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Diagnosis and clinical severity markers of bullous pemphigoid

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Abstract

The use of a broad spectrum of novel detection systems for autoantibodies to the basement membrane proteins BP180 and BP230 has greatly facilitated the diagnosis of bullous pemphigoid, which most likely explains its increasing incidence in central Europe. Because the pathogenic relevance of antibodies to human BP180 has been convincingly shown both *in vitro* and *in vivo*, repeated testing for these antibodies appears to be helpful in guiding treatment decisions during the course of the disease.

Introduction and context

Bullous pemphigoid (BP) is the most frequent autoimmune bullous disorder and is clinically characterized by tense blisters and crusts on erythematous or apparently uninvolved skin. Direct immunofluorescence (IF) microscopy of a perilesional skin biopsy that typically reveals linear deposits of complement component 3 (C3) and/or immunoglobulin G (IgG) at the basal membrane zone (BMZ) is still the diagnostic gold standard. The most sensitive substrate for screening of serum autoantibodies in subepidermal blistering autoimmune diseases is 1 molar sodium chloride (1 M NaCl) split human skin. In BP, the circulating IgG autoantibodies label the epidermal side of the artificial split. These autoantibodies are directed against two hemidesmosomal proteins, BP180 and BP230 [1,2]. BP180, also known as type XVII collagen or BP antigen 2, is a transmembrane glycoprotein of about 1500 amino acids which ultrastructurally spans the lamina lucida before kinking back from the lamina densa to the lamina lucida. In contrast, BP230 is an intracellular constituent of the hemidesmosomal plaque and belongs to the plakin family of proteins. In recent years, analysis of the fine specificities of various isotypes of autoantibodies against the two target antigens has led to both better diagnostic tools and new insights into the pathogenicity of autoantibodies in BP.

Recent advances

Diagnosis of bullous pemphigoid

For more than a decade, B-cell epitopes have been known to be unequally distributed on BP180 in BP. The extracellular portion of the 16th non-collagenous (NC16A) domain was shown to be recognized by about 80% of BP sera [3]. It then became obvious that the majority of BP patients also raise antibodies against epitopes outside the NC16A domain [4–9]. Interestingly, IgG reactivity with C-terminal epitopes appeared to be associated with the presence of both skin and mucosal lesions [6–8] whereas the intracellular domain was preferentially targeted at an early clinical stage [8]. Testing for anti-BP180 antibodies outside the NC16A domain therefore is required in those patients unreactive with the immunodominant domain and has been included in our routine work-up. Most recently, the extent of phosphorylation of BP180 was shown to be important for binding of antibodies to its ectodomain [10]. Because antibodies raised against the murine homolog to the NC16A domain, expressed in *Escherichia coli*, induce subepidermal blisters when injected into neonatal mice, the relevance of phosphoepitopes on BP180 needs to be determined.

The other major target antigen in BP, BP230, is recognized in between 60 and 70% of patients by enzyme-linked immunosorbent assay (ELISA)

