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Detection And Characterization Of Igg, Ige And Iga Autoantibodies In Patients With Bullous Pemphigoid Associated With Dipeptidyl Peptidase-Iv Inhibitors

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**DETECTION AND CHARACTERIZATION OF IGG, IGE AND IGA AUTOANTIBODIES IN
PATIENTS WITH BULLOUS PEMPHIGOID ASSOCIATED WITH DIPEPTIDYL
PEPTIDASE-IV INHIBITORS**

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BP180, BP230

Research letter

Bullous pemphigoid (BP) is a rare autoimmune subepidermal blistering disease characterized mainly by autoantibodies against two hemidesmosomal proteins, BP180 and BP230. Recent reports suggest that dipeptidyl peptidase (DPP)-IV inhibitors, administered for type-2 diabetes mellitus, could be involved in the pathogenesis of BP. IgG is the most common autoantibody isotype detected in BP, but also IgE against BP180 and BP230 have been identified. Some authors have recently demonstrated IgG against the mid-portion of BP180 in the BP patient's serum induced by DPP-IV inhibitors.^{1,2} We report five cases of BP associated with DPP-IV inhibitors for which we characterized IgG, IgE and IgA antibodies against the two hemidesmosomal proteins by ELISA and Western Blot (WB). The BP cases were admitted at IDI-IRCCS between August 2016 and February 2017. Relationships between drugs and BP were assessed using the Naranjo algorithm, and severity of BP using the ABSIS score.^{3,4} Two cases were associated to linagliptin, one to sitagliptin, one to vildagliptin and metformin, and another to alogliptin and metformin (**Table 1**). Circulating IgG against BP180 NC16A were detected in all cases. IgG autoantibodies against BP230 were found in 4/5 sera (**Table 2**). Circulating IgG binding to epitopes mapped in the mid-portion (AA 1080-1107) and C-terminus (AA 1331-1404) of BP180 were positive by ELISA in 2/4 and 3/4 patients, respectively. All 4 sera tested contained IgG that bound BP180 and the ectodomain LAD1 on WB (**Table 2**). Three of the four patients had IgE against BP230, whereas only 1/4 bound the BP180. No IgA reactivity was detected to BP180 and BP230 by ELISA and LAD1 by WB in any patient. Of note, in 3 of 4 patients, we detected complement deposition (C3) by complement-binding IIF (**Table 2**), suggesting a role for complement in the pathogenesis of BP. In patient 3, the reactivity against BP180 decreased in parallel with disease severity, whereas BP230 reactivity remained elevated. In the same patient, no reactivity to mid-portion and C-terminus epitopes was found during the course of the disease; as for IgE, the reactivity started with BP230 and after two months spread to a weak reactivity to BP180 (data not shown).

In our study, epitope mapping showed that IgG reacted with multiple epitopes distributed over BP180 antigen, in addition to NC16A. A group of BP patients associated with DPP-IV inhibitors have been characterized for both IgE and IgA autoimmune response.

Although several studies have demonstrated a pathogenic role for anti-BP180 IgE in BP,⁵ in these patients the IgE humoral response, mainly directed against an intracellular antigen (BP230), could assume a secondary role in the disease onset. Interestingly, the IgG humoral response is directed against multiple epitopes of BP180, not mainly against the BP180 mid-portion as reported.^{1,2} In contrast to the patients in Izumi's report,¹ all patients presented with inflamed phenotypes. Thus, the typical NC16A reactivity and the classical inflamed phenotype reported in our patients suggest that the pathogenesis in DPP-IV inhibitors-induced BP have several overlapping features with classical BP. Further studies are needed to clarify the pathogenesis of DPP-IV inhibitor-induced disease.

Abbreviation: BP: bullous pemphigoid, DPP-IV inhibitors: dipeptidyl peptidase-IV inhibitors, AA: amino acid; ELISA: enzyme linked immunosorbent assay; WB: Western Blot.

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Table 1: Clinical characteristics and histology of bullous pemphigoid cases associated with DPP-IV inhibitor drugs.

Table 2: Immunologic findings in bullous pemphigoid cases associated with DPP-IV inhibitor drugs

Table 1 legend: DIF: direct immunofluorescence; BMZ: basal membrane zone; T2DM: Type 2 Diabetes mellitus; Y: Yes; N: No; M: Male; F: Female; ADR: adverse drug reaction; * Resumption of the drug when the patient has previously interrupted this therapy; ** Clinical degree of inflammation observed in patient lesional skin. ABSIS: Autoimmune Bullous Skin Disorder Intensity Score.

