

Ceftazidime-induced drug reaction with eosinophilia and systemic symptoms (DRESS) complicated by hemophagocytic lymphohistiocytosis

Matthieu Picard, MD, FRCPC^a, PhD, M. Isabel Fernandez^{b,c}, Anne Des Roches, MD, FRCPC, FAAAAI^{c,d}, Philippe Bégin, MD, MSc, FRCPC^a, Jean Paradis, MD, FRCPC^a, Louis Paradis, MD, FRCPC, FAAAAI^a, and Françoise Le Deist, MD, PhD^{b,c,d}

Clinical Implications

- Hemophagocytic lymphohistiocytosis is a rare but potentially fatal complication of drug reaction with eosinophilia and systemic symptoms (DRESS), which could be mediated by drug-induced CD8⁺ T-cell activation and IFN- γ secretion.
- Prompt diagnosis and treatment of DRESS are key to prevent disease progression and possibly associated complications such as hemophagocytic lymphohistiocytosis and multiple drug hypersensitivity.

TO THE EDITOR:

Drug reaction with eosinophilia and systemic symptoms (DRESS) is among the most severe drug hypersensitivity reactions with an estimated mortality of 10%.¹ Its diagnosis can be challenging, given its rarity and atypical presentation for an adverse drug reaction. For instance, DRESS may progress despite stopping the implicated agent(s) and may persist for weeks with relapses even months after onset.¹ Prompt diagnosis and treatment are key to prevent clinical deterioration. Herein, we present a case of DRESS, most likely induced by ceftazidime, for which diagnosis was delayed and which progressed to hemophagocytic lymphohistiocytosis (HLH), a seldom-described but potentially deadly complication characterized by hypercytokinemia and multiple organ infiltration by activated CD8⁺ T cells.²

A previously healthy 55-year-old woman developed bacterial meningitis after ventricular drain placement for subarachnoid hemorrhage. The diagnosis of bacterial meningitis was based on the presence of a marked neutrophilic pleocytosis (white blood cell count, $5280 \times 10^6/L$; normal $\leq 5 \times 10^6/L$; neutrophils, 90%), elevated protein concentration (4.14 g/L; normal, <0.4 g/L) and low glucose concentration (9.0 mg/dL; normal, 50.5–75.7 mg/dL) on cerebrospinal fluid (CSF) analysis although cultures were negative, because antibiotics had been started beforehand. She improved rapidly on vancomycin and ceftazidime. However, after 13 days of treatment, fever recurred, and on the next day (day 14) she developed a maculopapular rash at which point ceftazidime was replaced with meropenem (Figure 1, A). Vancomycin and meropenem were stopped when repeat blood and CSF cultures (drawn on day 13) came back negative on day 16. Resolution of the initial bacterial meningitis was further supported by a repeat CSF analysis drawn on day 13 (white blood cell, $12 \times 10^6/L$; protein, 0.43 g/L; glucose, 55.9 mg/dL). Fever abated on the next day and she was discharged.