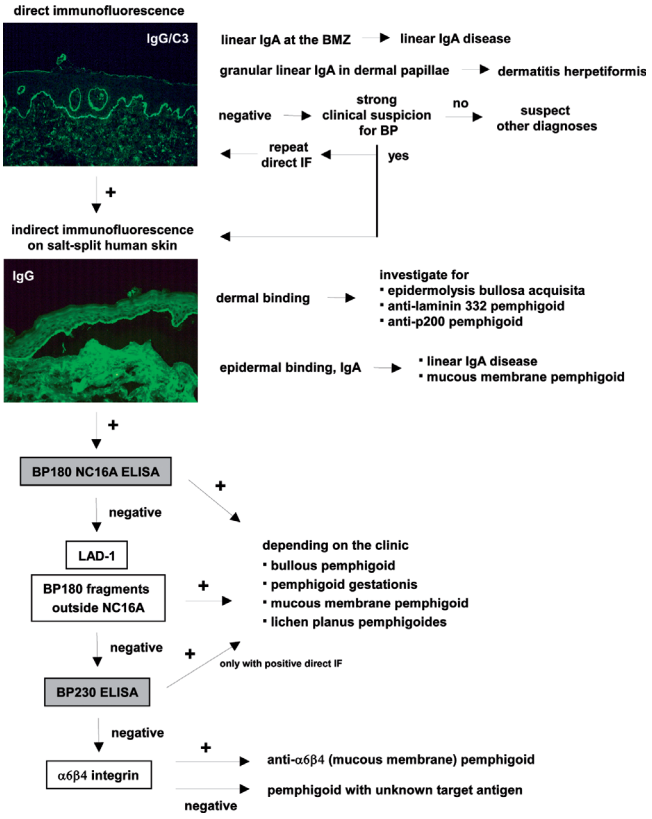
[4,9,11,12]. As for BP180, B-cell epitopes are not equally distributed but preferentially localize to the globular C-terminal domain of BP230 that mediates interaction with keratin filaments and is targeted by about 80% of BP230-reactive BP sera [9,13,14]. Interestingly, reactivity to BP230 was predominantly found in patients with limited BP, and only less than 5% of BP sera exclusively recognized BP230 [4]. By the combined use of detection systems for anti-BP180 and anti-BP230 antibodies, all 49 BP sera were reactive in a recent multicenter prospective study [9].

The novel diagnostic assays have greatly facilitated the diagnosis of BP (Figure 1), which may explain the increased incidence of BP that was recently observed in a prospective study with patients from a well-defined region, Lower Franconia in Germany. Compared with a study of the same region 10 years earlier, the incidence of BP has increased from 6.6 to 13.4 new cases per million per year [15,16]. An even higher incidence (including all subepidermal bullous diseases) of 43 new cases per million per year, which has increased almost fivefold within the last 10 years, was recently reported in the UK [17]. The data, however, were retrieved from a database for general practitioners and have been challenged by the report that in St. John's Institute of Dermatology, London, the diagnosis of BP, based on direct IF microscopy, has not increased within the last decade [18].

Whereas IgG reactivity to BP180 has been studied intensively, the importance of BP180-specific antibodies of the IgA and IgE isotypes has been highlighted only recently. Indeed, the majority of BP sera contain IgA antibodies to BP180 and most sera of patients with linear IgA disease raise IgG anti-BP180 antibodies in addition to IgA reactivity [19]. There appears to be a spectrum of the autoimmune response against BP180, ranging from pure IgG to pure IgA reactivity, with mixtures in between. This hypothesis is supported by the observation in Uganda that younger patients with BP/linear IgA disease were more likely to develop IgA anti-BMZ antibodies whereas older patients tended to raise IgG autoantibodies [20]. IgE reactivity against BP180 has been reported in 30 to 95% of BP patients [21–25]. The majority of sera contain IgE reactivity to the NC16A domain, and IgE antibodies to the intracellular domain of BP180 have also been detected recently [23,25].

In addition to reactivity against BP180 and BP230, antibodies to the α6 chain of α6β4 integrin and laminin 332, also known as laminin 5, have been reported in 50% and 40% of BP patients, respectively [26,27]. The relevance of these findings is elusive and both findings await confirmation. A more recent report demonstrates that the detection of antibodies to laminin 332 is rather uncommon in BP [28].

