rabbit

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model of *S. scabiei* var. *canis* (Pasay *et al.* 2006). In the event of continued emergence of acaricide resistance in human scabies, vaccine or immunotherapy may provide another way to overcome this problem.

Crusted scabies, the severe form of scabies infection, responds poorly to treatment and the death rate of crusted scabies remains high (Walton *et al.* 1999, 2008, 2010; Roberts *et al.* 2005). Even in the event of treatment, crusted scabies patients are often highly susceptible to re-infection from community contacts, and recrudescence is common. Some patients have several episodes of crusted scabies per year, requiring quarantine and hospitalization, severely impacting on quality of life. Immune suppressed patients, including patients with acquired immunodeficiency syndrome (AIDS) and organ transplant patients, tend to develop atypical scabies manifestations and crusted scabies (Orkin, 1993*b*; Taplin and Meinking, 1997). Interestingly however, a large subset of patients with crusted scabies has no known risk factor or immune deficit, and the immune mechanisms underlying the development of crusted scabies remain poorly understood (Walton, 2010; Walton *et al.* 2010). Subsequent bacterial infection can result in life-threatening conditions. As such, effective immunotherapy against early scabies infection would be very helpful to patients that tend to develop into crusted scabies.

Institutional outbreaks of scabies in childcare centres, aged care centres, prisons and hospital wards are not uncommon in developed countries. Current life expectancy in Australia reaches 80 years old for males and 84 for females. The likelihood of older people living in aged care centres will increase in future, and scabies is a common problem in aged care facilities (Paules *et al.* 1993; Makigami *et al.* 2011, 2012; Ariza *et al.* 2012). In a survey conducted in Japan to investigate risk factors of scabies in psychiatric and long-term care hospitals, 44·9% had one or more scabies cases in a year, with both patients and medical professionals affected (Makigami *et al.* 2011). Scabies tends to manifest atypically in older people, which complicates diagnosis, and some even progress to crusted scabies. Therefore, older people living in aged care centres and people working in the aged care centres would benefit from a vaccine against scabies infection, which could prevent problematic outbreaks in these settings. Similarly, more and more children are now attending childcare centres in developed countries (Churchill and Pickering, 1997). As infants and young children commonly carry the highest burden of scabies within a community (Clucas *et al.* 2008), outbreaks of scabies in childcare centres may cause significant problems to families, similar to those caused by head lice. Vaccination to children attending childcare centres or child carers may prevent the disease transmission among them.

In summary, an effective vaccine, either prophylactic or therapeutic, is needed for the control and treatment of this neglected skin disease in defined populations, and would benefit multiple groups in the community (Hengge *et al.* 2006).

PROTECTIVE IMMUNE RESPONSES IN SCABIES

A vaccine against scabies has yet to be developed, and research in this area has been extremely limited. It is clear however that the development of a scabies vaccine is unlikely to be successful without an understanding of the protective arm of immune responses against *S. scabiei* infection and modulating mechanisms utilized by the parasite to survive in human skin.

Protective immune responses develop after scabies infection. In human scabies, the immune response is usually able to control mite numbers and in some cases clear the infestation. People with a previous infestation of ordinary scabies are generally infested with fewer mites and more rapid development of symptoms after re-infection (Mellanby, 1977), suggesting that protective immune responses do develop after infestation with scabies mites (Walton *et al.* 1999, 2008, 2010). In animal studies, dogs previously infested with scabies developed protective immunity to subsequent infestation (Arlian *et al.* 1996). Goats challenged experimentally with mites were shown to be resistant to reinfestation (Tarigan and Huntley, 2005). Sheep following the secondary challenge, developed a smaller area of mange lesion than that seen following primary challenge and live *S. scabiei* mites were not detected in skin samples (Rodriguez-Cadenas *et al.* 2010*a*). However, the exact immune protective mechanisms against scabies infection have not been demonstrated.

Humoral immune responses

Data from both human and animal studies demonstrate that antibody levels are increased after *S. scabiei* infection (Roberts *et al.* 2005; Walton *et al.* 2008). In humans, some reports suggest that there are no differences in scabies-specific IgM, IgG antibody responses between ordinary scabies patients and controls, but only a limited number of antigens have been tested (Walton *et al.* 2010).

Crusted scabies patients develop stronger antibody responses than those of ordinary scabies (Salo *et al.* 1982; Walton *et al.* 2008). In one study, it was demonstrated that sera from all of the crusted scabies patients tested showed strong IgE binding to 11–21, and IgG binding to 1–7 scabies proteins. In contrast, only three of the seven patients with ordinary scabies showed IgE binding to 1–6 scabies proteins, and their antibody binding was much weaker, although the authors tested reactivity to a

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Table 1. Vaccination studies against scabies

Species	Antigen	Protection against mite challenge	Induced antibody	References
Rabbit	House dust mite extract	Partial protection	Protected animals have reduced antibody levels	(Arlian *et al*. 1995)
Canine	*Scabiei* var. *canis* (infection)	Yes	IgG1, IgG2, Ig M	(Arlian *et al*. 1996)
Rabbit	Human scabies Ssag1/Ssag 2	No	IgG,	(Harumal *et al*. 2003)
Goat	Soluble mite protein extract	No	IgG, no IgE	(Tarigan and Huntley, 2005)
Sheep	*Scabiei* var. *ovis* (infection)	Partial protection	IgG, IgE	(Rodriguez-Cadenas *et al*. 2010*b*)
Rabbit	Tropomyosin	No	IgG, low IgE	(Zhang *et al*. 2012)

var. *canis* mite extract (Arlian *et al*. 2004). Serum IgA levels usually decrease in ordinary scabies compared with controls (Salo *et al*. 1982; Walton *et al*. 2010), while in crusted scabies, IgA levels were elevated in 64% of patients (Roberts *et al*. 2005).

