THREE JAPANESE PATIENTS WITH CRIGLER-NAJJAR SYNDROME TYPE I CARRY AN IDENTICAL NONSENSE MUTATION IN THE GENE FOR UDP-GLUCURONOSYLTRANSFERASE

Osamu Koiwai,1 Yoshihiro Yasui,1 Kayo Hasada,2 Sachiko Aono,3 Hiroshi Sato,4 Morihiko Fujikake,5 and Tsugutoshi Aoki6

1Department of Biochemistry and 2Ultrastructure Research, Aichi Cancer Center Research Institute, Chikusa-ku, Nagoya 464, Japan
3Department of Perinatology, Institute for Developmental Research, Aichi Prefecture Colony, Kasugai 480-03, Japan
4Department of Biology, Shiga University of Medical Science, Ohtsu 520-21, Japan
5Department of Pediatrics, Seirei Hospital, Showa-ku, Nagoya 466, Japan
6Department of Pediatrics, Toho University School of Medicine and Ohashi Hospital, Ohashi, Meguro-ku, Tokyo 153, Japan

Key Words mutation, UDP-glucuronosyltransferase, bilirubin, hereditary disease, jaundice

Crigler-Najjar syndrome type I (CN-I) is a severe disorder caused by chronic nonhemolytic unconjugated hyperbilirubinemia due to the complete absence of hepatic bilirubin UDP-glucuronosyltransferase activity (UGT; Crigler and Najjar, 1952; Arias et al., 1969). The disease is inherited as an autosomal recessive trait (Roy-Chowdhury et al., 1982). Bilirubin levels in the serum reach more than 342 μmol/liter and the patients succumb to kernicterus during the neonatal period unless treated with phototherapy, plasmapheresis or liver transplantation (Wolkoff et al., 1979). Treatment with phenobarbital has no effect on the bilirubin level of CN-I.

Bilirubin UGT exists as a tetramer on the luminal surface of the endoplasmic reticulum in liver cells (Peters et al., 1984) and it catalyzes the conversion of insoluble bilirubin to a water-soluble form by glucuronidation. Recently, cDNAs for rat and human bilirubin UGT were isolated (Sato et al., 1990; Ritter et al., 1991) and the structure of the gene for human bilirubin UGT in terms of exon-
intron organization was determined (Ritter et al., 1992; Bosma et al., 1992a). The
gene is located on chromosome 2 (van Es et al., 1993) and consists of five exons
(Bosma et al., 1992a, 1994). On the basis of the structure of the gene for human
bilirubin UGT, the genetic backgrounds of patients with CN-I have been elucidated.
While almost all reported patients with the disease have homozygous nonsense
or deletion mutations in the coding region of the gene (Brierley and Burchell,
1993), a few cases with homozygous missense mutations were found recently (Bosma
et al., 1992b; Erps et al., 1994). By screening, we found four patients who suffered
from CN-I in Japan. We already reported that one of them, patient A, carried
a homozygous nonsense mutation in exon 1 of the gene for bilirubin UGT (Aono
et al., 1994). We have now analyzed the genetic backgrounds of the other three
patients, B, C, and D, and, as we report herein, all three patients have a mutation
in exon 1 identical to that found in patient A.

The diagnosis of CN-I was based on a markedly elevated level of unconjugated
bilirubin in the serum that did not respond to treatment with phenobarbital and
on the absence of hepatic bilirubin UGT activity. Liver tissues were obtained
from the patients by biopsy and bilirubin UGT activities were assayed. No biliru-
bin UGT activity was detected in patient B at 14 days after birth or in patient C
at 5 months after birth by HPLC method (Kawade, 1980). In patient D, no en-
zymatic activity was detected at 51 days after birth by the method of Heirwegh

![Fig. 1. Mutations detected in patients with CN-I. Nucleotide sequences of the gene for
bilirubin UGT of patients B, C, and D were determined. Products of PCR were
sequenced directly. The sites of mutations detected in the three patients were
confirmed by sequencing of the subcloned products of PCR in pUC vectors.
Since the mutation detected in patient B and his elder sister C were identical to
each other, only the typical pattern from patient B is depicted. The wedges in-
dicate the sites of mutations.](image)