〈Research Report〉

Comprehensive analysis of antibiotic-induced agranulocytosis using the Japanese adverse drug event report database

Yuki Asai, Takanori Yamamoto and Yasuharu Abe

Pharmacy, National Hospital Organization Mie Chuo Medical Center

(Received for publication July 1, 2021)

Although infrequent, drug-induced agranulocytosis can be caused by antibiotics. Here, we analyzed the Japanese Adverse Drug Event Report (JADER) database to identify profiles of antibiotic-induced agranulocytosis.

We analyzed reports of agranulocytosis from April 2004 to January 2021 from the JADER database. The reporting odds ratio and 95% confidence interval were used to detect agranulocytosis signals. We evaluated the time-to-onset profile and hazard type using the Weibull shape parameter.

Ten out of 60 antibiotics showed signals for agranulocytosis; the reporting odds ratios (95% confidence intervals) for ampicillin/sulbactam, amikacin, cefmetazole, cefozopran, clindamycin, ciprofloxacin, imipenem/cilastatin, kanamycin, teicoplanin, and vancomycin were 2.65 (1.79–3.80), 2.94 (1.91–4.34), 4.48 (2.27–6.92), 2.77 (1.88–3.95), 1.64 (1.04–2.47), 2.01 (1.40–2.82), 2.78 (2.11–3.60), 6.05 (2.16–13.7), 2.05 (1.31–3.07), and 3.54 (2.73–4.54), respectively. The median times-to-onset of agranulocytosis for ampicillin/sulbactam, cefmetazole, cefozopran, clindamycin, imipenem/cilastatin, kanamycin, teicoplanin, and vancomycin were 20, 6, 10, 16, 12, 3, 18, and 13 days, respectively. The 95% confidence intervals of the Weibull shape parameter β for these antibiotics were over and excluded 1, indicating that the antibiotics were the wear out failure type.

We identified 10 antibiotics that may be associated with high risk of agranulocytosis, suggesting that absolute neutrophil counts in patients taking these drugs should be monitored carefully in the clinical setting.

Corresponding author: Yuki Asai, Ph.D., Pharmacy, National Hospital Organization Mie Chuo Medical Center; 2158–5 Hisaimyojincho, Tsu, Mie 514–1101, Japan, Tel: +81–59–259–1211, Fax: +81–59–256–2651, E-mail: yuki0715asai@gmail.com

Introduction

Neutropenia is characterized as a decrease in the absolute neutrophil count, categorized as mild (1,000–1,500/μL), moderate (500–1,000/μL), or severe ($<$ 500/μL)[1]. The most severe form, called agranulocytosis, is a life-threating condition in which the patient becomes highly susceptible to infectious diseases and shows a mortality rate of 10–20%[2]. Agranulocytosis can be caused by a wide range of medications, including clozapine[3] and antibiotics[1], and infection control becomes particularly difficult in patients with antibiotic-induced agranulocytosis. Furthermore, in patients who continue to have a fever, it is necessary to switch to another antibiotic and add concomitant treatment with granulocyte-colony-stimulating factor[4]. Accordingly, there is an urgent need to identify antibiotics that have a low risk of inducing agranulocytosis.

Although the mechanisms of antibiotic-induced agranulocytosis remain unknown, Weitzman and Stossel[5] speculated that antibiotic-induced agranulocytosis may be caused by opsonizing antineutrophil antibodies, i.e., an IgG or IgM immune-mediated hypersensitivity reaction. However, because agranulocytosis is a rare adverse event (AE)[6], it is difficult to collect a sufficient number of samples to conduct clinical studies. Recently, spontaneous reporting systems (SRSs) have been shown to be useful for detection of rare AEs and used as primary tools in post marketing surveillance. In Japan, individual AEs are collected by healthcare workers at the Pharmaceuticals and Medical Devices Agency (PMDA). The Japanese Adverse Drug Event Report (JADER) database, which is available online, was established in April 2004 by the PMDA and reflects the realities of clinical practice[7]. Thus, data mining using the JADER database can be used as a screening tool for antibiotic-induced agranulocytosis.