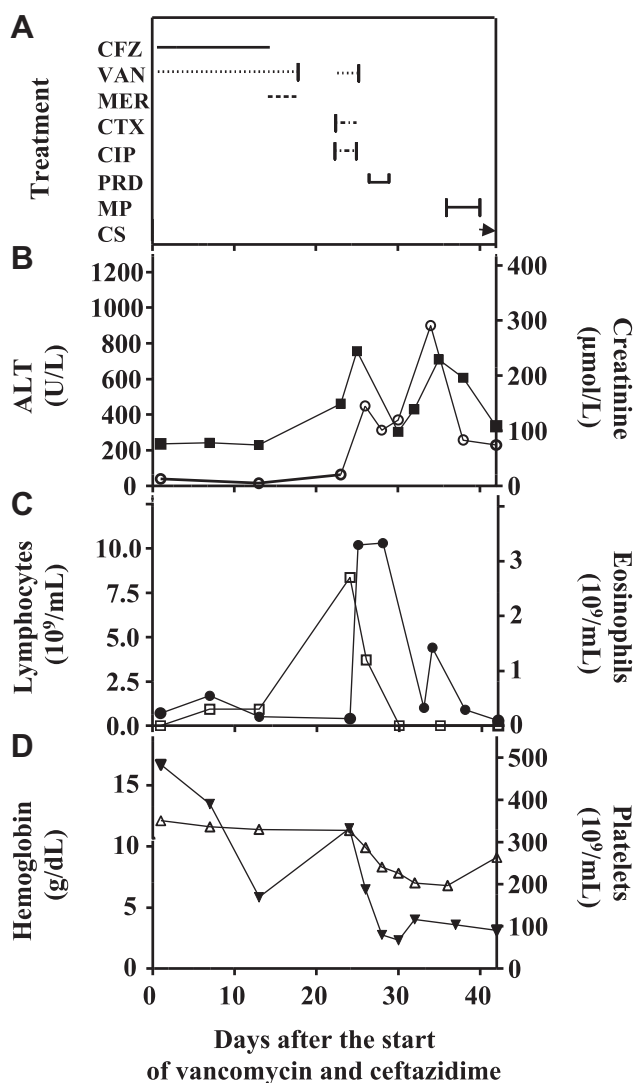


FIGURE 1. Clinical parameters over time. **A**, Medication given before and after the onset of DRESS. CFZ, ceftazidime; VAN, vancomycin; MER, meropenem; CTX, ceftriaxone; CIP, ciprofloxacin; PRD, prednisone 40 mg; MP, methylprednisolone; CS, long-term corticosteroid treatment. **B**, ALT (alanine aminotransferase) (open circle) and creatinine (filled square) levels over time. **C**, Absolute lymphocyte (filled square) and eosinophil (open square) counts over time. **D**, Hemoglobin (open triangle) and platelet (filled triangle) counts over time.

On day 22, the patient was re-admitted for recurrent fever. Vancomycin, ceftriaxone, and ciprofloxacin were given for 48 hours until another set of blood and CSF cultures came back negative. In parallel, the fever persisted and the rash progressed with several associated blisters but without mucosal involvement. Hematologic, hepatic, and renal abnormalities also developed (Figure 1, B–D). On day 26, prednisone was given for suspected interstitial nephritis and then stopped, after 2 days, because gallium scan was normal and eosinophiluria was absent.

TABLE I. RegiSCAR diagnostic score for DRESS

	Score				Patient score
	-1	0	1	2	
Fever $\geq 38.5^{\circ}\text{C}$	No/U	Yes			0
Enlarged lymph nodes		No/U	Yes		0
Eosinophilia ($\times 10^9/\text{L}$)			0.7-1.499	≥ 1.5	2
Atypical lymphocytosis		No/U	Yes		1
Skin involvement (maximum = 2)					2
Skin rash extent (% body surface area)		No/U	$>50\%$		$>50\%$
Skin rash suggesting DRESS	No	U	Yes		Yes
Biopsy suggesting DRESS	No	Yes/U			Yes
Organ involvement (maximum = 2)					2
Liver		No/U	Yes		Yes
Kidney		No/U	Yes		Yes
Lung		No/U	Yes		No
Muscle/heart		No/U	Yes		No
Pancreas		No/U	Yes		No
Other organ		No/U	Yes		No
Resolution ≥ 15 d	No/U	Yes			0
Evaluation of other potential causes					
Antinuclear antibody					Negative
Blood culture					Negative
Serology for HAV/HBV/HCV					Negative
Chlamydia/mycoplasma					U
If none positive and ≥ 3 negative			Yes		1
Total score (maximum = 9)					8

HAV, Hepatitis A; HBV, hepatitis B; HCV, hepatitis C; RegiSCAR, Registry of severe cutaneous adverse reactions; U, unknown.

Final score <2 , no case; 2-3, possible case; 4-5, probable case; >5 , definite case. Based on data from Cacoub et al.¹

An allergy consultation was requested on day 34 after which DRESS was diagnosed on the basis of the presence of characteristic findings: fever and rash, hematologic abnormalities (eosinophilia, atypical lymphocytes), and systemic involvement (hepatic and renal failure) (Table I). Hepatosplenomegaly or enlarged lymph nodes, which are frequently found in DRESS, were absent. Superimposed HLH was suspected because the patient showed bicytopenia, hypofibrinogenemia, and hyperferritinemia. A marked increase in peripheral blood activated CD8⁺ T cells confirmed the diagnosis³ (Table II). Serum level of IFN- γ was elevated (571 ± 62 pg/mL), but hemophagocytosis was not observed on a bone marrow biopsy performed 7 days after starting corticotherapy. It should be noted that demonstration of hemophagocytosis is not required for HLH diagnosis.² Other causes of HLH were sought, but no evidence of malignancy, autoimmune disease, or infection was found. Notably, Epstein-Barr virus and cytomegalovirus viral loads were undetectable in whole blood. Antinuclear antibody, antineutrophil cytoplasmic antibody, and rheumatoid factor were negative as well as serology for hepatitis viruses and HIV.