Figure 1. Diagnostic pathway in bullous pemphigoid (BP)



Direct immunofluorescence (IF) microscopy of a perilesional biopsy is the gold standard for the diagnosis of BP and differentiates subepidermal blistering autoimmune diseases from pemphigus. By indirect IF microscopy on 1 M NaCl spilt human skin, BP patients' sera are screened for anti-basal membrane zone (BMZ) autoantibodies. Whereas sera from patients with epidermolysis bullosa acquisita, anti-laminin 332 mucous membrane pemphigoid, and anti-p200 pemphigoid label the dermal side of the artificial split, sera of BP patients bind to the blister roof. Anti-BP180 antibodies can be detected by BP180 NC16A-specific enzyme-linked immunosorbent assay (ELISA), Western blotting with conditioned concentrated medium of cultured HaCaT cells, which detects reactivity against LAD-1 (linear IgA disease antigen 1) that corresponds to the cell-derived ectodomain of BP180, and Western blotting with various other recombinant fragments of BP180. Since four different entities are associated with IgG antibodies to BP180, the clinical phenotype determines the final diagnosis. When no BP180 reactivity is found, sera are assayed for BP230-specific antibodies that, only in conjunction with a positive direct IF microscopy and compatible clinical features support the diagnosis of BP. In case of epidermal binding by indirect IF microscopy and failure to detect IgG reactivity to both BP180 and BP230, testing for antibodies against α6β4 integrin is recommended (for example, by Western blotting of keratinocyte extract) [44]. The shaded boxes represent commercially available test systems. NC16A, the extracellular portion of the 16th noncollagenous (domain).

Clinical severity markers of bullous pemphigoid

The first evidence for a pathogenic role of human antibodies to human BP180 came from the observation that levels of circulating antibodies to the NC16A domain correlate with disease activity in BP patients [29]. This notion has been corroborated in different patient populations within recent years [30–34]. Thus, the NC16A domain appears to be an important target for monitoring circulating autoantibody levels and for guiding treatment decisions; for example, the reduction of the corticosteroid dose during the course of the disease. By the use of an NC16A tetramer, a second commercially available ELISA that is a highly sensitive and specific tool for the detection of serum antibodies in BP has been established [35]. Autoantibody levels to a C-terminal stretch of BP180 have subsequently been shown to parallel clinical activity in BP [9].

The pathogenic relevance of human antibodies to human BP180 had previously been suggested *in vitro* when the incubation of cultured human keratinocytes and cryosections of human skin with BP IgG resulted in increased mRNA levels followed by the release of proinflammatory cytokines or dermal-epidermal separation, respectively [36,37]. Most recently, a decrease of cell-surface BP180 that led to weakened cell attachment has been observed in response to the incubation of cultured human keratinocytes with anti-BP180 antibodies [38]. In addition, *in vivo* data have been provided by Nishie *et al*. [39], who induced BP-like skin lesions by injection of IgG from BP patients into Col17 knockout mice rescued by the human ortholog.

The importance of IgE autoantibodies to BP180 NC16A has been highlighted by demonstrating a correlation between clinical activity and IgE NC16A-specific circulating antibodies that were detected in 60% of BP patients [21]. Subsequently, IgE antibodies directed against this domain were linked to both a greater extent of skin lesions and a longer time period until remission was induced [23]. These observations have been substantiated in two passive-transfer mouse models. The injection of BP IgE or an IgE-producing hybridoma against the BP180 ectodomain into human skin grafted on to athymic nude mice or SCID (severe combined immunodeficiency disease) mice, respectively, resulted in an erythematous itchy plaque, which showed dermal-epidermal separation by histopathology [40,41]. Whether measurement of IgE anti-BP180 NC16A antibodies will be included in the routine analysis of BP sera needs to be addressed in future studies.

Whereas the pathogenic relevance of antibodies to BP180 is well established, the role of anti-BP230 reactivity is less clear. Serum levels of anti-BP230 antibodies repeatedly failed to parallel disease activity in BP patients [4,9,12]. In addition, experimental evidence for the pathogenicity of anti-BP230 antibodies is inconclusive and needs further exploration.