There are limited studies demonstrating whether antibody responses to scabies antigens are protective. In a goat model of scabies, vaccination with soluble proteins of *S. scabiei* invoked high levels of scabies-specific IgG in the serum of all animals but failed to induce specific IgE (Table 1). These immunized goats were not protected against mite challenge. In contrast, goats challenged experimentally with mites developed strong serum IgE and IgG antibody responses to scabies antigens and were shown to be resistant to reinfestation (Tarigan and Huntley, 2005). Recently, these observations have been confirmed in a similar study, where sheep were challenged with *S. scabiei* var. *ovis*. The challenged sheep developed both IgG and IgE responses to mites. Following the secondary challenge, sheep developed a smaller area of mange lesion than that seen following primary challenge and live *S. scabiei* mites were not detected in skin samples (Rodriguez-Cadenas *et al*. 2010*a*). Interestingly, it has been shown that host antibody can be detected in mite gut (Rapp *et al*. 2006).

Taken together, the above results indicate that scabies infection evokes antibody responses to *S. scabiei* antigens. *Sarcoptes scabiei*-specific IgE may serve a marker for the development of protective immunity against the mite infection, at least in the above animal studies. However, high levels of IgE are characteristic of crusted scabies, where protective immunity does not develop (Walton, 2010). Therefore, it is still not known whether antibody against selected scabies specific antigens, especially scabies-specific IgE, may have a protective role against scabies infection.

Antigen-specific CD4+T cell responses may play a critical role for the control of scabies

T cell infiltrates to scabies infected skin are observed in wombats (Skerratt, 2003), dogs (Stemmer *et al*. 1996; Arlian *et al*. 1997), pigs (Van Neste and Staquet, 1986) and humans (Reunala *et al*. 1984; Walton *et al*. 2008; Walton, 2010). CD4+T cells are found to dominate the lymphocytic infiltrate of inflammatory skin lesions in ordinary scabies (Cabrera *et al*. 1993; Arlian *et al*. 1997). In dogs infested with *S. scabiei* var. *canis*, CD4+T cells were abundant in fluctuating densities in the dermis, epidermis and follicular epidermis during the sensitizing infection and these cells became the dominant cell type early during the challenge infection. The density of CD4+T cells in the infiltrate was much greater during the challenge than during the sensitization infection (Arlian *et al*. 1997). In individuals with crusted scabies, the main skin-infiltrating T cells are CD8+T cells. The proportions of T and B lymphocytes and T cell subsets in the blood of these patients were reported within normal ranges, indicating a selective movement of CD8+T cells into the skin (Walton *et al*. 2008; Walton, 2010), which appear unable to control the infection. When infected with scabies AIDS patients often develop crusted scabies (Orkin, 1993*a*; Orkin and Maibach, 1993; Fuchs *et al*. 2007). These results argue that scabies-specific CD4+T cells may be necessary for the control of the disease and that crusted scabies may result from the deficiency of CD4+T cells in the skin; CD4+T cells may be required to reach infected skin to eradicate the parasite, through release of cytokines, or to attract other types of cells or cytokines to facilitate the killing of mites.

Th1 vs Th2 responses

It has been suggested that a Th2 biased immune response against *S. scabiei* infection may account for the development of crusted scabies (Walton, 2010). It has been demonstrated that scabies mite infection in mice promotes the production of IL4 in lymph nodes while immunization with scabies extract induces production of IFNγ, suggesting that mites develop mechanisms inhibiting Th1 responses (Lalli *et al*. 2004). In a study conducted by Arlian *et al*. (1995), when rabbits were immunized with house dust mite extract and challenged by *S. scabiei* var. *canis*,

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protected rabbits developed lower levels of antibodies than those that were not immunized and developed an increased Th1, and reduced Th2 responses. Peripheral blood mononuclear cells isolated from patients with crusted scabies secreted higher levels of IL-4, IL-5 and IL-13, and lower levels of IFNγ, compared with those of ordinary scabies patients, showing an increased allergic Th2 response to recombinant *S. scabiei* antigens (Walton *et al*. 2008, 2010). Crusted scabies patients develop stronger IgE antibody responses than those of ordinary scabies, which indicate Th2 biased immune responses in uncontrolled scabies infestation (Salo *et al*. 1982; Walton *et al*. 2008; Mounsey and McCarthy, 2013; Mounsey *et al*. 2013; Rampton *et al*. 2013). Therefore, a vaccine that elicits robust Th1 immune responses may protect hosts from infection, and may provide therapeutic effects for crusted scabies patients.

Role of innate immune cells

It is clear that *S. scabiei* proteins play a role in modulating the host skin immune responses. *Sarcoptes scabiei* mite extracts regulate skin pro-inflammatory processes (Arlian *et al*. 2003; Kato *et al*. 2005; Walton, 2010). Human PBMCs, whether from naïve or sensitized donors, when stimulated with mite extract, produce high levels of IL-10 (Arlian *et al*. 2006; Walton *et al*. 2010) which may be produced by antigen-specific T cells (Arlian *et al*. 2006), but may also come from innate immune cells. Mast cells, basophils and eosinophils are all recruited to scabies-infected skin (Skerratt, 2003; Noviana *et al*. 2004; Roberts *et al*. 2005; Walton *et al*. 2008; Walton, 2010). Recently, it was shown that mast cells and regulatory T cells contribute to each other's immune suppressive function, mediated through membrane-bound TGF-β (Blatner *et al*. 2010; Su *et al*. 2012). Therefore, the function of mast cells, basophils or eosinophils attracted into the infection site are likely influenced by the skin environment, and modulate infiltrating skin dendritic cell function to elicit either a Th1 biased or a Th2 biased immune response. These attracted mast cells, basophil or eosinophil may also influence adapted immune responses either at the priming stage, or the function of effector T cells when these cells migrate to infected skin at the effector stage. However, this hypothesis needs more thorough exploration.

