

Anaphylactic shock to fresh-frozen plasma inactivated with methylene blue

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BACKGROUND: Methylene blue allergy is a well-known entity associated with food or lymph node location. Inactivation of viruses by methylene blue in fresh-frozen plasma (FFP-MB) has been recently introduced in France after many years of use in other parts of Europe.

CASE REPORT: We describe here two anaphylactic shock reactions occurring during FFP-MB infusion in patients after cardiac surgery. The follow-up was favorable using epinephrine infusion in one patient and extracorporeal circulatory assistance in the other.

RESULTS: The allergy was suspected based on the following arguments: chronology of the event, absence of other allergen infused to patients, and an associated symptom (generalized rash). Methylene blue allergy was documented by prick tests and/or intradermal reactions with methylene blue and patent blue. Additional allergic tests with basophil activation tests confirm the role of the dye in the anaphylactic reaction.

CONCLUSION: These two cases questioned the safety of FFP-MB.

Methylene blue is a commonly used dye to detect digestive or urinary fistulas. It has been recently used in France to inactivate viruses in fresh-frozen plasmas (FFP-MB). We report here the first two cases of anaphylactic shock occurring during FFP-MB infusion resulting from methylene blue-patent blue crossed allergy.

CASE REPORT

The first case occurred in a 64-year-old woman undergoing coronary artery bypass grafting in September 2008. She previously suffered from a breast neoplasm that required chemotherapy and lumpectomy with sentinel lymph node biopsy after patent blue injection. She was allergic to shellfish and hymenopteran.

Preoperative evaluation showed myocardial infarction on electrocardiogram and altered left ventricle function (ejection fraction, 40%) on echocardiography. Her daily treatment consisted of 2.5 mg of bisoprolol, 2.5 mg of ramipril, and 300 mg of aspirin.

After an uneventful surgery of 3 hours 39 minutes' duration, the patient was transferred to the intensive care unit and maintained on mechanical ventilation. She then had stable hemodynamic variables without cardiovascular support (heart rate 80 bpm and arterial blood pressure 115/65 mmHg). Owing to a sudden diffuse oozing of the surgical bed and of puncture points and slightly increased thoracic drainages (60 mL/hr), one red blood cell unit and 2 FFP-MB units (EFS-Etablissement Français du Sang, Bordeaux, France) were ordered. During the second FFP-MB infusion, a cardiovascular collapse (45/32 mmHg) suddenly occurred with generalized rash and a severe drop of SpO₂ not associated with a bronchospasm (SpO₂ 85% with 70% FiO₂). Five boluses of 0.1 mg of epinephrine 1:100 aqueous solution followed by a 1 µg/kg/min continuous infusion associated to 1 µg/kg/min norepinephrine were required to restore an acceptable hemodynamic status (arterial pressure 100/48 mmHg). Transesophageal echocardiography revealed an overall hypokinetic left ventricle with an ejection fraction of 18%. After hemodynamic stabilization, chest X-rays showed

ABBREVIATIONS: ECMO = extracorporeal circulatory assistance; FFP-MB = methylene blue in fresh-frozen plasma.

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Received for publication February 2, 2010; revision received May 14, 2010, and accepted May 17, 2010.
doi: 10.1111/j.1537-2995.2010.02800.x

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right unilateral interstitial infiltrates. Owing to this poor hemodynamic status despite high doses of catecholamine, extracorporeal circulatory assistance (ECMO) was required for 4 days. Catecholamine doses were maintained for 48 hours and ECMO for 4 days before weaning with an ejection fraction near 30%. She still required mechanical ventilation during an additional 7 days before leaving hospital on Day 20 for rehabilitation. Further FFP required while on ECMO was inactivated by the solvent/detergent (S/D) technique.

During this episode, the serum histamine level reached 25.8 nmol/L (limit of positive values 6 nmol/L), and tryptase 26.7 µg/L (limit of positive value 12.5 µg/L). Seven weeks after, skin tests for all the drugs administered during anesthesia were performed as recommended.¹ Undiluted patent blue V (2.5%; Guerbet, Roissy, France) was used for prick test. A decreasing dilution scale was used for intradermal tests (starting dose 1:1000, maximal concentration 1:10). Prick tests with patent blue were considered positive (papule of 3 mm diameter with patent blue and of 5 mm with codeine control positive test) but prick tests with methylene blue were negative. Prick tests with latex were also negative. Intradermal reaction tests with four dilutions of pure methylene blue (10^{-4} to 10^{-1}) were positive at a 10^{-2} dilution. In vitro basophil activation in presence of allergens was measured by flow cytometry. Activation was positive for patent blue (activation index reaching 758) but equivocal with methylene blue (activation index 58 with positive control index of 75 and negative control test of 1.5). This test gave same results several weeks after. Additional tests were positive with FFP-MB from the same donor (activation index 758) and equivocal with FFP-SD (activation index 48) (positive control with FMLP of 75 and with anti-IgE 10.3, negative control 1.5).

