

Trimethoprim-sulfamethoxazole induces acute pancreatitis associated with drug-specific cytotoxic T lymphocytes

Minyoung Jung, MD^{a,b,*}, Jihyun Kim, MD, PhD^{a,b,*},
Ji Young Lee, MD, PhD^c, Minji Kim, MD^d,
Seung-Hyun Kim, MD, PhD^e, and
Kangmo Ahn, MD, PhD^{a,b}



Clinical Implications

- With a diagnosis of drug-induced acute pancreatitis, it is difficult to establish causality. We report a case of a pediatric patient who experienced trimethoprim/sulfamethoxazole-induced pancreatitis confirmed through both *in vivo* and *in vitro* tests.

TO THE EDITOR:

Drug-induced acute pancreatitis (DIAP) is a rare adverse reaction. More than 100 drugs are known to cause DIAP, and they are classified into 5 categories (Ia, Ib, II, III, IV) on the basis of available case reports¹ due to the difficulty in establishing causality. Although the pathogenesis of DIAP is not well understood, potential mechanisms include structural damage, toxins, metabolic changes, ischemia, and immune-mediated reactions.² Trimethoprim/sulfamethoxazole (TMP/SMX) is commonly used for the treatment of urinary tract infections and *Pneumocystis jiroveci* pneumonia in children. Among the adverse events associated with TMP/SMX, acute pancreatitis (AP) has been rarely reported in children.^{3,4} However, the diagnosis is typically not confirmed using a controlled challenge test, and the exact mechanism is not usually reported.² Herein, we describe a pediatric patient with TMP/SMX-induced AP, which was confirmed using both *in vivo* and *in vitro* tests.

A 12-year-old girl was referred to our hospital for the evaluation of severe abdominal pain and vomiting. She was initially treated with amoxicillin/clavulanate and TMP/SMX for 1 week under the diagnosis of acute rhinosinusitis. The amoxicillin/clavulanate was then changed to clarithromycin because of a poor response to the previous antibiotics. After 5 days, the patient manifested abdominal pain, high fever, frequent vomiting, steatorrhea, and lip edema. She had neither notable medical history nor family history of allergic diseases.

On hospitalization, her blood pressure was 98/52 mm Hg with a heart rate of 87 beats/min, a respiratory rate of 20 beats/min, and a body temperature of 38.2°C. On palpation, tenderness was found in the right upper quadrant of the abdomen with the presence of muscle guarding. There was no hepatosplenomegaly or jaundice. A blood test revealed a white blood cell count of 14,430/mm³ (reference, 4,000-10,500/mm³), a C-reactive protein level of 12.54 mg/dL (reference, 0.08-0.76 mg/dL), aspartate aminotransferase/alanine aminotransferase levels of 117/138 U/L (reference: aspartate aminotransferase, 10-40 U/L; alanine aminotransferase, 5-45 U/L),

total/direct bilirubin levels of 3.9/3.0 mg/dL (reference: total bilirubin, 0-1 mg/dL; direct bilirubin, 0-0.2 mg/dL), a calcium level of 8.4 mg/dL (reference, 8.8-10.8 mg/dL), and a triglyceride level of 54 mg/dL (reference, 41-138 mg/dL). The serum levels of amylase and lipase were 522 and 1160 U/L (reference: amylase, 30-100 U/L; lipase, 145-216 U/L), respectively. No respiratory virus was detected in the nasopharyngeal aspirate, and there was no bacterial growth in the blood culture. Ultrasonography showed mild pancreatic enlargement without evidence of gallstones or a biliary tract obstruction. Because DIAP was highly suspected, possible etiologic drugs, such as TMP/SMX (class Ia) and clarithromycin (class III), were withheld. The patient was not allowed anything per the oral route and received intravenous hydration. Fever and angioedema disappeared 2 days later. Four days after the discontinuation of the drugs, her abdominal pain subsided along with a decrease in the levels of serum amylase and lipase, and she was discharged.

To identify the exact cause among 3 different antibiotics and to prevent another attack, she was readmitted for a provocation test after informed consent was obtained. The patient was challenged with clarithromycin first, but no reactions occurred. She was then challenged with TMP/SMX via the oral route. The starting dose was 2/10 mg, which was gradually increased at 1-hour intervals to a final dose of 160/800 mg. Four hours after administration of the final dose, the patient developed fever of 38°C, vomiting, and epigastric pain. The serum amylase and lipase levels were 101.8 and 65.4 U/L, increasing from the baseline amylase and lipase levels of 83.9 and 31.4 U/L, respectively. The next day, fever and epigastric pain continued and the levels of amylase and lipase were markedly elevated to 572 and 1416 U/L, respectively, although the patient was fasting and received intravenous hydration. Her epigastric pain and increased levels of amylase and lipase recovered 3 days after the drug challenge test. The Naranjo Adverse Drug Reaction Probability Scale⁵ scored 9 points, which indicates a definite diagnosis of a TMP/SMX-induced adverse reaction. After 6 months, we performed amoxicillin challenge test to the case patient, and the results was negative.

