

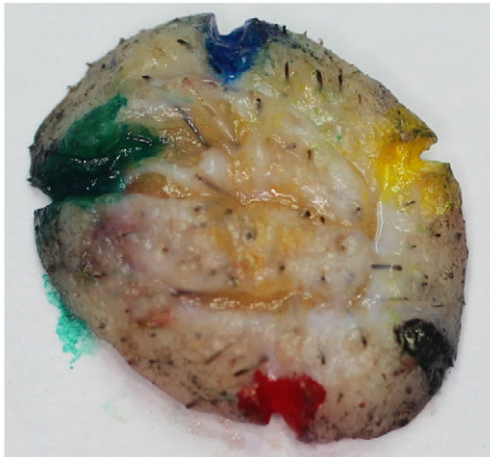


Figure 2 Tissue specimen after chromacoding. The author typically colours a one-fragment tumour as illustrated (12:00 – blue; 2:30 – yellow; 4:30 – black; 6:00 – red; 9:00 – green).

Dermoscopy ('non-mapped') has been used as an adjunctive method in MMS to demarcate preoperative margins and reduce the number of stages; however, its results are conflicting.^{8–12} One factor that may influence these results is that some studies included well-defined tumours, for which dermoscopy is 'less needed' for tumour demarcation. Furthermore, such studies probably included Mohs surgeons who were already trained and interested in dermoscopy, which increased the difficulty of evaluating the extent to which dermoscopy was helpful. It would be interesting to perform a study comparing the number of stages necessary to achieve clear margins in ill-defined tumours before and after dermoscopy training.

Mapping dermoscopic findings of ill-defined tumours treated by MMS is an important addition because it allows for a precise correlation of dermoscopy and histology findings. With repeated practice, this simple, extra preoperative step may help Mohs surgeons to correctly delineate tumour margins and hopefully reduce the number of stages, although further evaluation is needed. The main contribution of 'dermoscopy' mapping is the evaluation of dermoscopy findings when histological margins are positive for tumours ('subdermoscopic extension'). Additional studies with an extensive image database will help to identify subtle dermoscopic findings that may be missed when demarcating tumour margins.

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References

- Leibovitch I, Huilgol SC, Selva D, Richards S, Paver R. Basal cell carcinoma treated with Mohs surgery in Australia II. Outcome at 5-year follow-up. *J Am Acad Dermatol* 2005; **53**: 452–457.
- Ad Hoc Task Force, Connolly SM, Baker DR *et al*. AAD/ACMS/ASDSA/ASMS 2012 appropriate use criteria for Mohs micrographic surgery: a report of the American Academy of Dermatology, American College of Mohs Surgery, American Society for Dermatologic Surgery Association, and the American Society for Mohs Surgery. *J Am Acad Dermatol*. 2012;**67**:531–550.
- Shriner DL, McCoy DK, Goldberg DJ, Wagner Jr RF. Mohs micrographic surgery. *J Am Acad Dermatol* 1998; **39**: 79–97.
- Tolkachjov SN, Brodland DG, Coldiron BM *et al*. Understanding Mohs micrographic surgery: a review and practical guide for the nondermatologist. *Mayo Clin Proc* 2017; **92**: 1261–1271, Review.
- Silapunt S, Peterson SR, Alcalay J, Goldberg LH. Mohs tissue mapping and processing: a survey study. *Dermatol Surg* 2003; **29**: 1109–1112; discussion 1112.
- Altamura D, Menzies SW, Argenziano G *et al*. Dermatoscopy of basal cell carcinoma: morphologic variability of global and local features and accuracy of diagnosis. *J Am Acad Dermatol* 2010; **62**: 67–75.
- Carducci M, Bozzetti M, De Marco G, Foscolo AM, Betti R. Usefulness of margin detection by digital dermoscopy in the traditional surgical excision of basal cell carcinomas of the head and neck including infiltrative/morpheaform type. *J Dermatol* 2012; **39**: 326–330.
- Yeom SD, Lee SH, Ko HS *et al*. Effectiveness of dermoscopy in Mohs micrographic surgery (MMS) for nonmelanoma skin cancer (NMSC). *Int J Dermatol* 2017; **56**: e136–e139.
- Gurgen J, Gatti M. Epiluminescence microscopy (dermoscopy) versus visual inspection during Mohs microscopic surgery of infiltrative basal cell carcinoma. *Dermatol Surg* 2012; **38**(7 Pt 1): 1066–1069.
- Terushkin V, Wang SQ. Mohs surgery for basal cell carcinoma assisted by dermoscopy: report of two cases. *Dermatol Surg* 2009; **35**: 2031–2035.
- Jayasekera PSA, Dodds J, Oliphant T, Langtry JAA, Lawrence CM. Dermoscopy prior to Mohs Micrographic Surgery does not improve tumour margin assessment and lead to fewer Mohs stages. *Br J Dermatol* 2018; **178**: 565–566.
- Suzuki HS, Serafini SZ, Sato MS. Utility of dermoscopy for demarcation of surgical margins in Mohs micrographic surgery. *An Bras Dermatol* 2014; **89**: 38–43.