The patient was treated with intravenous methylprednisolone (1 g/d for 3 days) starting on day 34, followed by oral prednisone

(1 mg/kg per day) with rapid improvement in all parameters. Two months later, on decreasing the dose from 10 to 5 mg/d, the rash recurred without any systemic involvement. It responded promptly to an increase in prednisone to 20 mg/d. A slower prednisone taper, over 7 months, led to complete remission. More than 3 years after the event the patient remains healthy.

Vancomycin and ceftazidime were suspected as the DRESS-inducing drugs. Because of the severity of the clinical reaction and risk of disease reactivation with skin testing, which in this particular patient was felt to be prohibitive, *in vitro* tests were performed to identify the culprit drug as previously described.⁴ Methods used for the *in vitro* tests are detailed (see this article's Methods section in the Online Repository at www.jaci-inpractice.org). Tests were performed 5 and 13 months after corticosteroid withdrawal and in healthy controls concomitantly. Ceftazidime and vancomycin induced marked lymphocyte proliferation, IFN- γ secretion and T-cell activation in the patient (Figure 2), indicating sensitization to both drugs. Therefore, one possibility is that DRESS was induced by both ceftazidime and vancomycin. However, most cases of DRESS are induced by a single drug, and patients with DRESS are known to be at risk for multiple drug hypersensitivity (MDH).^{1,5} Indeed, some patients develop sensitization to drugs given during the course of their reaction that are unrelated to the inciting agent and can, as a result, experience exacerbations of their disease.⁵ Here, vancomycin was re-administered later during the reaction, on day 22, after which the patient had a clinical deterioration. Therefore, an alternative explanation is that DRESS was solely induced by ceftazidime and that sensitization to vancomycin results from MDH.

Given the possibility of MDH in this patient, sensitivity to other drugs administered during DRESS was assessed. Ceftriaxone and meropenem but not ciprofloxacin induced IFN- γ secretion and T-cell activation in the patient (Figure 2). Whereas the observed sensitivity to ceftriaxone might depend on cross-reactivity with ceftazidime (two cephalosporins), sensitivity to meropenem (a carbapenem) is likely explained by MDH.⁶ In addition, ceftriaxone administration on day 22 could have contributed to the clinical deterioration that followed. These findings support the need to limit, to a minimum, drugs given to patients with DRESS because they are at increased risk of sensitization.⁵

The patient was advised to strictly avoid cephalosporins, carbapenems, and vancomycin. In addition, we advised caution with penicillins because cross-reactivity with cephalosporin and carbapenems is a possibility and reactivity to penicillins was not assessed.⁶ Caution was advised about ciprofloxacin as well because the negative predictive value of *in vitro* tests is unknown.⁴ If penicillins or ciprofloxacin were to be required, skin testing followed by a drug challenge could be considered.

At last, we sought to further define the *in vitro* drug-induced cytokines and chemokines that could be involved in DRESS-related HLH. Ceftazidime and ceftriaxone induced secretion of CCL2, CCL4, CXCL8, IL-5, GM-CSF, and G-CSF (see Figure E1 in this article's Online Repository at www.jaci-inpractice.org). Interestingly, CCL2, CCL4, and CXCL8 may play an important role in HLH pathophysiology,⁷ and IL-5, GM-CSF, and G-CSF are known to induce eosinophilia.⁸ IL-2, IL-6, IL-10, IL-13, and TNF- α secretion was induced by all implicated drugs, except for meropenem, which did not induce IL-13 secretion (see Figure E2 in this article's Online Repository

TABLE II. Hemophagocytic lymphohistiocytosis diagnostic criteria

Criteria	Patient
Fever	Yes
Splenomegaly	No
Cytopenias*	Hemoglobin, 5.9-8.5 g/dL (days 28-40) Platelets, 65-92 × 10 ⁹ /L (days 28-30)
Hypofibrinogenemia (<150 mg/dL) and/or hypertriglyceridemia (fasting >265 mg/dL)	Fibrinogen, 55-115 mg/dL (days 30-41)
Hemophagocytosis in bone marrow, spleen, lymph nodes, or liver	Absent on bone marrow biopsy (7 d after starting corticosteroid therapy)
Low or absent NK-cell activity	Not done
Ferritin >500 ng/mL	Ferritin, 3708 ng/mL (day 31)
Elevated soluble CD25 (soluble IL-2 receptor-α)	Activated T cells [†] (as judged by expression of HLA class II; normal <15%): HLA-DR/CD8 ⁺ , 90% HLA-DR/CD4 ⁺ , 32%

NK, Natural killer.