We performed 3 *in vitro* tests using peripheral blood samples from the patient and 5 healthy donors as the negative controls: (1) a lymphocyte transformation test (LTT) to examine drug-specific T-cell activation,⁶ (2) enzyme-linked immunoSpot assay (AID Diagnostika GmbH, Strassberg, Germany) to measure cytolytic granzyme B secretion,⁶ and (3) flow cytometry measurement of CD107a, a lysosomal-associated membrane glycoprotein-1,⁷ under drug stimulation. This study was approved by the Institutional Review Board of Samsung Medical Center (Samsung Medical Center institutional review board file no. 2016-02-094). In LTT using ³H-thymidine incorporation assay, lymphocyte proliferation (9.37-fold increase) was strongly enhanced in the concomitant incubation of TMP (10 µg/mL) and SMX (25 µg/mL) with PBMCs from the patient (Figure 1, A). In contrast, a single drug stimulation did not show similar effect; 3-fold increase in proliferation by SMX alone at 100 µg/mL and no response to TMP alone at 10 µg/mL. Granzyme B-secreting PBMCs were detected on 48 hours of TMP/SMX stimulation in the patient's sample (230 spots per 5 × 10⁵ cells)

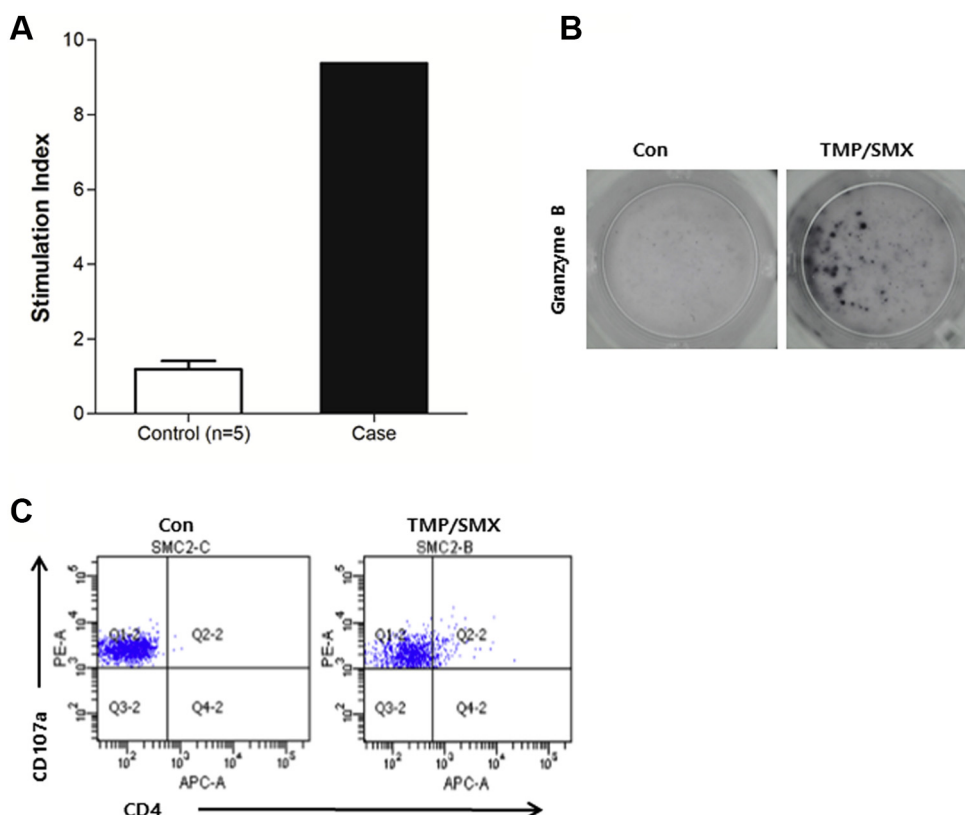


FIGURE 1. *In vitro* tests. **A**, Proliferation of PBMCs against TMP/SMX (drug stimulation, 6 days) measured with [³H] thymidine incorporation assay. The stimulation index is the ratio of counted radioactivity of PBMC culture with and without TMP/SMX. **B**, Granzyme B secretion by PBMCs using enzyme-linked immunoSpot (TMP/SMX stimulation, 48 hours). **C**, CD107a expression on T cells measured using flow cytometry.

(Figure 1, B), whereas less effect was found under single stimulation with SMX at 25 µg/mL (56 spots) and TMP at 10 µg/mL (26 spots). The cytotoxic activity of T cells after drug stimulation was also assessed by measuring CD107a using flow cytometry. Surface expression of CD107a on both CD3⁺CD4⁺ and CD3⁺CD4⁻ T cells from the patient was further increased under TMP/SMX stimulation compared with the control group (Figure 1, C).