CURRENT STATUS OF SCABIES VACCINE DEVELOPMENT

TickGARD and GAVAC are marketed commercial vaccines against the cattle tick *Boophilus microplus*. These vaccines are based on a *B. microplus* derived glycoprotein Bm86 that is expressed on the surface of mid-gut cells, which is not exposed to the host immune system (Odongo *et al*. 2007; Parizi *et al*. 2012*a*, *b*). Bm86 specific antibody binds to Bm86 when the tick sucks blood from immunized cattle, damages the digestive system of a tick and subsequently causes parasite death by antibody mediated killing. Since both ticks and scabies mites are ectoparasites, the success of tick vaccines is encouraging for the development of a vaccine against scabies. Although *S. scabiei* does not directly feed on blood, it feeds on serum that seeps into the burrow. Immunohistochemistry demonstrates that host immunoglobulins are present in the mite gut (Rapp *et al*. 2006; Willis *et al*. 2006). Although there is little evidence that natural antibodies play a direct role in protective immunity to scabies, vaccine-induced cellular immune responses to scabies, or antibodies directed to selected antigens are more likely to provide the ultimate protective role.

As described above, in animal studies, vaccination with soluble proteins of *S. scabiei* invoked high levels of scabies-specific IgG in the serum of all animals but failed to induce specific IgE, and immunized animals were not protected (Tarigan and Huntley, 2005). Recently, it was demonstrated that hydrophobic, but not hydrophilic antigens from *S. scabiei* reacted with sera from infected pigs (Hejduk *et al*. 2011), indicating these antigens are more immunogenic than soluble antigens that have previously been tested. Rabbits immunized with dust mite extract were resistant to infection by *S. scabiei* var. *canis*. Interestingly, resistant hosts exhibited increased cell infiltration, significantly lower scabies-specific immunoglobulin titres and produced antibody to fewer scabies mite antigens than did non-resistant hosts (Arlian *et al*. 1995). The same has been observed in naïve rabbits infested with *S. scabiei* var. *canis* with less severe clinical symptoms ('resistant individuals') (Arlian *et al*. 1994*a*, *b*). In a recent report, sera from scabies-infected patients reacted with scabies mite recombinant proteins with sequence homology to house dust mite allergens, and sera from crusted scabies patients had stronger immune responses against these allergens (Walton *et al*. 2010). The above results suggest that the host can be protected from scabies infection through vaccination; cross reactivity to antigens from dust mite may protect the host from pathological scabies infection. Finally, protection is achieved through generating immune responses to unidentified antigen/antigens.

One of the key obstacles currently preventing advances in scabies vaccine development is the knowledge and selection of protective antigen or antigens. Both exposed and/or concealed antigens of scabies may provide protective responses, if these are important proteins for parasite survival or/and pathogenesis. The construction of comprehensive *S. scabiei* cDNA libraries has enabled the identification of several candidate antigens. Recombinant

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apolipoprotein (Harumal *et al*. 2003), glutathione S transferases (Pettersson *et al*. 2005), serine proteases (Holt *et al*. 2003; Beckham *et al*. 2009), cysteine proteases (Walton *et al*. 2010) and serine protease inhibitors (Mika *et al*. 2012) have been expressed and purified *in vitro*, and their location in mites and human skin determined. However, immunogenicity of these antigens has yet to be characterized in detail. The scabies mite itself inhibits Th1 responses and mite proteins such as glutathione S transferases have immunomodulatory functions (Ouaissi *et al*. 2002; Lalli *et al*. 2004; Arlian *et al*. 2006), Their influences on the efficacy of vaccine, especially therapeutic vaccine, has to be carefully considered.

In summary, vaccination using dust mite extracts protected immunized animals from mites challenge, although whether protection is mediated by a specific protective antibody, or by induced cellular immune responses, is not clear. Protective antigens are also not known. It is therefore necessary to investigate how the immune system controls scabies, and identify the antigen or antigens that elicit protective immune responses. Increased understanding of the immune-pathogenesis of crusted scabies will also lead to the development of specific immunotherapy for this group of patients.

FUTURE DIRECTIONS: GENERATING A ROBUST PROTECTIVE IMMUNE RESPONSE AGAINST SELECTED *S. SCABIEI* ANTIGEN/ANTIGENS AND LESSONS FROM OTHER PARASITIC DISEASES

Vaccination with cysteine proteinases has been shown to be effective against a number of parasites, and in some cases the efficacy of the vaccine is also associated with the inhibition of cysteine proteinase activity by IgG from the immune sera (Nisbet and Huntley, 2006). Glutathione S-transferases also have immunomodulatory functions and seem a promising vaccine candidate in human schistosomiasis and other parasite infections including scabies (Ouaissi *et al*. 2002). The genomes of schistosomes are now available and have helped researchers to identify new antigens for schistosomiasis vaccine development (Kupferschmidt, 2013). Hence, selection of appropriate antigens may not be possible until the entire genome of scabies mite is sequenced, although preliminary sequencing has commenced (Mounsey *et al*. 2012). Other 'next generation' approaches, such as systematic and functional genomics, reverse vaccinology and selection of proper expression systems are likely required to identify the right scabies vaccine antigen candidates (Geldhof *et al*. 2007; Maritz-Olivier *et al*. 2012).