In this study, we evaluated the expression profiles of antibiotic-induced agranulocytosis using the JADER database.

Materials and Methods

Data source

The JADER database, including data from April 2004 to January 2021, was searched from the PMDA website (https://www.info.pmda.go.jp/fukusayoudb/CsvDownload.jsp). The JADER database consisted of four data tables, as follows: 1) DEMO (patients' demographic information, including sex, age, and weight); 2) DRUG (drug name, causality, etc.); 3) REAC (AEs, outcomes, etc.); and 4) HIST (medical history, primary illness, etc.). We created relational database from the four data tables according to patient identification numbers using Microsoft Access 2019 (Microsoft, Redmond, WA, USA). Because duplicate reports in the SRS database may affect the evaluation from the relational database, we extracted only the latest of AE reports based on the DEMO file. Data with inaccurate dates were excluded.

Drugs and AE names

The 60 antibiotics analyzed in the current study are shown in Table 1. We focused on agranulocytosis, the most life-threatening type of neutropenia. Agranulocytosis was defined using the preferred term (PT) code (10001507) according to the Medical Dictionary for Regulatory Activities version 23.1J.

Data analysis

We selected the reporting odds ratio (ROR) for the detection of signals associated with antibiotic-induced agranulocytosis. As shown Fig. 1, the ROR and 95% confidence interval (95% CI) were calculated from a two-by-two contingency table. Signal detection was defined as follows: ROR greater than 1, lower limit of the corresponding 95% CI greater than 1, and number of cases three or more[8]. Conversely, the condition of ROR less than 1 was considered no exposure-event association. The adjusted odds ratio was not calculated by multivariate analysis because the data mining by JADER cannot evaluate risk factors for the development of agranulocytosis. Statical analysis was conducted with SPSS Statistics version 27 (IBM Japan, Tokyo, Japan). In addition, the clinical outcomes from agranulocytosis develops were investigated for signal-detected antibiotics.

The median of period until agranulocytosis onset from the time of first administration for each patient was evaluated using the Weibull shape parameter (WSP), which does not require a reference population[9]. The elapsed time from the first administration was calculated by subtracting the first administration date from the agranulocytosis expression date. Fifty cases were excluded because the date of first administration or onset of agranulocytosis was not available. The combination of WSP β and 95% CI could be interpreted as follows: when β was equal to 1 (random failure) and 95% CI of β included the 1, the hazard was estimated to be constant over time; if β was greater than 1 (wear out failure) and the 95% CI (lower) of β exceeded 1, the hazard was considered to increase over time; if β was less than 1 (early failure) and the 95% CI (upper) of β was less than 1, the hazard was considered to increase at an early stage and then decrease. Time-to-onset analyses were performed using JMP version 15 software (SAS Institute, Cary, NC, USA).

Ethics approval and consent to participate

Ethics approval and consent to participate were not applicable to this study because we used publicly available voluntarily provided reports.

Results

As shown in Fig. 2, we extracted 72,723 reports of suspected AEs related to antibiotic treat-