The second case in April 2009 was a 75-year-old man who underwent coronary artery bypass grafting surgery with atrial fibrillation correction. His past history included hypertension, atrial fibrillation, carotid stenosis, and atopic rhinitis. He had no previous surgery or food or drug allergy. His usual treatment associated 15 mg of fluindione, 8 mg of perindopril, 40 mg of atorvastatine, 5 mg of bisoprolol, 10 mg of amlodipine, and 40 mg of furosemide. Preoperative ejection fraction reached 70%, and echocardiography revealed pulmonary hypertension with slight aortic and mitral regurgitation. Surgery was uneventful (5-hr duration), then the patient was transferred to the intensive care unit and maintained under sedation. Low doses of epinephrine (0.3 mg/hr, 3 mL/hr of a 1:1000 solution) were required to maintain hemodynamic stability (heart rate 70 bpm, arterial pressure 110/60 mmHg). Six hours after surgery, thoracic drains losses increased and one FFP-MB (EFS) was ordered. As soon as it was infused, a hemodynamic shock occurred with generalized urticaria but without bronchospasm (systolic arterial pressure was 56 mmHg).

FFP-MB was stopped immediately and five 0.1-mg bolus injections of epinephrine were given followed by epinephrine continuous infusion (1.5 mg/hr, 15 mL/hr of a 1:1000 solution) to restore arterial pressure to 140/60 mmHg with a heart rate of 110 bpm. Epinephrine infusion was maintained until Day 3 and the patient left intensive care on Day 4 without sequelae. Plasma histamine and tryptase were not obtained at this moment but the patient was scheduled to immunologic testing 7 weeks thereafter. Intradermal tests with four dilutions of undiluted product (10^{-4} to 10^{-1}) were positive to patent blue at 10^{-2} and to methylene blue at 10^{-1} . Latex prick tests were negative. In vitro basophil activation test quantification by flow cytometry was positive for methylene (degranulation index 123) and patent blue (degranulation index 295).

DISCUSSION

Clinical symptoms exhibited by these two patients were consistent with Grade III anaphylaxis to FFP-MB. This hypothesis was confirmed by histamine and tryptase levels in Patient 1 and by in vivo or in vitro tests with patent and methylene blue. In these two cases, patients were allergic to both dyes. Cross reaction between them is highly suspected because the two patients exhibited a marked reaction to patent blue more than to methylene blue.

At the time of the reaction, no medications other than FFP-MB were administered. Prick tests with latex were negative. The increase in histamine and tryptase levels confirms the anaphylaxis reaction in the first patient. We did not obtain these dosages in the second patient. However, the positive reaction to prick tests or dermal injection to patent blue and methylene blue in the patients confirm that both of them had cross-allergies to these two dyes. Basophil activation quantification by flow cytometry in the presence of patent blue and methylene blue add further certitude to the results of cutaneous tests.

Neither of our patients was known to have allergy to one of the dyes. The first patient was previously exposed to patent blue at the time of lymph node localization. First exposure and subsequent sensitization to patent or methylene blue may have gone unnoticed in the second patient because both of them are used as common dyes for industry and food adjuvant (E131).² Allergy to methylene blue is supposed to be very rare because the small size of the molecule reduces the risk of sensitization to a hapten-protein complex.³ In contrast, patent blue may bind to proteins and gives more frequent and severe allergic reactions than methylene blue.⁴ It has been suggested that 1 per thousand to 2.7% of patients receiving patent blue may develop an anaphylactic reaction.^{5,6} The occurrence of anaphylaxis may also be dose-related.⁷ Anaphylaxis is triggered when bridging between IgE present at the mas-

toocytes and basophil surface is done by a sufficient number of allergens. Specific IgE to patent blue has been described but not to methylene blue.⁸ Because methylene and patent blue are not supposed to cross-react despite similarities in their structures, methylene blue has been recommended for lymph node detection before biopsy.^{2,5,9} However, in a series of 14 patients, Mertes and colleagues¹⁰ observed a cross-reaction between methylene and patent blue while performing prick tests in one patient. Such a cross-reactivity was also described in three melanoma patients.¹¹ In the present two cases, patient skin positively reacted to both dyes and clinical or *in vitro* tests exhibited a higher immunization to patent blue than methylene blue suggesting a cross-sensitization. The use of *in vitro* basophil activation test in presence of a suspected allergen confirm its implication in the anaphylactic reaction and its interest to analyze *ex vivo* allergens potentially implicated into the clinical reaction without any risk for patients to be exposed again to the allergen. This test quantifies by flow cytometry the membrane receptors CD63 and IgE at the surface of basophils after a previous phase of immunofluorescence labeling.¹² Results are expressed as an activation index of basophils according to the percentage of CD63 and IgE receptors present at the surface of basophils measured by fluorescence intensity. Three controls are done to compare results and to make sure there is no basophils spontaneous activation. The negative control activation index must be less than 5 and the two positive controls with MFI-CD63 and MFI-IgE higher than 15 (%CD63).