Pre-existing immune responses to selected antigen in endemic areas also need careful evaluation. Recently, a hookworm vaccine candidate, based on the protein of Ancylostoma Secreted Protein 2 of *Necator americanus* (Na-ASP-2), was suspended from phase I clinical trial, because of severe adverse events caused by pre-existing IgE antibody to the antigen before vaccination (Schneider *et al*. 2011). Pre-existing immune responses to antigen incorporated in vaccine has also been shown to inhibit T cell responses, a phenomenon called 'original antigenic sin' (Liu *et al*. 2003, 2006). As scabies infection rates in endemic areas reaches 40–80% in some high-risk groups in Africa, indigenous communities in Australia and New Zealand, the influence of pre-existing circulating antibody is a critical consideration (Heukelbach and Feldmeier, 2006).

Traditionally, a vaccine utilizes antigens plus an adjuvant to induce immune responses. Currently, the strategy has been extended to firstly optimize antigenicity of an antigen, and secondly to push the vaccine-induced immune response to higher quantity and quality by using a synergistic combination of cytokines, Toll-like receptor ligands and co-stimulatory molecules, and finally by removing the braking mechanisms mediated by regulatory T cells, NKT cells and suppressive molecules such as CTLA-4, PD-1 and TGF-β (Berzofsky, 2012). Previously, it was demonstrated that blockage of IL-10 signalling at the time of immunization generated robust Th1 and CD8 + T cell responses (Liu *et al*. 2006, 2009; Chen *et al*. 2011*a*). IL-10 signalling blockage protects the host from infection with viral (Brooks *et al*. 2006), bacterial (Pitt *et al*. 2012) and parasitic infection (Fairfax *et al*. 2012), and has therapeutic effects against tumours (Berezhnoy *et al*. 2012). It is likely that careful selection of proper antigens, together with administration of Toll-like receptor ligands and blockage of negative immune-signalling, may protect vaccinated people from scabies infection and may have therapeutic effects against crusted scabies, which is hard to manage using current modalities.

As scabies is generally not life threatening, vaccination compliance is a limiting factor if it is used to control scabies in a community. Nanotechnology provides some exciting ways in developing novel application-friendly vaccines. Recently, it was shown that oral administration of nanotechnology based vaccine, or needle-free skin vaccination, generated effective immune responses against cancer and viral infection (Chen *et al*. 2011*b*; Zhu *et al*. 2012).

Taken together, a scabies vaccine would be useful for controlling infection in both developing and developed countries. Understanding the immuno-pathogenesis of scabies and host immunological protective mechanisms, and combining new concepts and technology in vaccination will promote the generation of an effective vaccine against scabies infection.

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REFERENCES

Ariza, L., Walter, B., Worth, C., Brockmann, S., Weber, M. L. and Feldmeier, H. (2012). Investigation of a scabies outbreak in a kindergarten in Constance, Germany. *European Journal of Clinical Microbiology and Infectious Diseases* **32**, 372–380.

Arlian, L. G., Morgan, M. S., Vyszenski-Moher, D. L. and Stemmer, B. L. (1994*a*). *Sarcoptes scabiei*: the circulating antibody response and induced immunity to scabies. *Experimental Parasitology* **78**, 37–50.

Arlian, L. G., Rapp, C. M., Vyszenski-Moher, D. L. and Morgan, M. S. (1994*b*). *Sarcoptes scabiei*: histopathological changes associated with acquisition and expression of host immunity to scabies. *Experimental Parasitology* **78**, 51–63.

Arlian, L. G., Rapp, C. M. and Morgan, M. S. (1995). Resistance and immune response in scabies-infested hosts immunized with Dermatophagoides mites. *American Journal of Tropical Medicine and Hygiene* **52**, 539–545.

Arlian, L. G., Morgan, M. S., Rapp, C. M. and Vyszenski-Moher, D. L. (1996). The development of protective immunity in canine scabies. *Veterinary Parasitology* **62**, 133–142.

Arlian, L. G., Rapp, C. M., Stemmer, B. L., Morgan, M. S. and Moore, P. F. (1997). Characterization of lymphocyte subtypes in scabietic skin lesions of naive and sensitized dogs. *Veterinary Parasitology* **68**, 347–358.

Arlian, L. G., Morgan, M. S. and Neal, J. S. (2003). Modulation of cytokine expression in human keratinocytes and fibroblasts by extracts of scabies mites. *American Journal of Tropical Medicine and Hygiene* **69**, 652–656.

Arlian, L. G., Morgan, M. S., Estes, S. A., Walton, S. F., Kemp, D. J. and Currie, B. J. (2004). Circulating IgE in patients with ordinary and crusted scabies. *Journal of Medical Entomology* **41**, 74–77.

Arlian, L. G., Morgan, M. S. and Paul, C. C. (2006). Evidence that scabies mites (Acari: Sarcoptidae) influence production of interleukin-10 and the function of T-regulatory cells (Tr1) in humans. *Journal of Medical Entomology* **43**, 283–287.

Beckham, S. A., Boyd, S. E., Reynolds, S., Willis, C., Johnstone, M., Mika, A., Simerska, P., Wijeyewickrema, L. C., Smith, A. I., Kemp, D. J., Pike, R. N. and Fischer, K. (2009). Characterization of a serine protease homologous to house dust mite group 3 allergens from the scabies mite *Sarcoptes scabiei*. *Journal of Biological Chemistry* **284**, 34413–34422. doi: 10.1074/jbc.M109.061911.