Diagnosis of hemophagocytic lymphohistiocytosis is made when at least 5 of the 8 criteria are fulfilled.

*Affecting at least two of three lineages in the peripheral blood: hemoglobin, <9 g/dL; platelets, <100 × 10⁹/L; neutrophils, <1 × 10⁹/L.

[†]Expression of HLA class II on T cells is used as a surrogate for measurement of soluble CD25.

Based on data from Filipovich.²

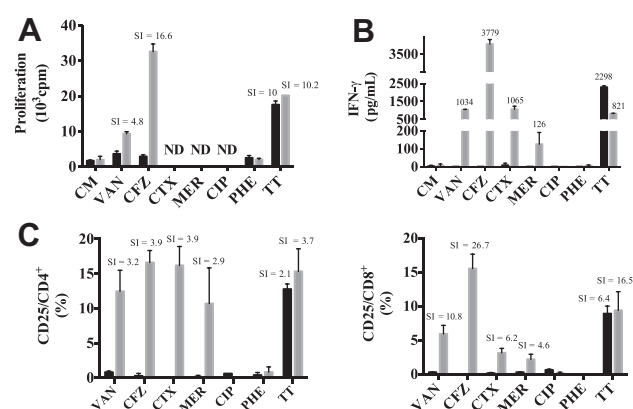


FIGURE 2. Drug stimulation of lymphocytes from the patient (gray bar) and a healthy control (black bar). **A**, Proliferation; cpm, counts per minute; ND, not done. **B**, IFN-γ secretion. **C**, CD25 induced expression on CD4⁺ (left) and CD8⁺ (right) cells. Means ± SDs were calculated from quadruplicate (proliferation) and triplicate (IFN-γ and CD25) results. A positive result, which indicates sensitization, is defined as a stimulation index (SI) >3 for proliferation (cpm with drug/cpm with CM) and >2 for CD25 expression (% positive cells with drug/% positive cells with CM). A level of IFN-γ secretion two SDs above the mean of control samples (threshold calculated at 13.2 pg/mL) is considered a positive result. SI and IFN-γ values are shown only for positive results. VAN, vancomycin; CFZ, ceftazidime; CTX, ceftriaxone; MER, meropenem; CIP, ciprofloxacin; PHE, phenytoin; TT, tetanus toxoid; CM, culture medium.

at www.jaci-inpractice.org). These cytokines are commonly found in DRESS and, except for IL-13, also in HLH and reflect lymphocyte activation.^{2,8} Finally, no drug induced the secretion of eotaxin, IL-1β, IL-4, IL-7, IL-12, IL-17A, and vascular endothelial growth factor.

Although several cases of DRESS and HLH have been reported, most ascribed HLH to a concomitant infection and none described the associated immunologic features or established the link between the two diseases.⁹ In our case, no other

cause for HLH, besides DRESS, was identified. In addition, the patient's immune profile at time of diagnosis, characterized by the marked CD8⁺ T-cell activation and IFN-γ secretion typical of HLH,^{2,3} was reproduced *in vitro* especially by ceftazidime more than a year after the event, suggesting that drug-induced T cells were the effectors of HLH in this patient. DRESS and HLH appear somewhat similar, being characterized by activated lymphocytes and hypercytokinemia.^{2,10} However, although a marked and predominant CD8⁺ T-cell activation is a hallmark of HLH, this is not a constant finding in DRESS, hence providing a possible explanation to why few patients with DRESS develop HLH.¹⁰ That being said, much remains to be learned on the mechanisms underlying the diverse clinical manifestations of DRESS. Epstein-Barr virus and cytomegalovirus were the only herpes viruses tested, and it cannot be excluded that another herpes virus reactivation could have contributed to HLH. However, most patients with DRESS reactivate herpes viruses and very few develop HLH, making this hypothesis unlikely.^{1,9}

This study also indicates the usefulness of *in vitro* tests as a completely safe mean of identifying drug sensitization in patients having experienced a severe drug hypersensitivity reaction for which skin testing is felt to be too high risk. Efforts should be made to bring those tests from research to clinical practice. Increasing awareness about DRESS is key to ensure disease recognition and to prompt institution of treatment, which might reduce the risk of complications such as HLH and hopefully reduce the associated mortality.