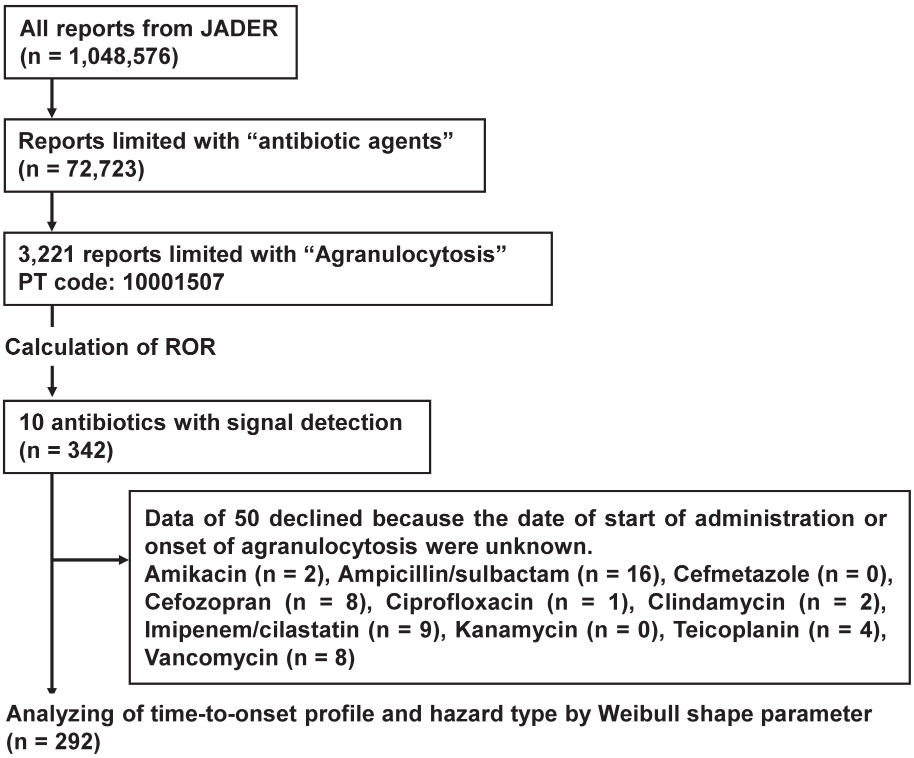
Fig. 1. Two-by-two contingency table for the calculation of reporting odds ratios

	Adverse Drug Event of Agranulocytosis	Other Adverse Drug Event
Target antibiotics	a	b
Other antibiotics	c	d
Total	a+c	b+d

$$ROR = \frac{a \times d}{b \times c} \quad SE = \sqrt{\frac{1}{a} + \frac{1}{b} + \frac{1}{c} + \frac{1}{d}}$$

$$95\%CI = \exp[\log(ROR) \pm 1.96 \times SE\{\log(ROR)\}]$$

Fig. 2. Flowchart for the construction of data analysis tables from the JADER database



ment among the 1,048,576 reports in the JADER database between April 2004 and January 2021. The number of reports of agranulocytosis was 3,221. As shown in Table 1, 10 out of 60 antibiotics were detected as signals; the RORs (95% CIs) for ampicillin/sulbactam, amikacin, cefmetazole, cefozopran, clindamycin, ciprofloxacin, imipenem/cilastatin, kanamycin, teicoplanin, and vancomycin were 2.65 (1.79–3.80), 2.94 (1.91–4.34), 4.48 (2.27–6.92), 2.77 (1.88–3.95),

Table 1. Number of reports and reporting odds ratios of agranulocytosis associated with antibiotics