Implications for clinical practice

Direct IF microscopy of a perilesional biopsy is still regarded as the gold standard in the diagnosis of BP. A large body of evidence, however, has been collected supporting the notion that, in a large majority of BP patients, the disease can be diagnosed serologically by the detection of BP180 NC16A-specific autoantibodies. Rarely, anti-BP180 antibodies have also been found in patients with pruritic disorders, diabetes mellitus, or dementia, even when there have been no associated clinical signs of BP and direct IF microscopy has been negative [42,43]. Usually, however, in patients with clinical lesions typical for BP, the detection of antibodies to BP180 NC16A suffices to establish the diagnosis of BP, whereas serum antibodies to BP230 require a positive direct IF microscopy to diagnose BP. Finally, based on our personal experience, repeated testing for serum anti-BP180 NC16A antibodies during the course of the disease may be a helpful guide for treatment decisions.

Abbreviations

BMZ, basal membrane zone; BP, bullous pemphigoid; C3, complement component 3; ELISA, enzyme-linked immunosorbent assay; IF, immunofluorescence; IgG, immunoglobulin G; NC16A, the extracellular portion of the 16th noncollagenous (domain).

Competing interests

The authors run a non-profit laboratory that is integrated into the Department of Dermatology at the University of Lübeck. This laboratory specializes in the diagnosis of autoimmune bullous diseases and receives sera from all over the world. In this laboratory, the test systems discussed in this report are applied.

References

1. Stanley JR, Hawley-Nelson P, Yuspa SH, Shevach EM, Katz SI: **Characterization of bullous pemphigoid antigen: a unique basement membrane protein of stratified squamous epithelia.** *Cell* 1981, **24:**897-903.
2. Labib RS, Anhalt GJ, Patel HP, Mutasim DF, Diaz LA: **Molecular heterogeneity of the bullous pemphigoid antigens as detected by immunoblotting.** *J Immunol* 1986, **136:**1231-5.
3. Zillikens D, Rose PA, Balding SD, Liu Z, Olague-Marchan M, Diaz LA, Giudice GJ: **Tight clustering of extracellular BP180 epitopes recognized by bullous pemphigoid autoantibodies.** *J Invest Dermatol* 1997, **109:**573-9.
4. Thoma-Uszynski S, Uter W, Schwietzke S, Hofmann SC, Hunziker T, Bernard P, Treudler R, Zouboulis CC, Schuler G, Borradori L,

Hertl M: **BP230- and BP180-specific auto-antibodies in bullous pemphigoid.** *J Invest Dermatol* 2004, **122:**1413-22.