Berezhnoy, A., Stewart, C. A., McNamara, J. O., II, Thiel, W., Giangrande, P., Trinchieri, G. and Gilboa, E. (2012). Isolation and optimization of murine IL-10 receptor blocking oligonucleotide aptamers using high-throughput sequencing. *Molecular Therapy* **20**, 1242–1250. doi: 10.1038/mt.2012.18.

Berzofsky, J. A. (2012). A push-pull vaccine strategy using Toll-like receptor ligands, IL-15, and blockade of negative regulation to improve the quality and quantity of T cell immune responses. *Vaccine* **30**, 4323–4327. doi: 10.1016/j.vaccine.2011.11.034.

Blatner, N. R., Bonertz, A., Beckhove, P., Cheon, E. C., Krantz, S. B., Strouch, M., Weitz, J., Koch, M., Halverson, A. L., Bentrem, D. J. and Khazaie, K. (2010). In colorectal cancer mast cells contribute to systemic regulatory T-cell dysfunction. *Proceedings of the National Academy of Sciences USA* **107**, 6430–6435. doi: 10.1073/pnas.0913683107.

Brooks, D. G., Trifilo, M. J., Edelmann, K. H., Teyton, L., McGavern, D. B. and Oldstone, M. B. (2006). Interleukin-10 determines viral clearance or persistence *in vivo*. *Nature Medicine* **12**, 1301–1309. doi: 10.1038/nm1492.

Cabrera, R., Agar, A. and Dahl, M. V. (1993). The immunology of scabies. *Seminars in Dermatology* **12**, 15–21.

Chen, J., Ni, G. and Liu, X. S. (2011*a*). Papillomavirus virus like particle-based therapeutic vaccine against human papillomavirus infection related diseases: immunological problems and future directions. *Cellular Immunology* **269**, 5–9. doi: 10.1016/j.cellimm.2011.03.003.

Chen, X., Fernando, G. J., Crichton, M. L., Flaim, C., Yukiko, S. R., Fairmaid, E. J., Corbett, H. J., Primiero, C. A., Ansaldo, A. B., Frazer, I. H., Brown, L. E. and Kendall, M. A. (2011*b*). Improving the reach of vaccines to low-resource regions, with a needle-free vaccine delivery device and long-term thermostabilization. *Journal of Controlled Release* **152**, 349–355. doi: 10.1016/j.jconrel.2011.02.026.

Churchill, R. B. and Pickering, L. K. (1997). Infection control challenges in child-care centers. *Infectious Disease Clinics of North America* **11**, 347–365.

Clucas, D. B., Carville, K. S., Connors, C., Currie, B. J., Carapetis, J. R. and Andrews, R. M. (2008). Disease burden and health-care clinic attendances for young children in remote aboriginal communities of northern Australia. *Bulletin of the World Health Organization* **86**, 275–281.

Currie, B. J., Harumal, P., McKinnon, M. and Walton, S. F. (2004). First documentation of *in vivo* and *in vitro* ivermectin resistance in *Sarcoptes scabiei*. *Clinical Infectious Diseases* **39**, e8–e12. doi: 10.1086/421776.

Fairfax, K. C., Amiel, E., King, I. L., Freitas, T. C., Mohrs, M. and Pearce, E. J. (2012). IL-10R blockade during chronic *Schistosomiasis mansoni* results in the loss of B cells from the liver and the development of severe pulmonary disease. *PLoS Pathogens* **8**, e1002490. doi: 10.1371/journal.ppat.1002490.

Fuchs, B. S., Sapadin, A. N., Phelps, R. G. and Rudikoff, D. (2007). Diagnostic dilemma: crusted scabies superimposed on psoriatic erythroderma in a patient with acquired immunodeficiency syndrome. *Skinmed* **6**, 142–144.

Geldhof, P., De Maere, V., Vercruysse, J. and Claerebout, E. (2007). Recombinant expression systems: the obstacle to helminth vaccines? *Trends in Parasitology* **23**, 527–532. doi: 10.1016/j.pt.2007.08.012.

Harumal, P., Morgan, M., Walton, S. F., Holt, D. C., Rode, J., Arlian, L. G., Currie, B. J. and Kemp, D. J. (2003). Identification of a homologue of a house dust mite allergen in a cDNA library from *Sarcoptes scabiei* var *hominis* and evaluation of its vaccine potential in a rabbit/*S. scabiei* var. *canis* model. *American Journal of Tropical Medicine and Hygiene* **68**, 54–60.

Hejduk, G., Hofstatter, K., Lowenstein, M., Peschke, R., Miller, I. and Joachim, A. (2011). Characterisation of *Sarcoptes scabiei* antigens. *Parasitology Research* **108**, 309–315. doi: 10.1007/s00436-010-2063-z.

Hengge, U. R., Currie, B. J., Jager, G., Lupi, O. and Schwartz, R. A. (2006). Scabies: a ubiquitous neglected skin disease. *Lancet Infectious Diseases* **6**, 769–779. doi: 10.1016/S1473-3099(06)70654-5.

Heukelbach, J. and Feldmeier, H. (2006). Scabies. *Lancet* **367**, 1767–1774. doi: 10.1016/S0140-6736(06)68772-2.

