Nilotinib-induced liver injury
A case report
Youwen Tan, PhD, MD∗, Yun Ye, PhD, Xingbei Zhou, PhD

Abstract
Introduction: Nilotinib is a selective inhibitor of the BCR-ABL tyrosine kinase receptor and is used in the management of chronic myelogenous leukemia (CML). Nilotinib therapy at high doses is associated with elevated serum bilirubin levels. If the serum bilirubin level exceeds 3 times the upper limit of normal, the recommendation is to either adjust nilotinib dosage or temporarily discontinue the treatment. However, it is unclear whether hyperbilirubinemia indicates obvious liver histology damage.

Patient concerns: A 24-year-old man with confirmed CML was treated with nilotinib therapy and developed hyperbilirubinemia after the treatment. Although the first remission of the hyperbilirubinemia was achieved after dose adjustment, the hematological parameters deteriorated. Thus, we initiated an antineoplastic therapy (at the standard dose) until complete remission of the CML was achieved. The pathogenic mechanism of hyperbilirubinemia may be related to the inhibition of uridine diphosphate-glucuronosyltransferase (UGT1A1) activity. Liver histological analysis revealed no significant liver damage. In addition, the patient had no family history of hyperbilirubinemia and liver disease.

Diagnosis: The patient was admitted to our hospital under the diagnosis of hyperbilirubinemia, and histopathology by liver biopsy showed no obvious damage. We also detected a UGT1A1 mutation [ex1 c.686C > A (p.Pro229Gln)] in the patient and his mother.

Interventions: When the nilotinib dose was decreased to 300 mg daily, the total bilirubin (TBIL) level decreased to 30 to 50 μmol/L for 1 month. However, because the Bcr-Abl/Abi ratio did not correspond to the major molecular response (MMR; <0.1%), the nilotinib dose was readjusted to 400 mg daily. One week later, the TBIL and indirect bilirubin levels increased to 89 and 79 μmol/L, respectively. The levels of alanine transaminase and other liver functional indicators were normal.

Outcomes: A Naranjo Adverse Drug Reaction (ADR) Probability Scale score of 13 indicates that hyperbilirubinemia is attributed to ADR caused by nilotinib rather than by drug-induced liver injury.

Conclusion: Although reducing the nilotinib dose can alleviate the occurrence of hyperbilirubinemia, the effect of MMR is also reduced. Treatment of CML without dose adjustment or discontinuation of nilotinib therapy may be more advantageous.

Abbreviations: ALT = alanine transaminase, AST = aspartate aminotransferase, CCyR = complete cytogenetic response, CML = chronic myelogenous leukemia, MMR = major molecular response, TBIL = total bilirubin, UGT1A1 = uridine diphosphate-glucuronosyltransferase, ULN = upper limit of normal.

Keywords: drug-induced liver injury, hyperbilirubinemia, nilotinib

1. Introduction
Nilotinib is a selective inhibitor of the BCR-ABL tyrosine kinase receptor[1] and is used to treat Philadelphia chromosome, which is associated with chronic myelogenous leukemia (CML).[2] Elevated serum aminotransferase levels are common in nilotinib therapy; however, only 4% to 9% of patients have been reported with levels 5 times higher than the upper limit of normal (ULN).[3] Nilotinib therapy at high doses is also associated with elevated serum bilirubin level.[4] If the alanine transamnise (ALT) or aspartate aminotransferase (AST) level increase significantly (i.e., if the ALT or AST level continues to be greater than 5 times the ULN, or the bilirubin level exceeds 3 times the ULN), the recommendation is to either adjust or temporarily discontinue the nilotinib dosage and initiate low-dose therapy.[5] We recently encountered a case of CML treated with nilotinib therapy, which led to hyperbilirubinemia. Although the first remission of the hyperbilirubinemia was achieved after dose adjustment, the hematological parameters deteriorated. Thus, we initiated an antineoplastic therapy (standard dose) until complete remission...
of the CML was achieved. The pathogenic mechanism of hyperbilirubinemia may be related to the inhibition of uridine diphosphate-glucuronosyltransferase (UGT1A1) activity. Liver histological analysis revealed no significant liver damage.