Acknowledgments

M.P., F.L.D., and L.P. contributed to conception and design. M.P., M.I.F., L.P., and J.P. contributed to data acquisition. All authors contributed to data analysis and interpretation. M.P. and M.I.F. drafted the article and A.D.R., P.B., L.P., J.P., and F.L.D. reviewed it critically. All authors have read and approved this version of the manuscript.

^aDivision of Allergy and Clinical Immunology, Department of Medicine, Centre Hospitalier de l'Université de Montréal (CHUM), Montreal, Quebec, Canada

^bDepartment of Microbiology and Immunology, CHU Sainte-Justine Research Center, Université de Montréal, Montreal, Quebec, Canada

^cCHU Sainte-Justine Research Center, Université de Montréal, Montreal, Quebec, Canada

^dDivision of Allergy and Clinical Immunology, Department of Pediatrics, CHU Sainte-Justine, Montreal, Quebec, Canada

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Corresponding author: Françoise Le Deist, MD, PhD, CHU Sainte-Justine, Department of Microbiology and Immunology, 3175 Chemin de la Côte-Sainte-Catherine, Montreal, QC H3T 1C5, Canada. E-mail: francoise.le.deist@umontreal.ca.

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REFERENCES

1. Cacoub P, Musette P, Descamps V, Meyer O, Speirs C, Finzi L, et al. The DRESS syndrome: a literature review. *Am J Med* 2011;124:588-97.
2. Filipovich AH. The expanding spectrum of hemophagocytic lymphohistiocytosis. *Curr Opin Allergy Clin Immunol* 2011;11:512-6.
3. Ouachee-Chardin M, Elie C, de Saint Basile G, Le Deist F, Mahlaoui N, Picard C, et al. Hematopoietic stem cell transplantation in hemophagocytic lymphohistiocytosis: a single-center report of 48 patients. *Pediatrics* 2006;117:e743-50.
4. Lochmatter P, Zawodniak A, Pichler WJ. In vitro tests in drug hypersensitivity diagnosis. *Immunol Allergy Clin North Am* 2009;29:537-54.
5. Daubner B, Groux-Keller M, Hausmann OV, Kawabata T, Naisbitt DJ, Park BK, et al. Multiple drug hypersensitivity: normal Treg cell function but enhanced in vivo activation of drug-specific T cells. *Allergy* 2012;67:58-66.
6. Depta JP, Pichler WJ. Cross-reactivity with drugs at the T cell level. *Curr Opin Allergy Clin Immunol* 2003;3:261-7.
7. Tamura K, Kanazawa T, Tsukada S, Kobayashi T, Kawamura M, Morikawa A. Increased serum monocyte chemoattractant protein-1, macrophage inflammatory protein-1beta, and interleukin-8 concentrations in hemophagocytic lymphohistiocytosis. *Pediatr Blood Cancer* 2008;51:662-8.
8. Lochmatter P, Beeler A, Kawabata TT, Gerber BO, Pichler WJ. Drug-specific in vitro release of IL-2, IL-5, IL-13 and IFN-gamma in patients with delayed-type drug hypersensitivity. *Allergy* 2009;64:1269-78.
9. Eshki M, Allanore L, Musette P, Milpied B, Grange A, Guillaume JC, et al. Twelve-year analysis of severe cases of drug reaction with eosinophilia and systemic symptoms: a cause of unpredictable multiorgan failure. *Arch Dermatol* 2009;145:67-72.
10. Nishio D, Izu K, Kabashima K, Tokura Y. T cell populations propagating in the peripheral blood of patients with drug eruptions. *J Dermatol Sci* 2007;48:25-33.

METHODS

Informed consent was obtained from the patient and healthy controls. This study was approved by the Centre Hospitalier de l'Université de Sainte-Justine Review Board.

A blood sample was collected at time of DRESS/HLH diagnosis, and serum was isolated and frozen at -20°C until use. Blood samples were also collected at 5 and 13 months after corticosteroid withdrawal from the patient and a healthy control at the same time and processed concomitantly. Two healthy controls were used: one tolerant to ceftazidime and vancomycin for all assays except proliferation and one unexposed to all these antibiotics for the proliferation assay.