Group	Antibiotics	Total (n)	Case (n)	Non-case (n)	ROR	95%CI
Anti-MRSA agents	Arbekacin	435	3	432	0.74	0.15–2.19
	Linezolid	74	0	74	NA	0.00–5.48
	Teicoplanin	1,356	25	1,331	2.05	1.31–3.07
	Vancomycin	2,428	72	2,356	3.54	2.73–4.54
Anti-fungal agents	Amphotericin B	1,055	0	1,055	NA	0.00–0.37
	Fluconazole	2,960	22	2,938	0.80	0.49–1.22
	Flucytosine	22	0	22	NA	0.00–19.6
	Fos-fluconazole	2,002	24	1,978	1.31	0.83–1.97
	Itraconazole	1,646	4	1,642	0.26	0.07–0.66
	Ketoconazole	125	0	125	NA	0.00–3.21
	Micafungin	2,482	3	2,479	0.13	0.03–2.40
	Miconazole	351	0	351	NA	0.00–1.13
	Voriconazole	378	0	378	NA	0.00–1.04
Aminoglycoside	Amikacin	1,037	27	1,010	2.94	1.91–4.34
	Dibekacin	9	0	9	NA	0.00–54.3
	Gentamicin	467	4	463	0.92	0.25–2.40
	Kanamycin	113	6	107	6.05	2.16–13.7
	Streptomycin	183	0	183	NA	0.00–2.18
Quinolone	Ciprofloxacin	2,005	36	1,969	2.01	1.40–2.82
	Gatifloxacin	673	0	673	NA	0.00–0.58
	Levofloxacin	4,267	15	4,252	0.36	0.20–0.61
	Norfloxacin	111	0	111	NA	0.00–3.62
	Pazufloxacin	1,072	3	1,069	0.30	0.06–1.34
	Tosufloxacin	448	1	447	0.24	0.01–0.92
Macrolide	Azithromycin	1,508	5	1,503	0.35	0.11–0.83
	Clarithromycin	3,921	17	3,904	0.45	0.26–0.73
	Erythromycin	651	1	650	0.16	0.004–0.92
Carbapenem	Imipenem/cilastatin	2,734	65	2,669	2.78	2.11–3.60
	Doripenem	18	0	18	NA	0.00–24.41
	Biapenem	535	4	531	0.81	0.22–2.08
	Meropenem	3,651	33	3,618	0.98	0.67–1.39
Tetracycline	Doxycycline	23	0	23	NA	0.00–18.67
	Minocycline	1,908	25	1,883	1.44	0.92–2.15
Anti-viral agents	Acyclovir	2,012	8	2,004	0.42	0.18–0.84
	Valaciclovir	1,927	1	1,926	0.05	0.00–10.30
Penicillin	Amoxicillin	805	1	804	0.13	0.003–0.74
	Amoxicillin/clavulanate	59	0	59	NA	0.00–6.92
	Ampicillin	341	0	341	NA	0.00–1.16
	Ampicillin/sulbactam	1,365	32	1,333	2.65	1.79–3.80
	Benzylpenicillin	18	0	18	NA	0.00–24.43
	Faropenem	187	1	186	0.58	0.01–3.26
	Piperacillin	1,670	14	1,656	0.90	0.50–1.53
	Piperacillin/tazobactam	535	6	529	1.22	0.44–2.68
Cephalosporin	Cefaclor	439	3	436	0.74	0.15–2.17
	Cefazolin	1,446	14	1,432	1.05	0.57–1.78
	Cefcapene	2,191	4	2,187	0.19	0.05–0.49
	Cefditoren	541	2	539	0.40	0.01–1.44
	Cefepime	1,572	9	1,563	0.61	0.28–1.17
	Cefmetazole	561	22	539	4.48	2.77–6.92
	Cefoperazone/sulbactam	1,565	12	1,553	0.82	0.43–1.45
	Cefotaxime	259	3	256	1.26	0.26–3.73
	Cefotiam	1,884	18	1,866	1.03	0.61–1.65
	Cefozopran	1,351	33	1,318	2.77	1.88–3.95
	Ceftazidime	1,408	5	1,403	0.38	0.12–0.89
	Cefteram	156	2	154	1.39	0.17–5.13
	Ceftriaxone	1,415	12	1,403	0.91	0.47–1.61
	Cephalexin	67	0	67	NA	0.00–6.07
	Flomoxef	959	11	948	1.25	0.62–2.26
Lincomycin	Clindamycin	1,612	24	1,588	1.64	1.04–2.47
Other	Trimethoprim/sulfamethoxazole	5,730	47	5,683	0.88	0.64–1.18

NA; not available. MRSA: methicillin-resistant *Staphylococcus aureus*. ROR: reporting odds ratio. 95%CI: 95% confidence interval.