Holt, D. C., Fischer, K., Allen, G. E., Wilson, D., Wilson, P., Slade, R., Currie, B. J., Walton, S. F. and Kemp, D. J. (2003). Mechanisms for a novel immune evasion strategy in the scabies mite *Sarcoptes scabiei*: a multigene family of inactivated serine proteases. *Journal of Investigative Dermatology* **121**, 1419–1424. doi: 10.1046/j.1523-1747.2003.12621.x.

Kato, T., Takai, T., Mitsuishi, K., Okumura, K. and Ogawa, H. (2005). Cystatin A inhibits IL-8 production by keratinocytes stimulated with Der p 1 and Der f 1: biochemical skin barrier against mite cysteine proteases. *Journal of Allergy and Clinical Immunology* **116**, 169–176. doi: 10.1016/j.jaci.2005.03.044.

Kupferschmidt, K. (2013). A worm vaccine, coming at a snail's pace. *Science* **339**, 502–503. doi: 10.1126/science.339.6119.502.

Lalli, P. N., Morgan, M. S. and Arlian, L. G. (2004). Skewed Th1/Th2 immune response to *Sarcoptes scabiei*. *Journal of Parasitology* **90**, 711–714. doi: 10.1645/GE-214R.

La Vincente, S., Kearns, T., Connors, C., Cameron, S., Carapetis, J. and Andrews, R. (2009). Community management of endemic scabies in remote aboriginal communities of northern Australia: low treatment uptake and high ongoing acquisition. *PLoS Neglected Tropical Diseases* **3**, e444. doi: 10.1371/journal.pntd.0000444.

Liu, X. S., Xu, Y., Hardy, L., Khammanivong, V., Zhao, W., Fernando, G. J., Leggatt, G. R. and Frazer, I. H. (2003). IL-10 mediates suppression of the CD8T cell IFN-gamma response to a novel viral epitope in a primed host. *Journal of Immunology* **171**, 4765–4772.

Liu, X. S., Dyer, J., Leggatt, G. R., Fernando, G. J., Zhong, J., Thomas, R. and Frazer, I. H. (2006). Overcoming original antigenic sin to generate new CD8T cell IFN-gamma responses in an antigen-experienced host. *Journal of Immunology* **177**, 2873–2879.

Liu, X. S., Leerberg, J., MacDonald, K., Leggatt, G. R. and Frazer, I. H. (2009). IFN-gamma promotes generation of IL-10 secreting CD4 + T cells that suppress generation of CD8 responses in an antigen-experienced host. *Journal of Immunology* **183**, 51–58. doi: 10.4049/jimmunol.0802047.

Makigami, K., Ohtaki, N., Ishii, N., Tamashiro, T., Yoshida, S. and Yasumura, S. (2011). Risk factors for recurrence of scabies: a retrospective study of scabies patients in a long-term care hospital. *Journal of Dermatology* **38**, 874–879. doi: 10.1111/j.1346-8138.2011.01199.x.

Makigami, K., Ohtaki, N. and Yasumura, S. (2012). A 35-month prospective study on onset of scabies in a psychiatric hospital: discussion on patient transfer and incubation period. *Journal of Dermatology* **39**, 160–163. doi: 10.1111/j.1346-8138.2011.01324.x.

Maritz-Olivier, C., van Zyl, W. and Stutzer, C. (2012). A systematic, functional genomics, and reverse vaccinology approach to the identification of vaccine candidates in the cattle tick, *Rhipicephalus microplus*. *Ticks and Tick-borne Diseases* **3**, 179–187. doi: 10.1016/j.ttbdis.2012.01.003.

McCarthy, J. S., Kemp, D. J., Walton, S. F. and Currie, B. J. (2004). Scabies: more than just an irritation. *Postgraduate Medical Journal* **80**, 382–387. doi: 10.1136/pgmj.2003.014563.

Mellanby, K. (1977). Scabies in 1976. *Royal Society of Health Journal* **97**, **32–36**, 40.

Mika, A., Reynolds, S. L., Mohlin, F. C., Willis, C., Swe, P. M., Pickering, D. A., Halilovic, V., Wijeyewickrema, L. C., Pike, R. N., Blom, A. M., Kemp, D. J. and Fischer, K. (2012). Novel scabies mite serpins inhibit the three pathways of the human complement system. *PLoS ONE* **7**, e40489. doi: 10.1371/journal.pone.0040489.

Mounsey, K. E. and McCarthy, J. S. (2013). Treatment and control of scabies. *Current Opinion in Infectious Diseases* **26**, 133–139. doi: 10.1097/QCO.0b013e32835e1d57.

Mounsey, K. E., Pasay, C. J., Arlian, L. G., Morgan, M. S., Holt, D. C., Currie, B. J., Walton, S. F. and McCarthy, J. S. (2010). Increased transcription of glutathione S-transferases in acaricide exposed scabies mites. *Parasites and Vectors* **3**, 43. doi: 10.1186/1756-3305-3-43.

Mounsey, K. E., McCarthy, J. S. and Walton, S. F. (2012). Scratching the itch: new tools to advance understanding of scabies. *Trends in Parasitology* **29**, 35–42. doi: 10.1016/j.pt.2012.09.006.

Mounsey, K. E., McCarthy, J. S. and Walton, S. F. (2013). Scratching the itch: new tools to advance understanding of scabies. *Trends in Parasitology* **29**, 35–42. doi: 10.1016/j.pt.2012.09.006.

Nisbet, A. J. and Huntley, J. F. (2006). Progress and opportunities in the development of vaccines against mites, fleas and myiasis-causing flies of veterinary importance. *Parasite Immunology* **28**, 165–172. doi: 10.1111/j.1365-3024.2006.00803.x.

