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DOI: https://doi.org/10.1016/j.pathol.2018.08.016

Acute Epstein–Barr virus hepatitis superimposed on drug induced liver injury causing severe hepatic dysfunction

Sir,

Epstein–Barr virus (EBV) induced hepatitis, drug induced liver injury (DILI) and choledocholithiasis are separate pathological entities that have been well described both clinically and histologically. While DILI and large duct obstruction from choledocholithiasis are well documented and several small cases series of acute EBV hepatitis are reported,[1,2] we present the first case report of all three of these entities in a single patient.

A 36-year-old female was seen by her general practitioner for coryzal symptoms and subjective fevers. She was subsequently treated with multiple antimicrobial agents including cephalexin and oseltamivir. She reported facial swelling after taking the cephalexin. The patient's regular medications included mesalazine as necessary for inactive Crohn's disease and escitalopram. One week later, she was admitted to hospital with a 5 day history of fevers and abdominal pain. On investigation, blood biochemistry showed a mixed hepatocellular and cholestatic liver function derangement. On ultrasound, there was sonographic tenderness with a thickened gallbladder wall and dilated common bile duct. Ceftriaxone and metronidazole were commenced to treat suspected acute cholecystitis with choledocholithiasis. The patient reported facial swelling and cutaneous rash after the administration of ceftriaxone. An endoscopic retrograde cholangiopancreatography (ERCP) showed choledocholithiasis in the common bile duct and a laparoscopic cholecystectomy was performed which showed chronic cholecystitis. Despite this, the patient's liver function continued to worsen: ALP 997 U/L (30–120 reference range), GGT 828 U/L (<51), AST 499 U/L (<41), ALT 459 U/L (<41), bilirubin 148 μmol/L (<25), and albumin 23 g/L (35–50). Due to the patient's drug reactions and deterioration with the risk of possible iatrogenic perforation, she was subsequently treated with benzylpenicillin, tazocin, clindamycin, and ciprofloxacin. A liver core biopsy was then performed to determine her risk of fulminant liver failure, as per Hy's law which predicts fulminant liver failure and death after DILI.[3]

Microscopically, the liver biopsy showed severe interface hepatitis and portal inflammation which included numerous eosinophils and lymphocytes with scattered loose, non-caseating granulomas. There was concomitant milder lobular inflammation and increased sinusoidal lymphocytes forming a 'string of beads' pattern (Fig. 1). No evidence of bile ductular proliferation was seen and no other features suggestive of acute or chronic large duct obstruction were identified. There was moderate macrovesicular steatosis. The liver architecture was intact with no significant fibrosis. An initial diagnosis was made favouring DILI but not ruling out the possibility of infection. In light of the above clinical information and liver biopsy findings, the gallbladder histopathology was also reviewed and showed an increased number of eosinophils superimposed on changes of chronic cholecystitis.

Initially, serological testing for EBV infection was performed and the result was consistent with past exposure to EBV (IgG positive IgM negative). However, over the next few days, blood films showed atypical lymphocytes suggestive of infectious mononucleosis, and this was confirmed with a positive Monospot. Repeat EBV serology performed 4 days later showed the appearance of EBV IgM, consistent with current acute EBV infection. All other tests for viral, autoimmune, and storage disorder aetiologies were negative. With this additional information, the prominent sinusoidal lymphocytes and endophlebitis were attributed to superimposed acute EBV hepatitis on DILI. The treating clinician subsequently ceased all unnecessary medications and the patient's liver function continued to improve with conservative management with normalisation of liver function.

Epstein–Barr encoding region (EBER) *in situ* hybridisation (ISH) was performed retrospectively which showed sparse positivity of lymphocytes (Fig. 1D). This pattern of staining was consistent with previously reported positivity for EBV hepatitis. EBER ISH is reported as a useful ancillary method for the detection of EBV hepatitis.[4,5] It has been shown to be at least as sensitive and specific as EBV DNA polymerase chain reaction (PCR). This result was also consistent with the patient's acute EBV seroconversion. EBV serology is also reported to show significant correlation with EBV PCR and ISH.[5]

EBV is a member of the herpesvirus family which infects 80–90% of the global population usually by late teens and early adulthood and remains latent with the possibility of reactivation.[4,6] EBV is usually asymptomatic in immunocompetent children or causes mild pharyngitis and constitutional symptoms in 30–50% of immunocompetent adolescents and adults. Acute infection usually involves the liver, however, generally it is mild and asymptomatic. There have been rare reports of severe hepatitis and liver failure attributed to EBV hepatitis,[1,7] and it may occur in young immunocompetent patients. Liver biopsies are rarely performed in the context of what is usually a self-limiting viral hepatitis which can be diagnosed by serological testing and routine blood analysis.