5. Perriard J, Jaunin F, Favre B, Büdinger L, Hertl M, Saurat JH, Borradori L: **IgG autoantibodies from bullous pemphigoid (BP) patients bind antigenic sites on both the extracellular and the intracellular domains of the BP antigen 180.** *J Invest Dermatol* 1999, **112:**141-7.
6. Mariotti F, Grosso F, Terracina M, Ruffelli M, Cordiali-Fei P, Sera F, Zambruno G, Mastrogiacomo A, Di Zenzo G: **Development of a novel ELISA system for detection of anti-BP180 IgG and characterization of autoantibody profile in bullous pemphigoid patients.** *Br J Dermatol* 2004, **151:**1004-10.
7. Hofmann S, Thoma-Uszynski S, Hunziker T, Bernard P, Koebnick C, Stauber A, Schuler G, Borradori L, Hertl M: **Severity and phenotype of bullous pemphigoid relate to autoantibody profile against the NH_2- and COOH-terminal regions of the BP180 ectodomain.** *J Invest Dermatol* 2002, **119:**1065-73.
8. Di Zenzo G, Grosso F, Terracina M, Mariotti F, De Pità O, Owaribe K, Mastrogiacomo A, Sera F, Borradori L, Zambruno G: **Characterization of the anti-BP180 autoantibody reactivity profile and epitope mapping in bullous pemphigoid patients.** *J Invest Dermatol* 2004, **122:**103-10.
9. Di Zenzo G, Thoma-Uszynski S, Fontao L, Calabresi V, Hofmann SC, Hellmark T, Sebbag N, Pedicelli C, Sera F, Lacour JP, Wieslander J, Bruckner-Tuderman L, Borradori L, Zambruno G, Hertl M: **Multicenter prospective study of the humoral autoimmune response in bullous pemphigoid.** *Clin Immunol* 2008, **128:**415-26.
10. Zimina EP, Hofmann SC, Fritsch A, Kern JS, Sitaru C, Bruckner-Tuderman L: **Bullous pemphigoid autoantibodies preferentially recognize phosphoepitopes in collagen XVII.** *J Invest Dermatol* 2008, **128:**2736-9.
11. Yoshida M, Hamada T, Amagai M, Hashimoto K, Uehara R, Yamaguchi K, Imamura K, Okamoto E, Yasumoto S, Hashimoto T: **Enzyme-linked immunosorbent assay using bacterial recombinant proteins of human BP230 as a diagnostic tool for bullous pemphigoid.** *J Dermatol Sci* 2006, **41:**21-30.
12. Kromminga A, Sitaru C, Hagel C, Herzog S, Zillikens D: **Development of an ELISA for the detection of autoantibodies to BP230.** *Clin Immunol* 2004, **111:**146-52.
13. Hamada T, Nagata Y, Tomita M, Salmhofer W, Hashimoto T: **Bullous pemphigoid sera react specifically with various domains of BP230, most frequently with C-terminal domain, by immunoblot analyses using bacterial recombinant proteins covering the entire molecule.** *Exp Dermatol* 2001, **10:**256-63.
14. Skaria M, Jaunin F, Hunziker T, Riou S, Schumann H, Bruckner-Tuderman L, Hertl M, Bernard P, Saurat JH, Favre B, Borradori L: **IgG autoantibodies from bullous pemphigoid patients recognize multiple antigenic reactive sites located predominantly within the B and C subdomains of the COOH-terminus of BP230.** *J Invest Dermatol* 2000, **114:**998-1004.
15. Bertram F, Bröcker EB, Zillikens D, Schmidt E: **Prospective analysis of the incidence of autoimmune bullous disorders in Lower Franconia, Germany [in German].** *J Dtsch Dermatol Ges* 2009, Jan 19. [Epub ahead of print].
16. Zillikens D, Wever S, Roth A, Weidenthaler-Barth B, Hashimoto T, Brocker EB: **Incidence of autoimmune subepidermal blistering dermatoses in a region of central Germany.** *Arch Dermatol* 1995, **131:**957-8.
17. Langan SM, Smeeth L, Hubbard R, Fleming KM, Smith CJ, West J: **Bullous pemphigoid and pemphigus vulgaris–incidence and mortality in the UK: population based cohort study.** *BMJ* 2008,, **337:**a180.

F1000 Factor 3.0 *Recommended*
Evaluated by Detlef Zillikens with Enno Schmidt 6 Aug 2008

18. Groves RW, Bhogal B, Taghipour K, Caposciutti P, Saha M: **Bullous pemphigoid: is the incidence of pemphigoid really increasing?** *BMJ* 2008, **337:**a1138.
19. Kromminga A, Scheckenbach C, Georgi M, Hagel C, Arndt R, Christophers E, Bröcker EB, Zillikens D: **Patients with bullous pemphigoid and linear IgA disease show a dual IgA and IgG autoimmune response to BP180.** *J Autoimmun* 2000, **15:**293-300.
20. Mulyowa GK, Jaeger G, Kabakyenga J, Brocker EB, Zillikens D, Schmidt E: **Autoimmune subepidermal blistering diseases in Uganda: correlation of autoantibody class with age of patients.** *Int J Dermatol* 2006, **45:**1047-52.
21. Dopp R, Schmidt E, Chimanovitch I, Leverkus M, Brocker EB, Zillikens D: **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:**577-83.
22. Ishiura N, Fujimoto M, Watanabe R, Nakashima H, Kuwano Y, Yazawa N, Echigo T, Okochi H, Tamaki K: **Serum levels of IgE anti-BP180 and anti-BP230 autoantibodies in patients with bullous pemphigoid.** *J Dermatol Sci* 2008, **49:**153-61.
23. Iwata Y, Komura K, Kodera M, Usuda T, Yokoyama Y, Hara T, Muroi E, Ogawa F, Takenaka M, Sato S: **Correlation of IgE autoantibody to BP180 with a severe form of bullous pemphigoid.** *Arch Dermatol* 2008, **144:**41-8.