2. Ethics Statement

Ethics statement is not applicable for case reports according to the medical ethics committee of the Third Hospital of Zhenjiang Affiliated Jiangsu University, but informed consent was obtained from the patient for publication of this case report and the accompanying images. The study was conducted in accordance with the Declaration of Helsinki.

3. Case report

A 24-year-old man was admitted to our hospital with a complaint of abdominal pain. His white blood cell count at admission was $3.61 \times 10^9/L$. CML was suspected and later confirmed on the basis of the bone marrow biopsy result and the expression of Philadelphia chromosome (Ph+) on the cytogenetic study. First, 400mg of imatinib mesylate (IM; Glivec, Novartis, Basel, Switzerland) daily was prescribed. The dose was reduced by 1 tablet daily when the patient first complained of leg ache. A rapid decline was observed in the platelet count from $231 \times 10^9/L$ to $54 \times 10^9/L$ in 20 days, and the dose was reduced by 2 tablets when the platelet count declined to $34 \times 10^9/L$. After a month since the discontinuation of the nilotinib treatment, the Bcr-Abl/Abl ratio according to the international scale (IS) increased from 0.8% to 1%, and the nilotinib therapy (400mg daily, twice a day; Tasigna; Novartis, Basel, Switzerland) was reinitiated.

The findings from the liver function test performed before treatment were as follows: ALT, 32 U/L (normal range, 4–40 U/L); AST, 25 U/L (4–40 U/L); total bilirubin (TBIL), 12.6 μmol/L (3.4–21.2 μmol/L); direct bilirubin, 6.4 μmol/L (0.8–5 μmol/L); albumin, 3.5 g/dL (3.5–5.5 g/dL); prothrombin time, 12 s (9–14 s); alkaline phosphatase (ALP), 46 U/L (35–105 U/L); and γ-glutamyl transpeptidase, 32 U/L (normal, 5–36 U/L; Fig. 1).

After 4 weeks of nilotinib treatment, the TBIL level increased to 91 μmol/L. When the nilotinib dose was decreased to 300mg daily, the TBIL level decreased to 32 μmol/L, which was maintained at approximately 30 to 50 μmol/L for 1 month. However, because the Bcr-Abl/Abl ratio did not correspond to the major molecular response (MMR, <0.1%), the nilotinib dose was readjusted to 400mg daily. One week later, the TBIL and indirect bilirubin (IBIL) levels increased to 89 and 79 μmol/L, respectively. The levels of ALT and the other liver functional indicators were normal. Liver biopsy was performed, and the histopathology showed no obvious liver damage (Fig. 2). The Bcr-Abl/Abl ratio was <0.01% after 4 months of 400-mg nilotinib treatment, but the TBIL and IBIL levels were 60 to 80 and 40 to 60 μmol/L, respectively (Fig. 1).

We also detected a UGT1A1 mutation [ex1 c.686C>A (p.Pro229Gln)] in the patient and his mother (Fig. 3). On the basis of the data from a database (https://www.ncbi.nlm.nih.gov/clinvar/), the gene had a missense mutation that caused the pathogenesis, and both the patient and his mother were asymptomatic carriers due to the heterozygous mutation. This finding showed that the hyperbilirubinemia observed in the patient was induced by the nilotinib administration and suggested that nilotinib therapy may inhibit UGT1A1 activity, including bilirubin glucuronidation. To this end, the liver histology and function showed normal manifestations. Moreover, viral hepatitis (A–E), Epstein–Barr virus, and cytomegalovirus infections, and hereditary metabolic liver diseases were ruled out.

4. Discussion

Imatinib is a revolutionary drug used in the treatment of CML.[6] A single-drug therapy with imatinib can achieve complete cytogenetic responses and has attained an unprecedented overall survival rate.[7,8] However, one-third of the patients who used 400-mg IM did not achieve satisfactory outcomes.[9] Side effects such as progressive decrease in platelet count in patients restrict the use of IM in standard dose.[10,11] In 2007, nilotinib was...
Figure 2. Microscopic features of the liver biopsy specimens. A: Normal hepatic lobule (hematoxylin and eosin staining, original magnification ×100). B: Bland cholestasis (hematoxylin and eosin staining, original magnification ×400).