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Bullous pemphigoid with the deposition of IgG2 but not IgG1, IgG3 nor IgG4 autoantibodies at the basement membrane zone

Editor

Bullous pemphigoid (BP) is the most common subepidermal autoimmune blistering skin disease, characterized by tense bullae and urticarial erythema with a generalized distribution. BP is induced by autoantibodies to structural proteins of the basement membrane zone (BMZ) such as BP180 and BP230.¹ Because the majority of BP skin shows complement deposition,²

complement activation is considered to be important for blister formation. Among IgG subclasses, IgG1 have a high ability to activate complements, whereas IgG2 and IgG4 have a low and no ability, respectively. Therefore, IgG1 autoantibodies may play an important role in blister formation. Previous studies demonstrated that autoantibodies to BP180 can induce blister formation via complement-independent pathways.³ Here, we describe the first case of BP with the deposition of IgG2 but not IgG1, IgG3 nor IgG4 autoantibodies at the BMZ.

A 66-year-old woman visited our clinic with a blister on her left leg. She had been suffering from depression. Physical examination revealed a blister on her left leg without erythema (Fig. 1a). No other blisters or erosions were observed on the skin or mucous membranes. A skin biopsy specimen from the blister showed subepidermal separation associated with perivascular inflammation (Fig. 1b). The inflammatory cells consisted of lymphocytes and neutrophils, with scant eosinophils. Serum IgG antibodies to BP180 NC16A and BP230 were undetectable by ELISA. Immunoblotting using epidermal extracts from healthy skin and the patient's serum was also negative. Direct immunofluorescence studies using FITC-conjugated rabbit anti-human IgG (Dako, diluted to 1/100) revealed the deposition of IgG at the BMZ (Fig. 1c), whereas the complement (C3) was only faintly positive (Fig. 1d). Indirect immunofluorescence using 1 mol/L NaCl-split skin was positive on the epidermal side (Fig. 1e). We examined IgG subclasses of the autoantibody to

the BMZ using FITC-conjugated mouse anti-human IgG1-4 antibodies (Sigma-Aldrich, diluted to 1/100). Notably, IgG2 but not IgG1, IgG3 nor IgG4 was detected at the BMZ of perilesional skin (Fig. 2). Finally, the diagnosis of BP induced by IgG2 autoantibodies was made. We treated her with topical clobetasol propionate, which improved the skin lesion without the recurrence.

This case showed faint deposition of complement at the BMZ. Previous studies reported that 98% of BP cases show complement deposition² and that IgG autoantibodies to BP180 fail to induce blister formation in complement-deficient animals.⁴ These findings suggest that complement activation is essential in the development of BP. In contrast, a previous study reported a case of BP in a patient with C4 deficiency.⁵ Furthermore, a recent study described non-inflammatory BP cases with the deposition of IgG4 autoantibodies but not complements.⁶ Thus, the pathogenicity of IgG subclasses without the ability to activate complements has become commonly known.

