

Abstract

Gallbladder cancer (GBC) while rare worldwide has a high prevalence in north, east and north-eastern parts of India. Pathogenesis is unclear and outcomes poor. No suitable models are available to study GBC pathogenesis and drug response. The aim of this study was to establish a biobank comprising patient derived organoids (PDOs) from a wide variety of gallbladder diseases including GBC and other rare pathologies.

PDOs were generated from freshly isolated and cryopreserved cholangiocytes obtained from surgically resected gallbladder tissue of Indian patients. Protocol optimisation achieved 58% (69/119) and 52% (10/19) success in organoid generation and expansion for all and invasive malignant pathologies, respectively – highest globally for GBC organoids reported to date. Organoids maintained tight junction integrity; P-glycoprotein pump and enzymatic activity; preserved tissue-specific gene and protein marker expression; histological features and genetic variations. Besides cryopreserved organoids from 62 patients, primary gallbladder tissue and high-quality DNA, RNA and protein derivatives have been banked. In gene expression analyses of tissue, xanthogranulomatous cholecystitis (XGC) samples clustered with malignant subtypes, separate from non-malignant pathologies. Derived XGC organoids also clustered with organoids derived from GBC. Enriched hallmark pathway analyses support chronic inflammatory processes in the aetiology of GBC. Drug response testing confirmed that banked PDO lines are suitable for evaluating drug responses.

WNT signalling plays a pivotal role in organogenesis and cancer progression. A comparative evaluation of PDOs developed in culture condition with canonical WNT activator (Type A) and inhibitor (Type B) demonstrated that while PDOs developed in Type A offered superior long-term expansion, PDOs developed in Type B better preserved native histopathological identity. Canonical WNT modulation exerts a significant yet reversible influence on drug-response phenotype.

The developed biobank of PDOs from normal, inflamed and malignant gallbladder pathologies provides a promising resource to study gallbladder diseases, investigate the pathogenesis of GBC and study drug responses. Our findings also highlight the importance of aligning organoid culture conditions with specific research objectives.

Key words: gallbladder cancer; patient-derived organoids; organoid biobank; disease biology; drug response; WNT signalling