Figure 3. Subject: ex1 c.686C > A (p.Pro229Gln) Sanger sequencing map. A: Patient: had ex1 c.686C > A (p.Pro229Gln) gene mutation. B: Patient’s father: had no ex1 c.686C > A (p.Pro229Gln) gene mutation. C: Patient’s mother: had ex1 c.686C > A (p.Pro229Gln) gene mutation.
approved in the United States for the treatment of Philadelphia chromosome-positive CML,\(^\text{13}\) which is resistant or intolerant to previous treatments, including imatinib, thus opening up the possibility of using new treatment strategies for patients with CML.\(^\text{12,13}\)

In a phase 1 dose-escalation study of nilotinib,\(^\text{14,15}\) 119 patients with imatinib-resistant CML were enrolled and received different nilotinib doses (50, 100, 200, 400, 600, 800, and 1200 mg). The results showed that the high-dose groups (17%, 6/35, 600–1200 mg) had a higher incidence of severely elevated bilirubin level than the 400-mg dose group (9%, 3/32).

In a randomized open-label multicenter study,\(^\text{15}\) nilotinib was shown to be a more potent inhibitor of BCR-ABL than imatinib. Moreover, the CCyR rates achieved in 12 months were shown to be a more potent inhibitor of BCR-ABL than imatinib.

Whether nilotinib-induced hyperbilirubinemia is a drug-induced liver injury (DILI) remains to be elucidated. An international DILI Expert Working Group of clinicians and scientists developed uniform clinical chemistry criteria for DILI as follows:\(^\text{25}\)

1. ALT level of ≥5 times the ULN
2. ALP levels of ≥2 times the ULN (particularly with accompanying elevations in 5'-nucleotidase or γ-glutamyl transpeptidase concentration in the absence of a known bone pathology underlying the increase in ALP level)
3. ALT level of ≥3 times the ULN
4. TBIL levels of ≥2 times the ULN. On the basis of these criteria, nilotinib-induced hyperbilirubinemia is not a DILI.

To assess the probability of adverse drug reactions (ADR), we used the Naranjo ADR Probability Scale.\(^\text{26}\) (Table 2). A score of 13 indicates that hyperbilirubinemia is attributed to ADR caused by nilotinib rather than to DILI.

In conclusion, nilotinib is a selective tyrosine kinase receptor-inhibitor used in the treatment of CML.

### Table 1

| Number of cases | Diagnosis | Sex | Age | Country | Dose (mg) | dose adjustment or discontinued | Symptoms | TBIL (μmol/L) | IBIL (μmol/L) | ALT (U/L) | MMR (≡ 0.1%) | UGT1A1 mutations | Yr | Reporter |
|-----------------|-----------|-----|-----|---------|-----------|-----------------------------|----------|--------------|--------------|-----------|---------------|----------------|----|----------|
| 1               | CML       | Man | 73  | China   | No        | No                         | No       | 104          | 88           | Normal   | No            | No             | 2011 | Sammy Pak-lam Chen |
| 4               | CML       | Female | 66 | Japan   | 150       | Yes                        | No       | ≥1.8 mg/dL  | No           | No        | Yes          | Hom            | 2014 | Takahisa Shibata |
| 4               | CML       | Female | 48 | Japan   | 400       | No                         | No       | ≥1.8 mg/dL  | No           | No        | Yes          | Hom            | 2014 | Takahisa Shibata |
| 7               | CML       | Female | 51 | Japan   | 300       | Yes                        | Aremia   | ≥1.8 mg/dL  | No           | No        | Yes          | Co het         | 2014 | Takahisa Shibata |
| 1               | CML       | No   | 45-64| Japan   | 300-400   | No                         | Hyperbilirubinemia | No          | No         | 5 MMR (R)  | 2014 | Takahisa Shibata |
| 7               | CML       | Female | 48 | Japan   | 400       | No                         | No       | 66.7         | 58.1         | No        | No           | Het            | 2009 | Mikitaka Inoue |

No, No description; MMR, major molecular response; CMR, complete molecular response; Hom, homozgyous; Co het, compound heterozygous; Het, heterozygous genotype.