Notably, only IgG2 autoantibodies were detected in our case. Previous studies showed that IgG2 autoantibodies are detected in <50% of BP patients.⁷ Although the pathogenicities of IgG1 and IgG4 autoantibodies have been experimentally shown,^{3,8} the pathogenicity of IgG2 autoantibodies has never been confirmed. It has been reported that the response to Th1 is associated with IgG1 and IgG3 subclasses, and the response to Th2 is associated with IgG4 subclass in human.⁹ Moreover, it is known that Th2-

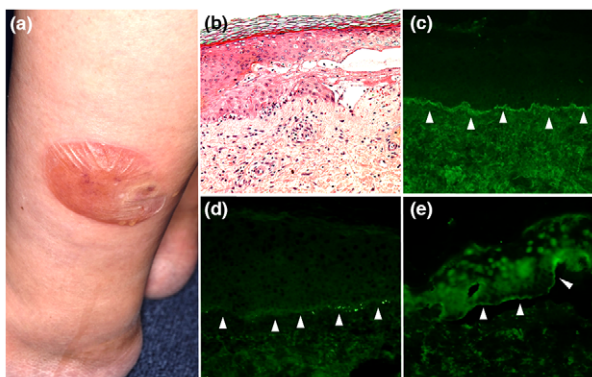


Figure 1 (a) Clinical findings of the present case. A tense blister without erythema is observed on the left leg. (b) Histopathological findings for that blister show subepidermal blister formation with infiltrations of lymphocytes and neutrophils in the dermis (haematoxylin and eosin, $\times 100$). (c) Direct immunofluorescence (DIF) for total IgG using a skin specimen from a periblistar region ($\times 200$). (Arrowheads indicate the BMZ.) (d) Direct immunofluorescence studies demonstrate the faint deposition of C3 at the BMZ ($\times 200$). (e) 1 mol/L NaCl-split human skin indirect immunofluorescence studies using the patient's serum are positive on the epidermal side ($\times 100$).

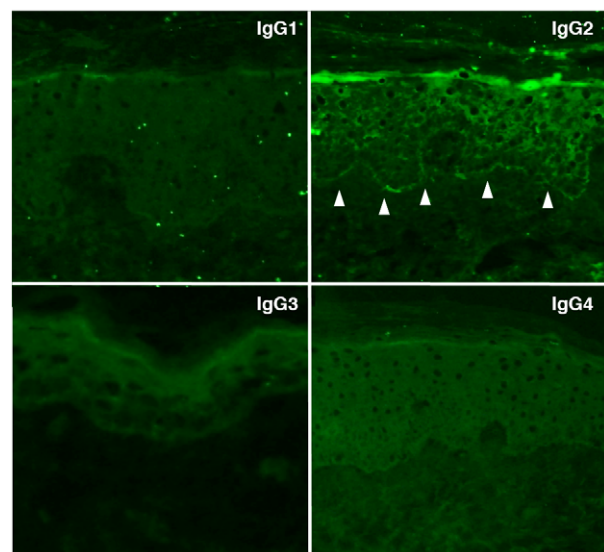


Figure 2 Direct immunofluorescence for IgG subclass antibodies reveals the linear deposition of IgG2 antibodies at the BMZ (arrowheads indicate the BMZ), but not of IgG1, IgG3, nor IgG4 ($\times 200$).

dependent IgG4 autoantibodies are preferentially seen in active stages of BP.¹⁰ Nevertheless, the relationship between IgG2 subclass and the disease phase of BP remains unclear. Despite the faint complement deposition in our case, IgG2 autoantibodies induced blister formation in association with a non-inflammatory phenotype with no erythema and little eosinophilic infiltration. These findings suggest that IgG2 autoantibodies to the BMZ have the ability to cause blister formation, possibly by complement-independent pathways. Hence, our case may give us a new insight into the pathomechanism of BP.

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References

- Schmidt E, Zillikens D. Pemphigoid disease. *Lancet* 2013; **381**: 320–332.
- Moriuchi R, Nishie W, Ujiie H, Natsuga K, Shimizu H. In vivo analysis of IgE autoantibodies in bullous pemphigoid: a study of 100 cases. *J Dermatol Sci* 2015; **78**: 21–25.
- Ujiie H, Sasaoka T, Izumi K *et al*. Bullous pemphigoid autoantibodies directly induce blister formation without complement activation. *J Immunol* 2014; **193**: 4415–4428.
- Liu Z, Guidice GJ, Swartz SJ *et al*. The role of complement in experimental bullous pemphigoid. *J Clin Invest* 1995; **95**: 1539–1544.
- Shiraishi S, Iio T, Shirakata Y *et al*. Bullous pemphigoid in a patient with a C4 deficiency. *Br J Dermatol* 1991; **124**: 296–298.
- Dainichi T, Nishie W, Yamagami Y *et al*. Bullous pemphigoid suggestive of complement-independent blister formation with anti-BP180 IgG4 autoantibodies. *Br J Dermatol* 2016; **175**: 187–190.
- Dopp R, Schmidt E, Chimanovitch I *et al*. IgG4 and IgE are the major immunoglobulins targeting the NC16A domain of BP180 in bullous pemphigoid: serum levels of these immunoglobulins reflect disease activity. *J Am Acad Dermatol* 2000; **42**: 577–583.
- Mihai S, Chiriac MT, Herrero-Gonzalez JE *et al*. IgG4 autoantibodies induce dermal-epidermal separation. *J Cell Mol Med* 2007; **5**: 1117–1128.
- Kawasaki Y, Suzuki J, Sakai N *et al*. Evaluation of T helper-1/-2 balance on the basis of IgG subclasses and serum cytokines in children with glomerulonephritis. *Am J Kidney Dis* 2004; **44**: 42–49.
- Hertl M. Humoral and cellular autoimmunity in autoimmune bullous skin disorders. *Int Arch Allergy Immunol* 2000; **122**: 91–100.