Table 2 legend:

IIF: Indirect immunofluorescence; normal skin: skin from normal healthy individual; sss: salt split-skin, positive staining is higher than 1:20; Com: complement fixing IIF on normal skin; Kc: keratinocyte; pos: positive; neg: negative; ND: not determined; 1080: a region encompassing amino acid residues 1080-1107; 1331: a region encompassing amino acid residues 1331-1404

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CASE	SEX	AGE	DPP-IV INHIBITORS	DELAY BEFORE ONSET OF PRURITUS (Months)	DELAY BEFORE BP DIAGNOSIS (Months)	RESUMPTION OF THE DPP- IV INHIBITORS*	CO-MEDICATIONS	HISTOLOGY, DIF AND DEGREE OF INFLAMMATION**	LEVEL OF CAUSALITY	A B S I S	A L I V E
1	M	62	LINAGLIPTIN	6	9	Y	INSULIN BROMAZEPAM CALCITRIOL FEBUXOSTAT AMLODIPINE FUROSEMIDE LANSOPRAZOLE	HISTOLOGY: DETACHMENT OF BMZ; DIF: LINEAR IGG AND C3 ON BMZ; SEVERE	PROBABLE ADR	8 0 · 5	Y
2	M	77	LINAGLIPTIN	1	3	N	INSULIN OMEPRAZOLE FUROSEMIDE BISOPROLOL CLOPIDOGREL DOXAZOSIN ACETYLSALICYLIC ACID EZETIMIBE + SIMVASTATIN PERINDOPRIL + AMLODIPINE ALLOPURINOL CALCITRIOL IVABRADINE	HISTOLOGY: DETACHMENT OF BMZ; DIF: LINEAR IGG AND C3 ON BMZ; SEVERE	PROBABLE ADR	3 6	Y
3	F	73	SITAGLIPTIN	8	14	N	INSULIN METFORMIN ESOMEPRAZOLE EZETIMIBE + SIMVASTATIN REPAGLINIDE PIOGLITAZONE PREGABALIN LACIDIPINE VALSARTAN OMEGA 3,6,9 FATTY ACID TICLOPIDINE	HISTOLOGY: DETACHMENT OF BMZ; DIF: LINEAR IGG AND C3 ON BMZ; SEVERE	PROBABLE ADR	5 4 · 5	Y
4	M	82	ALOGLIPTIN AND METFORMIN	2	6	N	ALFUZOSIN METFORMIN NEBIVOLOL + HYDROCHLOROTHIA ZIDE DIAZEPAM LORAZEPAM	HISTOLOGY: DETACHMENT OF BMZ; DIF: LINEAR IGG AND C3 ON BMZ; SEVERE	POSSIBLE ADR	5 5 · 5	N
5	F	63	VILDAGLIPTIN AND METFORMIN	1	4	N	RAMIPRIL PANTOPRAZOLE FUROSEMIDE GLICLAZIDE INSULIN DULOXETINE	HISTOLOGY: DETACHMENT OF BMZ; DIF: LINEAR IGG AND C3 ON BMZ; MODERATE	POSSIBLE ADR	4 4 · 5	Y

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118

119 **Table 2**

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C A S E	IgG IIF			EOSINOPHILS	TOTAL IgE (<100 kU/L neg)	IgE ELISA		IgG ELISA		IgG ELISA		Western Blot			IgA ELISA	
	normal skin	sss	Com			BP180	BP230	BP180	BP230	1 0 8 0	1 3 3 1	IgG on Kc extracts	IgG on LAD 1	IgA on LAD1	BP180	BP230
1	Pos	pos epid. side	pos	2.70 $10^3 \mu\text{L}$	4784	pos	pos	pos	pos	p o s	p o s	BP180	pos	neg	neg	neg
2	Pos	pos epid. side	pos	normal	662	neg	neg	pos	pos	p o s	p o s	BP180	pos	neg	neg	neg
3	Pos	pos epid. side	pos	1.61 $10^3 \mu\text{L}$	602	neg	pos	pos	pos	n e g	n e g	BP180	weak pos	neg	neg	neg
4	Pos	pos epid. side	neg	normal	1054	neg	pos	pos	pos	n e g	p o s	BP180	pos	neg	neg	neg
5	ND	ND		1.43 $10^3 \mu\text{L}$	158	ND	ND	pos	neg	N D	N D	ND	ND	ND	neg	neg