Nossal, G. J. (2003). Gates, GAVI, the glorious global funds and more: all you ever wanted to know. *Immunology and Cell Biology* **81**, 20–22. doi: 10.1046/j.0818-9641.2002.01139.x.

Noviana, D., Harjanti, D. W., Otsuka, Y. and Horii, Y. (2004). Proliferation of protease-enriched mast cells in sarcoptic skin lesions of raccoon dogs. *Journal of Comparative Pathology* **131**, 28–37. doi: 10.1016/j.jcpa.2004.01.001.

Odongo, D., Kamau, L., Skilton, R., Mwaura, S., Nitsch, C., Musoke, A., Taracha, E., Daubenberger, C. and Bishop, R. (2007). Vaccination of cattle with TickGARD induces cross-reactive antibodies binding to conserved linear peptides of Bm86 homologues in *Boophilus decoloratus*. *Vaccine* **25**, 1287–1296. doi: 10.1016/j.vaccine.2006.09.085.

Orkin, M. (1993*a*). Scabies in AIDS. *Seminars in Dermatology* **12**, 9–14.

Orkin, M. (1993*b*). Scabies in AIDS. *Seminars in Dermatology* **12**, 9–14.

Orkin, M. and Maibach, H. I. (1993). Scabies therapy – 1993. *Seminars in Dermatology* **12**, 22–25.

Ouaissi, A., Ouaissi, M. and Sereno, D. (2002). Glutathione S-transferases and related proteins from pathogenic human parasites behave as immunomodulatory factors. *Immunology Letters* **81**, 159–164.

Parizi, L. F., Githaka, N. W., Logullo, C., Konnai, S., Masuda, A., Ohashi, K. and da Silva Vaz, I., Jr. (2012*a*). The quest for a universal vaccine against ticks: cross-immunity insights. *Veterinary Journal* **194**, 158–165. doi: 10.1016/j.tvjl.2012.05.023.

Parizi, L. F., Reck, J., Jr., Oldiges, D. P., Guizzo, M. G., Seixas, A., Logullo, C., de Oliveira, P. L., Termignoni, C., Martins, J. R. and da Silva Vaz, I., Jr. (2012*b*). Multi-antigenic vaccine against the cattle tick *Rhipicephalus* (*Boophilus*) *microplus*: a field evaluation. *Vaccine* **30**, 6912–6917. doi: 10.1016/j.vaccine.2012.08.078.

Pasay, C., Walton, S., Fischer, K., Holt, D. and McCarthy, J. (2006). PCR-based assay to survey for knockdown resistance to pyrethroid acaricides in human scabies mites (*Sarcoptes scabiei* var. *hominis*). *American Journal of Tropical Medicine and Hygiene* **74**, 649–657.

Paules, S. J., Levisohn, D. and Heffron, W. (1993). Persistent scabies in nursing home patients. *Journal of Family Practice* **37**, 82–86.

Pettersson, E. U., Ljunggren, E. L., Morrison, D. A. and Mattsson, J. G. (2005). Functional analysis and localisation of a delta-class glutathione S-transferase from *Sarcoptes scabiei*. *International Journal for Parasitology* **35**, 39–48. doi: 10.1016/j.ijpara.2004.09.006.

Pitt, J. M., Stavropoulos, E., Redford, P. S., Beebe, A. M., Bancroft, G. J., Young, D. B. and O'Garra, A. (2012). Blockade of IL-10 signaling during Bacillus Calmette-Guerin vaccination enhances and sustains Th1, Th17, and innate lymphoid IFN-gamma and IL-17 responses and increases protection to *Mycobacterium tuberculosis* infection. *Journal of Immunology* **189**, 4079–4087. doi: 10.4049/jimmunol.1201061.

Rampton, M., Walton, S. F., Holt, D. C., Pasay, C., Kelly, A., Currie, B. J., McCarthy, J. S. and Mounsey, K. E. (2013). Antibody responses to *Sarcoptes scabiei* apolipoprotein in a porcine model: relevance to immunodiagnosis of recent infection. *PLoS ONE* **8**, e65354. doi: 10.1371/journal.pone.0065354.

Rapp, C. M., Morgan, M. S. and Arlian, L. G. (2006). Presence of host immunoglobulin in the gut of *Sarcoptes scabiei* (Acari: Sareoptidae). *Journal of Medical Entomology* **43**, 539–542. doi: 10.1603/0022-2585(2006)43[539:Pohiit]2.0.Co;2.

Reunala, T., Ranki, A., Rantanen, T. and Salo, O. P. (1984). Inflammatory cells in skin lesions of scabies. *Clinical and Experimental Dermatology* **9**, 70–77.

Roberts, L. J., Huffam, S. E., Walton, S. F. and Currie, B. J. (2005). Crusted scabies: clinical and immunological findings in seventy-eight patients and a review of the literature. *Journal of Infection* **50**, 375–381. doi: 10.1016/j.jinf.2004.08.033.

Rodriguez-Cadenas, F., Carbajal-Gonzalez, M. T., Fregeneda-Grandes, J. M., Aller-Gancedo, J. M. and Rojo-Vazquez, F. A. (2010*a*). Clinical evaluation and antibody responses in sheep after primary and secondary experimental challenges with the mange mite *Sarcoptes scabiei* var. *ovis*. *Veterinary Immunology and Immunopathology* **133**, 109–116. doi: 10.1016/j.vetimm.2009.07.004.

Rodriguez-Cadenas, F., Carbajal-Gonzalez, M. T., Fregeneda-Grandes, J. M., Aller-Gancedo, J. M. and Rojo-Vazquez, F. A. (2010*b*). Clinical evaluation and antibody responses in sheep after primary and secondary experimental challenges with the mange mite *Sarcoptes scabiei* var. ovis. *Veterinary Immunology and Immunopathology* **133**, 109–116. doi: 10.1016/j.vetimm.2009.07.004.