Table 2. Clinical outcomes after the onset of antibiotic-induced agranulocytosis

	Total (n)	Outcomes (n)			
		Recovery	Unrecovered	Death	Unknown
Amikacin	27	27	0	0	0
Ampicillin/sulbactam	32	29	0	3	0
Cefmetazole	22	19	0	2	1
Cefozopran	33	26	1	6	0
Ciprofloxacin	36	21	0	15	0
Clindamycin	24	22	0	2	0
Imipenem/cilastatin	65	58	0	7	0
Kanamycin	6	3	0	3	0
Teicoplanin	25	25	0	0	0
Vancomycin	72	71	0	1	0

Table 3. Time-to-onset analysis of antibiotics using Weibull distributions

Antibiotics	Case (n)	Median Day (25–75%)	Scale parameter: α		Scale parameter: β		Pattern
			α	95% CI	β	95% CI	
Amikacin	25	2 (1–3)	6.76	3.87–11.44	0.80	0.59–1.04	Random failure
Ampicillin/sulbactam	16	20 (14–29)	23.37	18.7–28.8	2.57	1.66–3.70	Wear out failure
Cefmetazole	22	6 (4–8)	6.59	6.62–7.68	2.95	2.09–3.90	Wear out failure
Cefozopran	25	10 (8–17)	12.55	9.32–12.65	1.49	1.03–2.06	Wear out failure
Ciprofloxacin	35	4 (1–8.5)	7.32	4.73–11.10	0.85	0.64–1.07	Random failure
Clindamycin	22	16 (10–23)	18.32	15.6–21.3	2.98	2.06–4.12	Wear out failure
Imipenem/cilastatin	56	12 (9–23)	16.56	14.16–19.27	1.81	1.43–2.24	Wear out failure
Kanamycin	6	3 (2–4)	3.36	2.47–4.48	3.46	1.61–6.35	Wear out failure
Teicoplanin	21	18 (6–20)	16.49	12.76–19.53	2.74	1.82–3.95	Wear out failure
Vancomycin	64	13 (9–20)	17.42	15.40–19.63	2.17	1.78–2.59	Wear out failure

$0<\beta<1$, early failure; $\beta=1$, random failure; $\beta>1$, wear out failure.
95% CI: 95% confidence interval.

1.64 (1.04–2.47), 2.01 (1.40–2.82), 2.78 (2.11–3.60), 6.05 (2.16–13.7), 2.05 (1.31–3.07), and 3.54 (2.73–4.54), respectively. The clinical outcomes after the onset of antibiotic-induced agranulocytosis were evaluated (Table 2). Most antibiotics were found to be recovered from agranulocytosis.

For the time-to-onset analysis, we extracted 292 cases for the 10 antibiotics with signal detection (Table 3). The median durations (interquartile ranges) of agranulocytosis onset following treatment with amikacin, cefmetazole, ciprofloxacin, and kanamycin were within 1 week of the first treatment. Although the 95% CIs of WSP β for eight antibiotics (excluding amikacin and ciprofloxacin) were over and excluded 1, indicating a wear out failure type, the profiles of amikacin and ciprofloxacin were consistent with the random failure type (Table 3).

Discussion

Since the occurrence of agranulocytosis is rare, retrospective study are difficult to evaluate the profiles of antibiotic-induced agranulocytosis. Therefore, data mining may be appropriate method as a pilot study for developing antibiotic-induced agranulocytosis.

Because neutrophils are produced in bone marrow, we speculated that antibiotics with high migration to bone marrow may have a high risk of inducing agranulocytosis before data mining. However, signals were not detected for antibiotics with high cerebrospinal fluid transferability, such as ceftriaxone[10] and cefotaxime[11], whereas teicoplanin[12], an antibiotic with poor cerebrospinal fluid transferability, was detected. Therefore, these findings suggested that the risk of agranulocytosis could not be clarified by analysis of migration to the bone marrow only. A retrospective study reported that the development of vancomycin-induced neutropenia was not associated with the dosage, trough concentration, or administration period[13]. Although other antibiotics also need to be investigated, antibiotic-induced agranulocytosis may not be a cumulative toxicity.