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Reply to: Kubiak K *et al*. Endosymbiosis and its significance in dermatology

Editor

We would like to begin by congratulating Kubiak K *et al*. on their work,¹ which has the potential to contribute to reviewing the question of endosymbiosis and its significance. We would

like to add some new data, notes and comments. Some data regarding viral endosymbiosis other than those discussed by the authors showed the transmission of a Gemycircularvirus – *Sclerotinia sclerotiorum* hypovirulence-associated DNA1-like virus (SsHADV-1–LV) – via insect vectors – *Lycoriella ingenua*. The DNA of SsHADV-1–LV was discovered in human samples like diarrhoeic faeces, brain, cerebrospinal fluid, serum of HIV positive and multiple sclerosis cases but also in insects: damselflies, dragonflies and mosquitoes.²

Body lice are vectors not just for *Riesia pediculicola* but also for *Acinobacter baumannii*, *Borrelia recurrentis*, *Rickettsia prowazekii* and *Bartonella quintana*. Experiments conducted with body and head lice showed an increased immunological response to *Staphylococcus aureus* and very little to *Escherichia coli*, and head lice showed better phagocytosis against *E. coli* than body lice explaining their vector activity.³

Other endosymbionts described as related to *Demodex* are as follows: (i) *Bacillus cereus*, instead of *B. oleronius*, was identified in a patient with secondary demodicosis associated with steroid-induced rosacea-like facial dermatitis (SIRLFD), but only in one positive culture.¹ (ii) *Bacillus simplex* was isolated from *Demodex folliculorum* of a single patient with primary demodicosis.⁴ (iii) In a patient with rosacea associated with *D. folliculorum*, were found positive cultures and the mass spectrometry positivity for *Bacillus pumilus*, which has haemolytic activity, cytotoxicity, produces *lecithinase* and is photolytic on casein, properties probably related with inflammation in rosacea.⁵ The difference of microbiome between demodicosis, SIRLFD and rosacea needs more and larger future studies for possible explanations of endosymbiosis between *Bacillus* species and *D. folliculorum*.⁶ Recently, *Demodex* was proposed as a delivery tool for topical targeted medications in the skin and for medications with cytotoxic properties directed to early non-melanoma skin cancer or melanoma.⁷

After reviewing the literature, we also intend to add some data, notes and comments about the authors subtitle tick-borne diseases. *Borellia garinii*, *B. burgdorferi sensu stricto* (s.s.) and *B. lusitaniae* were isolated from *Ixodes uriae*⁸ and also *B. garinii* was found related to *I. persulcatus* and *I. ricinus* in Asia and in Europe.⁸ Human disease is associated with three *Borrelia* species: *B. garinii*, *B. afzelii* (Europe) and *B. burgdorferi* s.s (North America).⁸ Climate change may affect disease spread: African trypanosomiasis, Lyme disease, malaria, tick-borne encephalitis, African trypanosomiasis, yellow fever, dengue and plague extended into wider range and higher latitude following the extension of ticks, mosquitos and midge vectors same as *Schistosoma japonicum* hosted by *Oncomelania hupensis* extended into the north of China or vectors carrying Lyme disease and West Nile virus extended into the Scandinavian Peninsula and Canada.⁹ Recent observations suggest that the historical number of tick bites (produced by *Ixodes monospinosus*, *Haemaphysalis*, *Amblyomma testudinarium*) experienced by an individual in