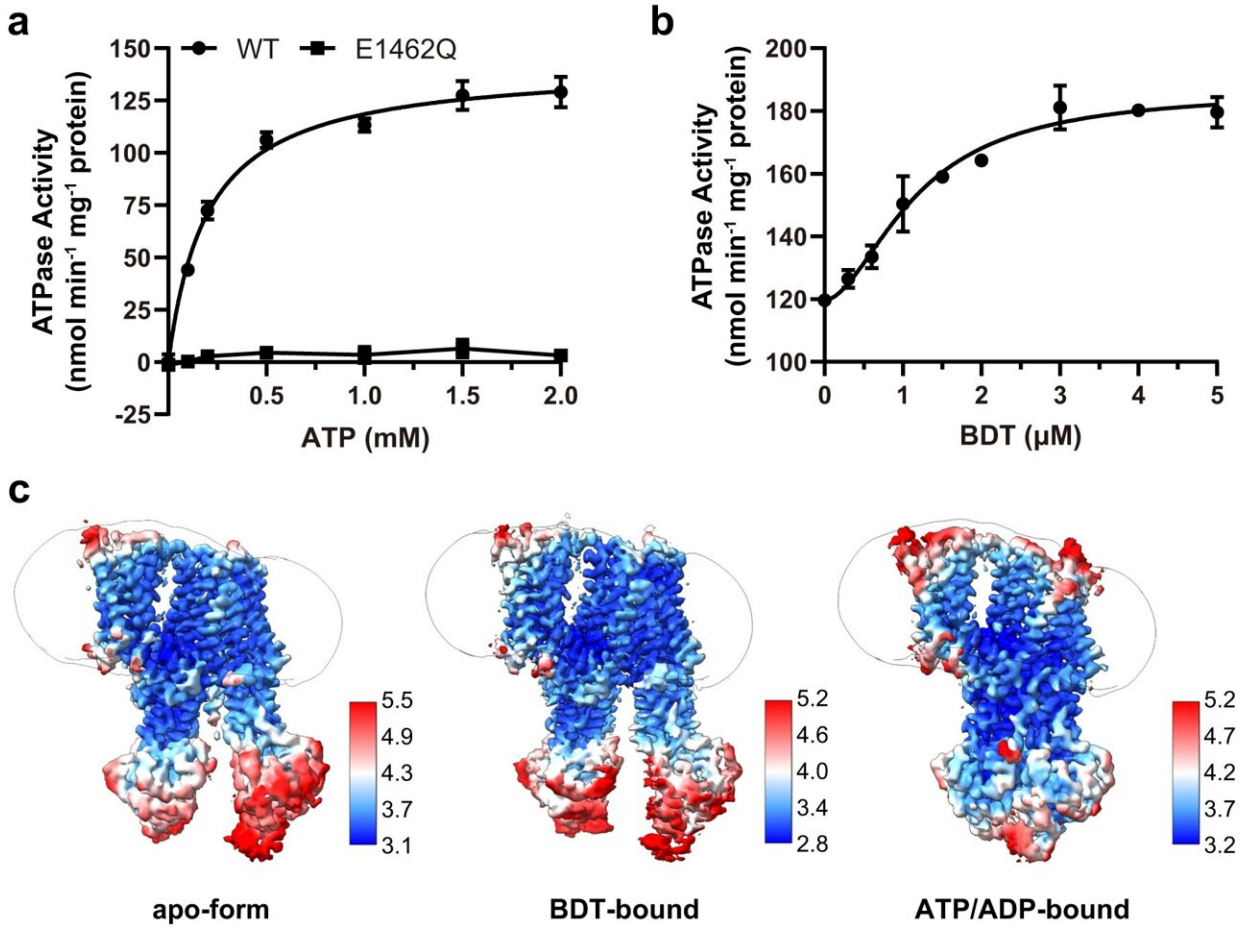
Molecular mechanism of transmembrane bilirubin transport by human ABCC2 transporter revealed

February 29 2024



Biochemical characterization and structure determination of ABCC2. **a** The ATPase activities of ABCC2 wild type (WT) and E1462Q mutant. The data points of WT activity are fitted with the Michaelis-Menten equation. **b** The substrate-stimulated ATPase activity of ABCC2 upon the addition of conjugated

bilirubin analog BDT. The data points are fitted with the Hill equation. All data points of (**a**) and (**b**) represent means of independent experiments ($n = 3$) and the error bars indicate the means ± standard deviation (SD). **c** The refined cryo-EM maps of three ABCC2 structures. The unsharpened maps are displayed as the outline to show the position of detergent micelle. The cryo-EM maps are colored by UCSF ChimeraX 1.5 according to the local resolution estimated by cryoSPARC 3.1. Credit: *Nature Communications* (2024). DOI: 10.1038/s41467-024-45337-5

The metabolic process of bilirubin has been a focus of medical research since the abnormal accumulation of bilirubin has been found to be associated with a variety of diseases. Bilirubin is a substance produced by the breakdown of aging or damaged red blood cells, and its effective removal is essential for human health.

A research team led by Prof. Chen Yuxing and Prof. Zhou Congzhao from the University of Science and Technology of China (USTC) of the Chinese Academy of Sciences has revealed the three-dimensional structure and working mechanism of the human bilirubin transporter ABCC2. The study was published in Nature Communications.

The researchers analyzed the structure of the ABCC2 protein determined by single-particle cryogenic electron microscopy (cryo-EM) in three different forms: the apo-form, the substrate-bound form, and the ATP/ADP-bound structures. They proposed a unique regulatory domain (R domain) that precisely controls ABCC2 substrate recognition and transport.

The R domain was folded into a hairpin structure that helped the ABCC2 protein stay still before it encountered bilirubin. But when it bound to a physiological substrate analog, bilirubin ditaurate (BDT), the hairpin part moved away. Additionally, the R domain helped proteins

select high-affinity conjugated bilirubin for preferential outward transport. The protein then changed its conformation in response to the hydrolysis of ATP, releasing the substrate into the duct.

In particular, the researchers pointed to a specific ABCC2 mutant (R1150H) that weakens the function of the R domain and leads to decreased transport activity, which explains the molecular mechanism of diseases such as Durbin-Johnson syndrome.

This study identified the key role of ABCC2 protein in the treatment of bilirubin under the fine regulation of the R domain, providing a new perspective for understanding bilirubin metabolism and a possibility for targeted treatment of the related genetic diseases.

More information: Yao-Xu Mao et al, Transport mechanism of human bilirubin transporter ABCC2 tuned by the inter-module regulatory domain, *Nature Communications* (2024). DOI: 10.1038/s41467-024-45337-5

Provided by University of Science and Technology of China