Peripheral blood mononuclear cells (PBMCs) were isolated from heparinized blood with the use of density gradient centrifugation (Ficoll/Hypaque). PBMCs were seeded at $2 \times 10^5/\text{well}$ into round-bottom 96-microwell plates in triplicates or quadruplicates with various drug concentrations, tetanus toxoid (TT; Aventis Pasteur, France; 1/250) as positive control and culture medium alone as negative control. Culture medium consisted of RPMI-1640 (45%), AIM V (45%) supplemented with human AB serum (10%). Drugs used were diluted from the commercially available intravenous formulations and consisted of vancomycin (100 $\mu\text{g}/\text{mL}$), ceftazidime (100 $\mu\text{g}/\text{mL}$), ceftriaxone (100 $\mu\text{g}/\text{mL}$), meropenem (50 $\mu\text{g}/\text{mL}$), ciprofloxacin (5 $\mu\text{g}/\text{mL}$), and phenytoin (50 $\mu\text{g}/\text{mL}$).

Lymphocyte transformation test was performed with vancomycin, ceftazidime, phenytoin, and TT in quadruplicates as previously described.^{E1} After 6 days of culture, 0.5 Ci of

^3H -thymidine was added to culture wells for 14 hours, and its uptake was calculated as counts per minute (cpm). Results represent the mean of quadruplicate cultures with SD. A positive result was defined as a stimulation index >3 , which is calculated by dividing the proliferation (cpm) induced by a drug by the proliferation induced by the culture medium as previously described.^{E1}

Flow cytometry was performed after 5 days of culture with vancomycin, ceftazidime, ceftriaxone, meropenem, ciprofloxacin, phenytoin, and TT in triplicates as previously described.^{E2} Briefly, culture supernatant fluids were collected and frozen at -20°C until use, and PBMCs were washed and stained with anti-human monoclonal antibodies (allophycocyanin-CD4, allophycocyanin-cyanine 7-CD8, phycoerythrin-CD25, and fluorescein isothiocyanate-CD71). Cells were analyzed with a FACSCanto cytometer and the software DIVA. Results represent the means and SDs of triplicates and are shown for CD25. CD71 induced an expression profile similar to CD25 (data not shown). Induced expression was calculated as follows for each triplicate: $(\% \text{CD}25/\text{CD}4^{+} \text{ or } \text{CD}8^{+} \text{ with drug}) / [100 - (\% \text{CD}25/\text{CD}4^{+} \text{ or } \text{CD}8^{+} \text{ with culture medium})]$. A positive result was defined as stimulation index >2 which is calculated by dividing the percentage of cells expressing CD25 when cultured with a drug by the percentage of cells expressing CD25 when cultured with the culture medium as previously described.^{E2}

Culture supernatant fluids, serum stored from the blood sample of the patient taken at time of DRESS diagnosis, and human AB serum (as a negative control for the patient serum)

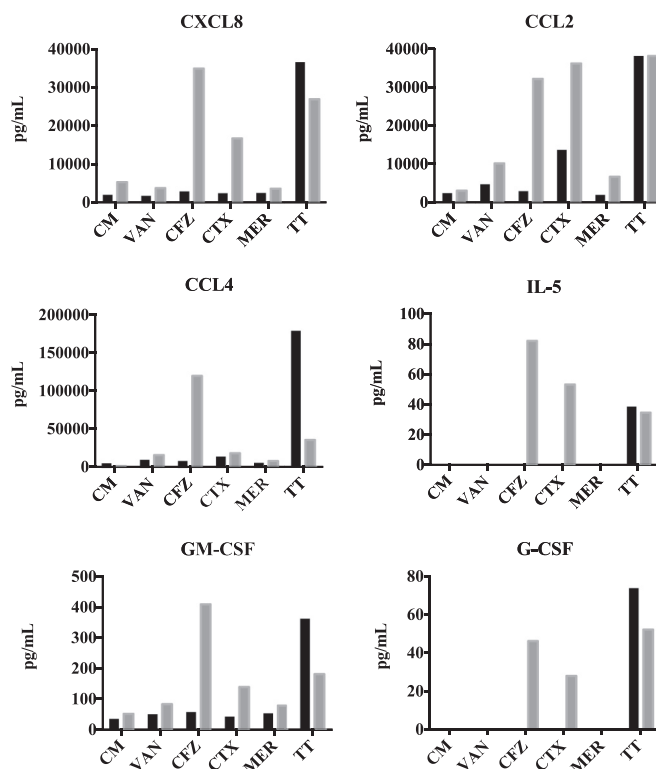


FIGURE E1. Levels of CXCL8, CCL2, CCL4, IL-5, GM-CSF, and G-CSF after drug stimulation of PBMCs from the patient (gray bar) and a control (black bar). VAN, vancomycin; CFZ, ceftazidime; CTX, ceftriaxone; MER, meropenem; CIP, ciprofloxacin; PHE, phenytoin; CM, culture medium.