The diagnosis of drug hypersensitivity is usually done using a detailed clinical history followed by skin or drug provocation tests. As a complementary approach to the diagnosis of delayed hypersensitivity reactions, it is helpful to use *in vitro* tests.⁶ Moreover, these tests can provide insights into the pathophysiology of the drug allergy, especially in various drug-induced skin diseases such as Stevens-Johnson syndrome.⁷ However, there is a lack of studies regarding the sensitivity and specificity of the above *in vitro* test and no established standard protocol in TMP/SMX-induced hypersensitivity reactions.⁶

In this case, a relapse of AP was confirmed after the provocation test with TMP/SMX. The acute reaction was reversible and resolved after discontinuation of the drug. From 3 *in vitro* tests showing uniformly a T-cell activation and releasing of cytotoxic molecules (granzyme and granzysin), we can deduce that TMP/SMX stimulation leads to differentiation of drug-specific T cells and apoptosis of the target cells by cytotoxic molecules (type IV hypersensitivity),⁸ although the

infiltration of drug-specific T cells in the pancreas was not confirmed. Because of the limited number of patients with DIAP, the mechanistic explanation of how drugs can stimulate T cells, that is, allergic/immune reactions, pharmacological interaction with immune receptors, or pseudoallergy,⁹ could not be demonstrated. Further investigations using drug-specific T-cell clones are needed for understanding the pathophysiology of DIAP.

In conclusion, we report a pediatric patient with TMP/SMX-induced AP, which may be mediated by drug-specific cytotoxic T cells. DIAP should be suspected when a child manifests acute abdominal pain during the administration of TMP/SMX. In addition, LTT, granzyme B enzyme-linked immunoSpot assays, and flow cytometry measurement of CD107a are helpful to confirm DIAP.

^aDepartment of Pediatrics, Samsung Medical Center, Sungkyunkwan University School of Medicine, Seoul, Korea

^bEnvironmental Health Center for Atopic Diseases, Samsung Medical Center, Seoul, Korea

^cDepartment of Pediatrics, Hallym University Hangang Sacred Heart Hospital, Seoul, Korea

^dDepartment of Pediatrics, Dongtan Sacred Heart Hospital, Hallym University of Medicine, Dongtan, Korea

^eTranslational Research Laboratory for Inflammatory Disease, Clinical Trial Center, Ajou University Medical Center, Suwon, Korea

*These authors contributed equally to this study.

This research was supported by the Korea Health Technology R&D Project through the Korea Health Industry Development Institute grant funded by the Ministry of Health & Welfare, Republic of Korea (grant no. HI16C0992).

Conflicts of interest: The authors declare that they have no relevant conflicts of interest.

Received for publication March 25, 2018; revised May 30, 2018; accepted for publication June 11, 2018.

Available online June 22, 2018.

Corresponding author: Kangmo Ahn, MD, PhD, Department of Pediatrics, Samsung Medical Center, Sungkyunkwan University School of Medicine, 81 Irwon-ro, Gangnam-gu, Seoul 06351, Korea. E-mail: kmaped@gmail.com. Or: Seung-Hyun Kim, MD, PhD, Translational Research Laboratory for Inflammatory Disease, Clinical Trial Center, Ajou University Medical Center, 164 World cup-ro, Youngtong-gu, Suwon, Korea. E-mail: kimsh@ajou.ac.kr.

2213-2198

© 2019 American Academy of Allergy, Asthma & Immunology

<https://doi.org/10.1016/j.jaip.2018.06.009>

REFERENCES

1. Badalov N, Baradaran R, Iswara K, Li J, Steinberg W, Tenner S. Drug-induced acute pancreatitis: an evidence-based review. *Clin Gastroenterol Hepatol* 2007;5:648-61.
2. Hung WY, Abreu Lanfranco O. Contemporary review of drug-induced pancreatitis: a different perspective. *World J Gastrointest Pathophysiol* 2014; 5:405-15.
3. Dickey SE, Mabry WA, Hamilton LA. Possible sulfamethoxazole/trimethoprim-induced pancreatitis in a complicated adolescent patient posttraumatic injury. *J Pharm Pract* 2015;28:419-24.
4. Kandula L, Lowe ME. Etiology and outcome of acute pancreatitis in infants and toddlers. *J Pediatr* 2008;152:106-110, 10.e1.
5. Farcas A, Bojita M. Adverse drug reactions in clinical practice: a causality assessment of a case of drug-induced pancreatitis. *J Gastrointest Liver Dis* 2009;18:353-8.
6. Mayorga C, Celik G, Rouzaire P, Whitaker P, Bonadonna P, Rodrigues-Cernadas J, et al. In vitro tests for drug hypersensitivity reactions: an ENDA/EAACI Drug Allergy Interest Group position paper. *Allergy* 2016;71:1103-34.
7. Zawodniak A, Lochmatter P, Yerly D, Kawabata T, Lerch M, Yawalkar N, et al. In vitro detection of cytotoxic T and NK cells in peripheral blood of patients with various drug induced skin diseases. *Allergy* 2010;65:376-84.
8. Pichler WJ, Naisbitt DJ, Park BK. Immune pathomechanism of drug hypersensitivity reactions. *J Allergy Clin Immunol* 2011;127:S74-81.
9. Pichler WJ, Hausmann O. Classification of drug hypersensitivity into allergic, p-i, and pseudo-allergic forms. *Int Arch of Allergy and Immunol* 2016;171:166-79.