

enabling us to diagnose a cell-mediated hypersensitivity to bosentan. This higher sensitivity of the LTT has been observed in other studies concerning non immediate reactions to β -lactams (7) and other drugs, such as non β -lactam antibiotics and anticonvulsants (8). On the other hand, the activation of drug-specific T cells plays a central role in many non immediate drug hypersensitivity reactions, such as MPE, toxic epidermal necrolysis (TEN), DRESS, and acute generalized exanthematous pustulosis (AGEP) (8, 9). Therefore, a more extensive diagnostic use of the LTT – which is still generally considered merely a research tool – could improve the sensitivity of the diagnostic workup for non immediate reactions to drugs, reducing the number of drug provocation tests, which are not only harmful, but are also contraindicated in severe reactions such as TEN, DRESS, and AGEP. However, a recent study by Kano et al. (9) proved that the LTT sensitivity varies depending on the type of drug reaction and the time interval between such reactions and testing. In fact, positive results were obtained when the test was performed at the acute stage, but not at the recovery stage, in MPE and TEN, while positive responses were observed at the recovery stage, but not at the acute one, in DRESS, as in our case.

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Anaphylaxis to amidotrizoate proved by skin testing and flow cytometry-based basophil activation test

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Key words: anaphylaxis; contrast media; flow cytometry; skin tests.

Following immediate hypersensitivity reaction to iodinated contrast media (ICM), the use of flow cytometry-based

basophil activation test associated with skin tests could be a useful diagnostic tool.

A 75-year-old man was scheduled for transurethral prostatectomy. He denied any drug allergy. At the end of the transurethral prostatectomy, a retrograde ureteropyelography was performed. Within one minute after amidotrizoate injection, sinus bradycardia occurred (from 60 to 30 b.min⁻¹); arterial pressure was not detectable. No bronchospasm was observed. Cardiovascular homeostasis was restored within 5 min following resuscitative treatment. Further clinical outcome was uneventful. Plasma histamine and tryptase were not measured at the time of the reaction.

With the patient's consent, skin tests were performed according to standardized procedures (1, 2). Prick-tests (PTs) with amidotrizoate, iobitridol, iodixanol, iothexol, iomeprol, ioxaglate and ioxitalamate remained negative as well as intradermal tests (IDTs) with iobitridol, iodixanol, iothexol, ioxaglate and ioxitalamate performed till the pure solution. In contrast, IDT with amidotrizoate was found positive since 10⁻² dilution (from 2 to 6 mm within 20 min). Cross-reactivity was found with iomeprol using IDT. PTs with latex remained negative.

Basophil activation test (BAT) was performed using an adaptation of a commercially available technique (Flow-CAST, Bühlmann, Allschwill, Switzerland). Briefly, a leucocyte-rich suspension was stimulated with or without positive control (anti-Fc ϵ R1 monoclonal antibody), or with various ICM (amidotrizoate, ioxitalamate, ioxaglate and iothexol, dilutions ranging from 6 mg/ml to 6 μ g/ml). Cells were then double stained using FITC-labelled anti-IgE antibody and PE-labelled anti-CD63 antibody (Bühlmann), or using FITC-labelled anti-IgE antibody (BioSource Europe, Nivelles, Belgium) and PE-labelled anti-CD203c antibody (Beckman-Coulter, Roissy, France); cells were analysed using an EPICS-XL flow cytometer (Beckman-Coulter). Two healthy controls unexposed to ICM were simul-

Immediate reactions following iodinated contrast media may be investigated through skin tests and basophil phenotype.

Table 1. Basophil activation test expressed in stimulation index (SI = ratio of stimulated to unstimulated values) (normal SI < 2)

ICM	CD63 (in % of + cells)			CD203c (in MFI)		
	Patient	Ctrl 1	Ctrl 2	Patient	Ctrl 1	Ctrl 2
Amidotrizoate	0.3	1.5	1.7	2.6	1.8	1.5
Ioxitalamate	1	1.9	1	1.9	1.9	1.75
Ioxaglate	1	0.7	1	1	1	1
Iohexol	0.8	0.7	1.5	1	1.6	1

ICM, iodinated contrast medium; SI, stimulation index; MFI, median of fluorescence intensity; Ctrl, healthy control

taneously tested, as negative controls. As shown in Table 1, BAT was negative for CD63, whatever the concentrations used for the four ICM tested (ratio of stimulated to unstimulated positive basophil percentage lower than 2). In contrast, an upregulation of CD203c was observed in response to amidotrizoate only, leading to a stimulation index of 2.6 (ratio of stimulated to unstimulated basophil median fluorescence intensity). This specific response was observed at 6 mg/ml.

Taken together, these results confirmed the onset of a grade III (3) anaphylactic reaction caused by amidotrizoate. The clinical arguments were the short onset delay and the severity of the reaction. The allergological arguments were the IDT positivity (4) and the evidence of anti-amidotrizoate sensitized basophils detected by CD203c upregulation.

To better investigate the drug hypersensitivity reactions, some *ex vivo* basophil activation tests have been developed such as the upregulation of several cell markers quantified by flow cytometry, in particular CD63 or CD203c (5). One single study recently reported CD63 upregulation in three patients with documented ICM anaphylaxis in correlation with skin tests (6). In our patient, only CD203c was upregulated, as already reported for other drugs, given the various mechanisms involved in the

regulation of these two markers (5). As this amidotrizoate-induced CD203c upregulation correlated with positive skin test, we can suggest the existence of anti-amidotrizoate IgE-bearing basophils in this patient; however, standardized methods for specific IgE against ICM are still lacking. No cross-reactivity with the different ICM tested *ex vivo* (ioxitalamate, ioxaglate and iohexol) was observed in correlation to the skin test results. In our patient, iobitridol, iodixanol, iohexol, ioxaglate and ioxitalamate might be therefore re-injected for further procedures.

The present study performed in one patient with anaphylactic reaction to amidotrizoate confirms that basophil activation analysis by flow cytometry can be a useful tool in addition to the clinical history and skin tests and thus needs to be further investigated.

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