Salo, O. P., Reunala, T., Kalimo, K. and Rantanen, T. (1982). Immunoglobulin and complement deposits in the skin and circulating immune complexes in scabies. *Acta Dermato-venereologica* **62**, 73–76.

Schneider, B., Jariwala, A. R., Periago, M. V., Gazzinelli, M. F., Bose, S. N., Hotez, P. J., Diemert, D. J. and Bethony, J. M. (2011). A history of hookworm vaccine development. *Human Vaccines* **7**, 1234–1244. doi: 10.4161/hv.7.11.18443.

Skerratt, L. F. (2003). Cellular response in the dermis of common wombats (*Vombatus ursinus*) infected with *Sarcoptes scabiei* var. *wombati*. *Journal of Wildlife Diseases* **39**, 193–202.

Stemmer, B. L., Arlian, L. G., Morgan, M. S., Rapp, C. M. and Moore, P. F. (1996). Characterization of antigen presenting cells and T-cells in progressing scabietic skin lesions. *Veterinary Parasitology* **67**, 247–258.

Su, W., Fan, H., Chen, M., Wang, J., Brand, D., He, X., Quesniaux, V., Ryffel, B., Zhu, L., Liang, D. and Zheng, S. G. (2012). Induced CD4+ forkhead box protein-positive T cells inhibit mast cell function and established contact hypersensitivity through TGF-beta1. *Journal of Allergy and Clinical Immunology* **130**, 444–452. e447. doi: 10.1016/j.jaci.2012.05.011.

Taplin, D. and Meinking, T. L. (1997). Treatment of HIV-related scabies with emphasis on the efficacy of ivermectin. *Seminars in Cutaneous Medicine and Surgery* **16**, 235–240.

Tarigan, S. and Huntley, J. F. (2005). Failure to protect goats following vaccination with soluble proteins of *Sarcoptes scabiei*: evidence for a role for IgE antibody in protection. *Veterinary Parasitology* **133**, 101–109. doi: 10.1016/j.vetpar.2005.03.044.

Terada, Y., Murayama, N., Ikemura, H., Morita, T. and Nagata, M. (2010). *Sarcoptes scabiei* var. *canis* refractory to ivermectin treatment in two dogs. *Veterinary Dermatology* **21**, 608–612. doi: 10.1111/j.1365-3164.2010.00895.x.

Van Neste, D. J. and Staquet, M. J. (1986). Similar epidermal changes in hyperkeratotic scabies of humans and pigs. *American Journal of Dermatopathology* **8**, 267–273.

Walton, S. F. (2010). The immunology of susceptibility and resistance to scabies. *Parasite Immunology* **32**, 532–540. doi: 10.1111/j.1365-3024.2010.01218.x.

Walton, S. F. and Currie, B. J. (2007). Problems in diagnosing scabies, a global disease in human and animal populations. *Clinical Microbiology Reviews* **20**, 268–279. doi: 10.1128/CMR.00042-06.

Walton, S. F., McBroom, J., Mathews, J. D., Kemp, D. J. and Currie, B. J. (1999). Crusted scabies: a molecular analysis of *Sarcoptes scabiei* variety *hominis* populations from patients with repeated infestations. *Clinical Infectious Diseases* **29**, 1226–1230. doi: 10.1086/313466.

Walton, S. F., Holt, D. C., Currie, B. J. and Kemp, D. J. (2004). Scabies: new future for a neglected disease. *Advances in Parasitology* **57**, 309–376. doi: 10.1016/S0065-308X(04)57005-7.

Walton, S. F., Beroukas, D., Roberts-Thomson, P. and Currie, B. J. (2008). New insights into disease pathogenesis in crusted (Norwegian) scabies: the skin immune response in crusted scabies. *British Journal of Dermatology* **158**, 1247–1255. doi: 10.1111/j.1365-2133.2008.08541.x.

Walton, S. F., Pizzutto, S., Slender, A., Viberg, L., Holt, D., Hales, B. J., Kemp, D. J., Currie, B. J., Rolland, J. M. and O'Hehir, R. (2010). Increased allergic immune response to *Sarcoptes scabiei* antigens in crusted versus ordinary scabies. *Clinical and Vaccine Immunology* **17**, 1428–1438. doi: 10.1128/CVI.00195-10.

Willis, C., Fischer, K., Walton, S. F., Currie, B. J. and Kemp, D. J. (2006). Scabies mite inactivated serine protease paralogues are present both internally in the mite gut and externally in feces. *American Journal of Tropical Medicine and Hygiene* **75**, 683–687.

Zhang, R., Jise, Q., Zheng, W., Ren, Y., Nong, X., Wu, X., Gu, X., Wang, S., Peng, X., Lai, S. and Yang, G. (2012). Characterization and evaluation of a *Sarcoptes scabiei* allergen as a candidate vaccine. *Parasites and Vectors* **5**, 176. doi: 10.1186/1756-3305-5-176.

Zhu, Q., Talton, J., Zhang, G., Cunningham, T., Wang, Z., Waters, R. C., Kirk, J., Eppler, B., Klinman, D. M., Sui, Y., Gagnon, S., Belyakov, I. M., Mumper, R. J. and Berzofsky, J. A. (2012). Large intestine-targeted, nanoparticle-releasing oral vaccine to control genitorectal viral infection. *Nature Medicine* **18**, 1291–1296.