The incidence of neutropenia induced by vancomycin is approximately 2–18%[14,15]. In contrast, Smith *et al.* reported that linezolid, an anti-methicillin-resistant *Staphylococcus aureus* (MRSA) agent, may be safe and effective for the treatment of febrile neutropenia[16]. In addition, carbapenems targeting *Pseudomonas aeruginosa* are empirically used for the treatment of febrile neutropenia[17], indicating that when using antibiotics with anti-*Pseudomonas aeruginosa* or anti-MRSA activity in the empirical treatment of high-risk patients, such as those with febrile neutropenia, it may be better to preferentially use antibiotics with the same spectrum for which no signal was detected in this study.

In the clinical setting, aminoglycosides are often administered with *β*-lactam antibiotics to induce synergistic effects[18,19]. Moreover, infective endocarditis caused by MRSA may be treated with vancomycin or teicoplanin in combination with aminoglycosides[20]. In the current study, because amikacin and kanamycin were detected as signals, we speculate that it may be better to use gentamicin as the aminoglycoside in combination therapy.

It was suggested that even if agranulocytosis develops, it may be recovered by discontinuing the causative drug (Table 2). Because the data mining method using the JADER database does not include the number of all patients who received antibiotics, the recovery rate of agranulocytosis cannot be compared among antibiotics. On the other hand, based on analysis using medical receipt data, the recovery rate can be calculated and may be compared among antibiotics. This should be considered to be an area for future study.

In recent reports of SRS data mining, WSP analysis has been performed to evaluate the time-to-onset data for target AEs[9]. Accordingly, time-to-onset analysis using WSP is likely to be a useful tool for determination of the specific safety monitoring period for agranulocytosis. The mechanisms of agranulocytosis involve immune responses toward opsonizing antineutrophil antibodies[5] or drugs themselves, suggesting that the differences in the time-of-onset of agranulocytosis may occur through different mechanisms.

Data mining using the JADER database has some limitations that should be considered because the JADER database has several biases. First, because the JADER database does not contain information for control patients, the intensity of RORs cannot be quantified and compared among antibiotics. Second, there are also some reporting biases, such as under-reporting and lack of data. Third, the number of reports was small and may have been underestimated because the PT codes 10018687 (granulocytopenia) and 10018681 (granulocyte count decreased) were not included in the current study. Fourth, because agranulocytosis may be induced by severe infections[21)], the agranulocytosis reported by SRS may not be caused by administration of antibiotics. Fifth, data mining methods cannot evaluate comorbidities that are involved in the risk of developing agranulocytosis.

In conclusion, this comprehensive analysis was the first report evaluating the incidences of antibiotic-induced agranulocytosis. Despite the various limitations of using the JADER database, we identified 10 antibiotics that may be associated with high risk of agranulocytosis, suggesting that absolute neutrophil counts in patients taking these drugs should be monitored carefully in the clinical setting. Finally, further clinical studies are needed to verify the mechanisms through which ampicillin/sulbactam, amikacin, cefmetazole, cefozopran, clindamycin, ciprofloxacin, imipenem/cilastatin, kanamycin, teicoplanin, and vancomycin may induce agranulocytosis. The difference in the onset time may have been related to the variations in pharmacokinetics of each antibiotic and the onset mechanism of agranulocytosis; however, the details are unknown, and further research is needed.