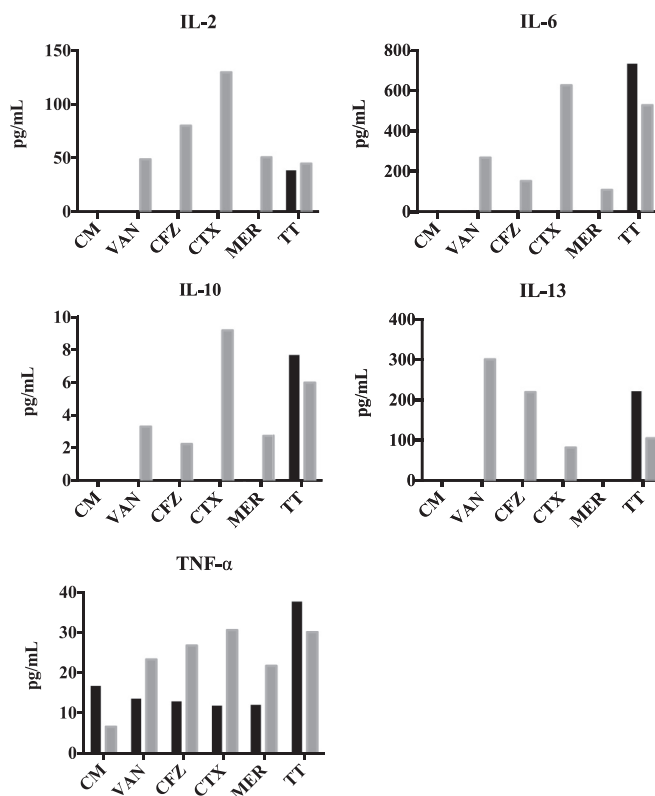


FIGURE E2. Levels of IL-2, IL-6, IL-10, IL-13, and TNF- α after drug stimulation of PBMCs from the patient (*gray bar*) and a control (*black bar*). Control was tolerant to vancomycin and ceftazidime. VAN, vancomycin; CFZ, ceftazidime; CTX, ceftriaxone; MER, meropenem; CIP, ciprofloxacin; PHE, phenytoin; CM, culture medium.

were tested for IFN- γ with the use of a commercial ELISA kit (e-Biosciences, San Diego, Calif) according to the manufacturer's instructions. Concentration in each triplicate was calculated from the standard curve, and results were expressed as means and SDS for each sample. A positive result was defined as detection of IFN- γ in presence of a drug at a level above the mean plus two SDs of all controls as previously described.^{E3} This level was calculated at 13.2 pg/mL in the present study.

Culture supernatant fluids were also tested in a multiplex human cytokine/chemokine assay kit (Affymetrix, Procarta, Santa Clara, Calif) according to the manufacturer's instructions. Concentrations of eotaxin, G-CSF, GM-CSF, IL-1 β , IL-2, IL-4, IL-5, IL-6, IL-7,

IL-10, IL-12, IL-13, IL-17A, TNF- α , CXCL8, CCL2, CCL4, and vascular endothelial growth factor were thus determined.

REFERENCES

- E1. Pichler WJ, Tilch J. The lymphocyte transformation test in the diagnosis of drug hypersensitivity. *Allergy* 2004;59:809-20.
- E2. Daubner B, Groux-Keller M, Hausmann OV, Kawabata T, Naisbitt DJ, Park BK, et al. Multiple drug hypersensitivity: normal Treg cell function but enhanced in vivo activation of drug-specific T cells. *Allergy* 2012;67:58-66.
- E3. Halevy S, Cohen AD, Grossman N. Clinical implications of in vitro drug-induced interferon gamma release from peripheral blood lymphocytes in cutaneous adverse drug reactions. *J Am Acad Dermatol* 2005;52: 254-61.