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F1000 Factor 3.2 *Recommended*
Evaluated by Akira Ishiko 4 Feb 2008, Cassian Sitaru 15 May 2008

24. Fairley JA, Fu CL, Giudice GJ: **Mapping the binding sites of anti-BP180 immunoglobulin E autoantibodies in bullous pemphigoid.** *J Invest Dermatol* 2005, **125:**467-72.
25. Dresow SK, Sitaru C, Recke A, Oostingh GJ, Zillikens D, Gibbs BF: **IgE autoantibodies against the intracellular domain of BP180.** *Br J Dermatol* 2008, Sep 19. [Epub ahead of print].
26. Kiss M, Perényi A, Marczinovits I, Molnár J, Dobozy A, Kemény L, Husz S: **Autoantibodies to human alpha6 integrin in patients with bullous pemphigoid.** *Ann N Y Acad Sci* 2005, **1051:**104-10.
27. Bekou V, Thoma-Uszynski S, Wendler O, Uter W, Schwietzke S, Hunziker T, Zouboulis CC, Schuler G, Sorokin L, Hertl M: **Detection of laminin 5-specific auto-antibodies in mucous membrane and bullous pemphigoid sera by ELISA.** *J Invest Dermatol* 2005, **124:**732-40.
28. Lazarova Z, Salato VK, Lanschuetzer CM, Janson M, Fairley JA, Yancey KB: **IgG anti-laminin-332 autoantibodies are present in a subset of patients with mucous membrane, but not bullous, pemphigoid.** *J Am Acad Dermatol* 2008, **58:**951-8.

F1000 Factor 3.0 *Recommended*
Evaluated by Detlef Zillikens 2 May 2008

29. Schmidt E, Obe K, Brocker EB, Zillikens D: **Serum levels of autoantibodies to BP180 correlate with disease activity in patients with bullous pemphigoid.** *Arch Dermatol* 2000, **136:**174-8.
30. Feng S, Wu Q, Jin P, Lin L, Zhou W, Sang H, Shao C: **Serum levels of autoantibodies to BP180 correlate with disease activity in patients with bullous pemphigoid.** *Int J Dermatol* 2008, **47:**225-8.
31. Amo Y, Ohkawa T, Tatsuta M, Hamada Y, Fujimura T, Katsuoka K, Hashimoto T: **Clinical significance of enzyme-linked immunosorbent assay for the detection of circulating anti-BP180 autoantibodies in patients with bullous pemphigoid.** *J Dermatol Sci* 2001, **26:**14-8.
32. Tsuji-Abe Y, Akiyama M, Yamanaka Y, Kikuchi T, Sato-Matsumura KC, Shimizu H: **Correlation of clinical severity and ELISA indices for the NC16A domain of BP180 measured using BP180 ELISA kit in bullous pemphigoid.** *J Dermatol Sci* 2005, **37:**145-9.
33. Kobayashi M, Amagai M, Kuroda-Kinoshita K, Hashimoto T, Shirakata Y, Hashimoto K, Nishikawa T: **BP180 ELISA using bacterial recombinant NC16a protein as a diagnostic and monitoring tool for bullous pemphigoid.** *J Dermatol Sci* 2002, **30:**224-32.
34. Izumi T, Ichiki Y, Esaki C, Kitajima Y: **Monitoring of ELISA for anti-BP180 antibodies: clinical and therapeutic analysis of steroid-treated patients with bullous pemphigoid.** *J Dermatol* 2004, **31:**383-91.
35. Sitaru C, Dähnrich C, Probst C, Komorowski L, Blöcker I, Schmidt E, Schlumberger W, Rose C, Stöcker W, Zillikens D: **Enzyme-linked**

immunosorbent assay using multimers of the 16th non-collagenous domain of the BP180 antigen for sensitive and specific detection of pemphigoid autoantibodies. *Exp Dermatol* 2007, **16:**770-7.