The occurrence of allergy to FFP-MB due to methylene blue in these two patients at a short time interval gives cause of concerns. During the same period of time, we observed these two reactions after administration of 2432 FFP-MB units (490 in 2008 and 1942 in 2009), but no comparable reactions after 2677 FFP units virus-attenuated by the S/D method. These consecutive events may have occurred solely by chance but the high frequency of events and the context of cardiac surgery are striking. Clinical consequences were probably potentiated by the immediate postoperative context of cardiac surgery. The associated cardiac treatments such as beta blockade or angiotensin-converting enzyme inhibitors decrease cardiac adaptation to hypovolemia induced by anaphylaxis vasodilation. Angiotensin-converting enzyme inhibitors also decrease the clearance of bradykinin released during anaphylactic shock.

These two cases describe here the first anaphylactic severe reaction to FFP inactivated by methylene blue. This raises concern about the use of this blood product at a large scale and its innocuousness in the field of hemorrhagic shock or cardiac surgery. In our institution the largest use of blood derivatives is for cardiac surgery. This may explain why we observed these reactions in this area. Moreover, the intense inflammatory reaction induced by

extracorporeal circulation (e.g., complement products activation) might have potentiated the anaphylactic reaction explaining thus why we observed these two events only after cardiac surgery. FFP virus-attenuated by methylene blue has been released into the French market since July 2008. In contrast to pooled FFP virus-attenuated by the S/D method, FFP-MB is extracted by apheresis from unit individual blood donation. Only men and women without previous pregnancy can give plasma to avoid the risk of transfusion-related acute lung injury. After preparation, methylene blue concentration must be less than 30 µg/L and the minimum volume of FFP-MB unit is 200 mL. Plasma illumination of methylene produces free radicals destroying the lipid envelope of virus such as hepatitis C and B or human immunodeficiency virus.¹³ This type of plasma was used formerly in Germany, Austria, and Switzerland but has recently been replaced by FFP secured by quarantine arguing the mutagenic risk of methylene blue.¹⁴ FFP-MB is supposed to share a similar effect to other FFP but the photoinactivation by methylene blue damage the histidine residues of fibrinogen altering their polymerizing ability into fibrin.^{15,16} Fibrinogen and factor VIII concentrations are lower in FFP-MB than in other FFP leading to increase the volume infused to obtain the same clinical effect.^{17,18} No other allergic reaction after FFP-MB infusion has previously been related in the literature to methylene blue. This may related to the large heterogeneity of how to declare adverse events leading to a 50-fold difference of reported adverse events with voluntary declarations.¹⁹ In Germany as in the United Kingdom, the hemovigilance system is based on voluntary declarations, whereas in France, adverse effects declaration is mandatory. Some systems are government regulated (France and Germany) but in others regulation is made by medical societies (United Kingdom) or public health authorities (Canada). In Germany, declarations are not centralized on a national basis but on a regional basis. Such system may lack or underevaluate some rare adverse effects such as rare allergy. Another hypothesis might be differences in the mode of production of FFP-MB.

We report the first two severe anaphylactic shock reactions triggered by FFP virus-attenuated by methylene blue infused after cardiac surgery. One patient was known to have been previously exposed to patent blue and exhibited cross-allergy between methylene blue and patent blue, suggesting that cross-allergy between the two dyes is possible. In the other case, no clear exposure to methylene blue was known but this dye, as well as patent blue, is commonly used in the industry and as a food adjuvant. These cases also raise questions about the risks of FFP-MB infusion especially in its common indication, hemorrhagic shock. The persistence of shock despite correct blood loss compensation should lead clinicians to investigate a possible allergy to FFP-MB, to stop its administration, and to sample blood to measure tryptase and

histamine levels and then to explore patients by in vivo skin tests and in vitro basophil degranulation tests. Previous exposure to patent blue for lymphography or lymph node localization should also probably be a contraindication to FFP-MB. The large use of blue dyes in the industry and as a food adjuvant questions the safety of methylene blue for medical purpose.

CONFLICT OF INTEREST

None.

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