Acknowledgments

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1） Boxer LA: How to approach neutropenia. Hematology Am Soc Hematol Educ Program. 2012; 2012: 174–82.
2） Andrès E, Maloisel F, Kurtz JE, *et al.*: Modern management of non-chemotherapy drug-induced agranulocytosis: a monocentric cohort study of 90 cases and review of the literature. Eur J Intern Med. 2002; 13: 324–8.
3） Mijovic A, MacCabe JH: Clozapine-induced agranulocytosis. Ann Hematol. 2020; 99: 2477–82.
4） Genchanok Y, Tolu SS, Wang H, Arora S: Agranulocytosis from outpatient antimicrobial treatment with ceftriaxone: a case report. Perm J. 2019; 23: 18–190.
5） Weitzman SA, Stossel TP: Drug-induced immunological neutropenia. Lancet. 1978; 1: 1068–72.
6） Olaison L, Belin L, Hogevik H, Alestig K: Incidence of beta-lactam-induced delayed hypersensitivity and neutropenia during treatment of infective endocarditis. Arch Intern Med. 1999; 159: 607–15.
7） Noguchi Y, Ueno A, Otsubo M, *et al.*: A simple method for exploring adverse drug events in pa-

tients with different primary diseases using spontaneous reporting system. BMC Bioinformatics. 2018; 19: 124.

8) Hashiguchi M, Imai S, Uehara K, Maruyama J, Shimizu M, Mochizuki M: Factors affecting the timing of signal detection of adverse drug reactions. PLoS One. 2015; 10: e0144263.

9) Sauzet O, Carvajal A, Escudero A, Molokhia M, Cornelius VR: Illustration of the weibull shape parameter signal detection tool using electronic healthcare record data. Drug Saf. 2013; 36: 995–1006.

10) Kotani A, Hirai J, Hamada Y, Fujita J, Hakamata H. Determination of ceftriaxone concentration in human cerebrospinal fluid by high-performance liquid chromatography with UV detection. J Chromatogr B Analyt Technol Biomed Life Sci. 2019; 1124: 161–4.

11) Chen XK, Shi HY, Leroux S, *et al.*: Penetration of cefotaxime into cerebrospinal fluid in Neonates and Young Infants. Antimicrob Agents Chemother. 2018; 62: e02448-17.

12) Wilson AP: Clinical pharmacokinetics of teicoplanin. Clin Pharmacokinet. 2000; 39: 167–83.

13) Pai MP, Mercier RC, Koster SA: Epidemiology of vancomycin-induced neutropenia in patients receiving home intravenous infusion therapy. Ann Pharmacother. 2006; 40: 224–8.

14) Farber BF, Moellering RC Jr: Retrospective study of the toxicity of preparations of vancomycin from 1974 to 1981. Antimicrob Agents Chemother. 1983; 23: 138–41.

15) Morris A, Ward C: High incidence of vancomycin-associated leucopenia and neutropenia in a cardiothoracic surgical unit. J Infect. 1991; 22: 217–23.

16) Smith PF, Birmingham MC, Noskin GA, *et al.*: Safety, efficacy and pharmacokinetics of linezolid for treatment of resistant Gram-positive infections in cancer patients with neutropenia. Ann Oncol. 2003; 14: 795–801.

17) Khoo AL, Zhao YJ, Teng M, *et al.*: Evaluation of a risk-guided strategy for empirical carbapenem use in febrile neutropenia. Int J Antimicrob Agents. 2018; 52: 350–7.

18) Kurtz TO, Winston DJ, Bruckner DA, Martin WJ: Comparative in vitro synergistic activity of new beta-lactam antimicrobial agents and amikacin against Pseudomonas aeruginosa and Serratia marcescens. Antimicrob Agents Chemother. 1981; 20: 139–243.

19) Scudamore RA, Goldner M: Penetration of the outer membrane of Pseudomonas aeruginosa by synergistic combinations of beta-lactam and aminoglycoside antibiotics. Antimicrob Agents Chemother. 1982; 21: 1007–10.

20) Baddour LM, Wilson WR, Bayer AS, *et al.*: Infective endocarditis in adults: diagnosis, antimicrobial therapy, and management of complications: a scientific statement for healthcare professionals from the American Heart Association. Circulation. 2015; 132: 1435–86.

21) Shi R, Zhou Q, Fang R, Xiong X, Wang Q: Severe acute pancreatitis with blood infection by Candida glabrata complicated severe agranulocytosis: a case report. BMC Infect Dis. 2018; 18: 706.