36. Schmidt E, Reimer S, Kruse N, Jainta S, Bröcker EB, Marinkovich MP, Giudice GJ, Zillikens D: **Autoantibodies to BP180 associated with bullous pemphigoid release interleukin-6 and interleukin-8 from cultured human keratinocytes.** *J Invest Dermatol* 2000, **115:**842-8.

37. Herrero-González JE, Brauns O, Egner R, Rönspeck W, Mascaró JM Jr., Jonkman MF, Zillikens D, Sitaru C: **Immunoadsorption against two distinct epitopes on human type XVII collagen abolishes dermal-epidermal separation induced *in vitro* by autoantibodies from pemphigoid gestationis patients.** *Eur J Immunol* 2006, **36:**1039-48.

38. Iwata H, Kamio N, Aoyama Y, Yamamoto Y, Hirako Y, Owaribe K, Kitajima Y: **IgG from Patients with Bullous Pemphigoid Depletes Cultured Keratinocytes of the 180-kDa Bullous Pemphigoid Antigen (Type XVII Collagen) and Weakens Cell Attachment.** *J Invest Dermatol* 2009, Jan 29. [Epub ahead of print].

39. Nishie W, Sawamura D, Goto M, Ito K, Shibaki A, McMillan JR, Sakai K, Nakamura H, Olasz E, Yancey KB, Akiyama M, Shimizu H: **Humanization of autoantigen.** *Nat Med* 2007, **13:**378-83.

F1000 Factor 8.3 *Exceptional*
Evaluated by Aimee Payne 16 Apr 2007, Cassian Sitaru 24 Apr 2007, Takashi Hashimoto 30 May 2007, Stefan Martin 29 Aug 2007

40. Zone JJ, Taylor T, Hull C, Schmidt L, Meyer L: **IgE basement membrane zone antibodies induce eosinophil infiltration and histological blisters in engrafted human skin on SCID mice.** *J Invest Dermatol* 2007, **127:**1167-74.

F1000 Factor 4.8 *Must Read*
Evaluated by Cassian Sitaru 8 Aug 2007, Takashi Hashimoto 20 Jun 2008

41. Fairley JA, Burnett CT, Fu CL, Larson DL, Fleming MG, Giudice GJ: **A pathogenic role for IgE in autoimmunity: bullous pemphigoid IgE reproduces the early phase of lesion development in human skin grafted to nu/nu mice.** *J Invest Dermatol* 2007, **127:**2605-11.

F1000 Factor 4.8 *Must Read*
Evaluated by Yumi Aoyama 28 Aug 2007, Stefan Martin 12 Sep 2007

42. Jedlickova H, Racovska J, Niedermeier A, Feit J, Hertl M: **Anti-basement membrane zone antibodies in elderly patients with pruritic disorders and diabetes mellitus.** *Eur J Dermatol* 2008, **18:**534-8.

43. Foureur N, Mignot S, Senet P, Verpillat P, Picard-Dahan C, Crickx B, Labarre C, Nicaise-Roland P, Descamps V: **Correlation between the presence of type-2 anti-pemphigoid antibodies and dementia in elderly subjects with no clinical signs of pemphigoid [in French].** *Ann Dermatol Venereol* 2006, **133:**439-43.

44. Leverkus M, Bhol K, Hirako Y, Pas H, Sitaru C, Baier G, Bröcker EB, Jonkman MF, Ahmed AR, Zillikens D: **Cicatricial pemphigoid with circulating autoantibodies to beta4 integrin, bullous pemphigoid 180 and bullous pemphigoid 230.** *Br J Dermatol* 2001, **145:**998-1004.