1. Chen SP, Poon WT, Mak CM, Lam CW, Kaung YL, Chan AT et al. Application of pharmacogenetics: UGT1A1*28 and nilotinib-induced unconjugated hyperbilirubinemia in a patient with chronic myeloid leukemia. Pathology2011 Apr;43(3):273-4.
2. Shibata T, Mirami Y, Mitsuma A, Morita S, Inada-Inoue M, Oguri T et al. Association between severe toxicity of nilotinib and UGT1A1 polymorphisms in Japanese patients with chronic myelogenous leukemia. Int J Clin Oncol2014 Apr;19(2):391-6.
3. Shibata T, Morita S, Maeda T, Oguri T, Inada-Inoue M, Oguri T et al. Association between severe toxicity of nilotinib and UGT1A1 polymorphisms in Japanese patients with chronic myelogenous leukemia. Int J Clin Oncol2011 Apr;16(2):132-6.
4. Fujita K, Sogami M, Akijima Y, Ando Y, Sasaki Y. The small-molecule tyrosine kinase inhibitor nilotinib is a potent non-competitive inhibitor of the SN-38 glucuronidation by human UGT1A1. Cancer Chemother Pharmacol2011 Jan;67(1):237-41.
Table 2
Naranjo Adverse Drug Reaction Probability Scale: Items and score.

| Question                                                                 | Yes | No  | Don’t know | Patient’s Score |
|--------------------------------------------------------------------------|-----|-----|------------|-----------------|
| 1. Are there previous conclusive reports on this reaction?               | +1  | 0   | 0          | +1              |
| 2. Did the adverse event appear after the suspected drug was administered? | +2  | −1  | 0          | +2              |
| 3. Did the adverse reaction improve when the drug was discontinued, or a specific antagonist was administered? | +1  | 0   | 0          | +1              |
| 4. Did the adverse reaction reappear when the drug was re-administered?  | +2  | −1  | 0          | +2              |
| 5. Are there alternative causes (other than the drug) that could on their own have caused the reaction? | −1  | +2  | 0          | +2              |
| 6. Did the reaction reappear when a placebo was given?                   | −1  | +1  | 0          | +1              |
| 7. Was the drug detected in the blood (or other fluids) in concentrations known to be toxic? | +1  | 0   | 0          | +1              |
| 8. Was the reaction more severe when the dose was increased, or less severe when the dose was decreased? | +1  | 0   | 0          | +1              |
| 9. Did the patient have a similar reaction to the same or similar drug in any previous exposure? | +1  | 0   | 0          | +1              |
| 10. Was the adverse event confirmed by any objective evidence?            | +1  | 0   | 0          | +1              |

Scoring: 3–9 = definite ADR; 5–8 = probable ADR; 1–4 = possible ADR; 0 = doubtful ADR. Patient’s Score = +13.

Author contributions
Investigation: Yun Ye and Xingbei Zhou
Supervision: Youwen Tan
Writing – original draft: Youwen Tan and Yun Ye
Writing – review & editing: Youwen Tan

References
[1] Kantarjian HM, Giles F, Gattermann N, et al. Nilotinib (formerly AMN107), a highly selective BCR-ABL tyrosine kinase inhibitor, is effective in patients with Philadelphia chromosome-positive chronic myelogenous leukemia in chronic phase following imatinib resistance and intolerance. Blood 2007;110:3540–6.
[2] Le Coutre P, Ottmann OG, Giles F, et al. Nilotinib (formerly AMN107), a highly selective BCR-ABL tyrosine kinase inhibitor, is active in patients with imatinib-resistant or -intolerant accelerated-phase chronic myelogenous leukemia. Blood 2008;111:1834–9.
[3] Kantarjian HM, Hochhaus A, Saglio G, et al. Nilotinib versus imatinib for the treatment of patients with newly diagnosed chronic phase, Philadelphia chromosome-positive, chronic myeloid leukemia: 24-month minimum follow-up of the phase 3 randomised ENESTnd trial. Lancet Oncol 2011;12:841–51.
[4] Breccia M, Alimena G. Nilotinib therapy in chronic myelogenous leukemia: the strength of high selectivity on BCR/ABL. Curr Drug Targets 2009;10:550–6.
[5] Deremer DL, Ustun C, Natarajan K. Nilotinib: a second-generation tyrosine kinase inhibitor for the treatment of chronic myelogenous leukemia. ClinTher 2008;30:1956–72.
[6] Uz B, Dolasik I. Comment: management of de novo chronic myelogenous leukemia and imatinib-induced acute rhabdomyolysis with the second-generati...