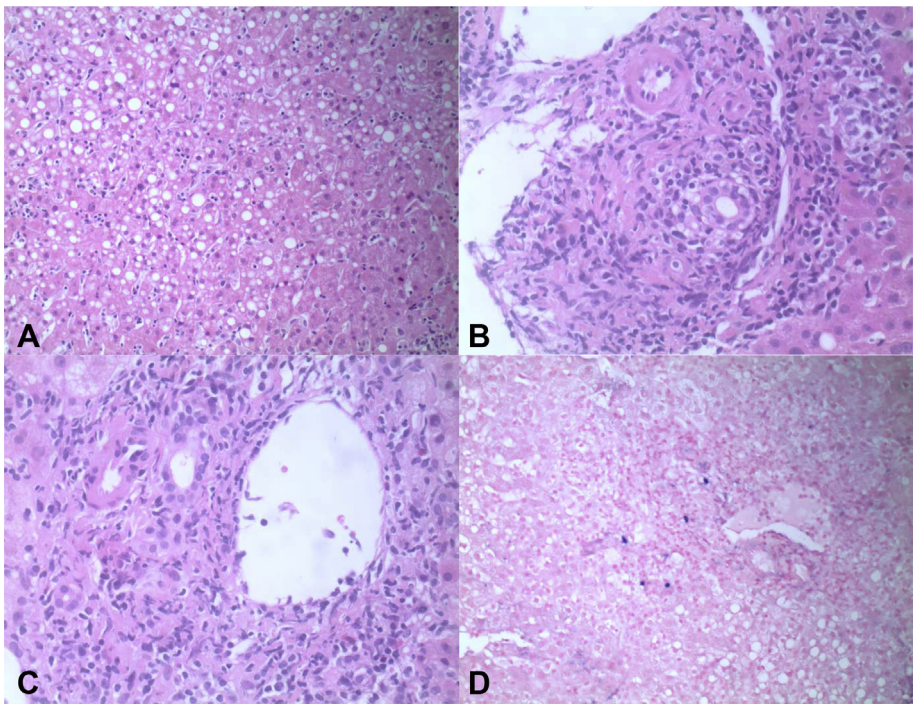

Fig. 1 (A) Hepatic parenchyma demonstrating sinusoidal lymphocytes arranged in a 'string of beads' pattern and steatosis. (B) Mild bile ductular damage. (C) Endophlebitis. (D) EBER ISH showing positive staining lymphocytes (purple).

Idiosyncratic drug reactions are an important cause of acute hepatic injury, and account for 13% of acute liver failure,[8] with many idiopathic cases that are likely not identified.[9] The mechanisms responsible are believed to be toxic metabolite accumulation or a hypersensitivity reaction, as in this case.[10] The antibiotics and escitalopram used by this patient have been previously implicated to cause DILI, even if rarely.[2] In this case, it is difficult to attribute the exact causative agent due to the number of different antimicrobials that had been prescribed prior to the development of DILI. However, given the documented acute hypersensitivity reaction, beta-lactam antibiotics are the most likely cause. The clinical, biochemical, and histopathological findings of DILI are relatively non-specific.[2] Histologically, inflammation with bile duct injury, eosinophils, granulomas, perivenular necrosis, and cholestasis out of proportion to hepatocellular injury are suggestive of but not specific for DILI and careful clinicopathological correlation is essential.[6]

Therefore, this case would be considered an atypical presentation of acute EBV hepatitis which was further complicated by the clinical history and possibility of DILI. There is significant histological overlap in the morphology of EBV hepatitis and DILI with portal inflammation, interface hepatitis, non-caseating granulomas, mild duct damage, mild lobular inflammation, and steatosis seen in both of these conditions (Fig. 1). However, sinusoidal infiltration by lymphocytes and endophlebitis are particularly classic of EBV hepatitis. There was a very prominent infiltrate of eosinophils, more typical of DILI than EBV (Fig. 2), which may be more pronounced in DILI due to an immuno-allergic response. The severity of the hepatic biochemical derangement was unusual for acute EBV hepatitis. The occurrence of an atypical serological profile where IgG appears before IgM is uncommon and clinically misleading but well described. This pattern may occur in approximately 7% of cases of acute infection due to late appearance of IgM, as occurred with this patient.[11] The contribution of the proven choledocholithiasis to the clinical situation was not apparent as classical histological features of large duct obstruction (bile ductular proliferation with neutrophilic infiltration, portal oedema, cholestasis and feathery degeneration) were not seen. An obstructive pattern of liver function derangement due to EBV hepatitis has been reported previously.[1,7]

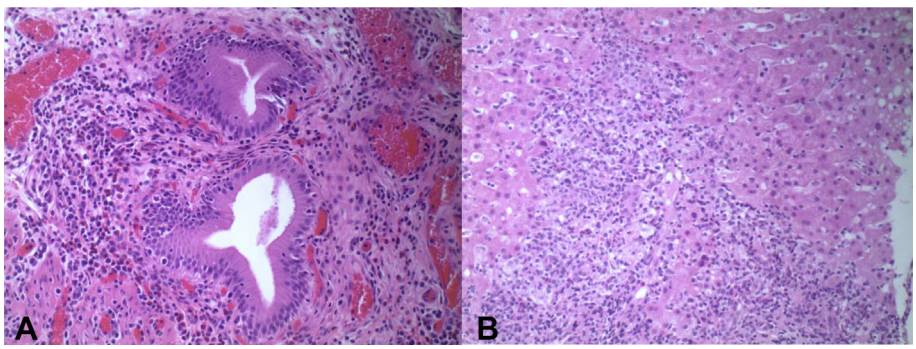

Fig. 2 (A) Gallbladder with chronic cholecystitis and prominent eosinophilic infiltrate. (B) Lobular hepatic inflammation with prominent eosinophilic infiltrate.

This case highlights the importance of a good clinical history and communication of relevant results between clinicians and pathologists. Without correlation of clinical, immunological, biochemical, and histological results the diagnosis could not be made and optimal management instituted. Throughout this patient's admission, the histological diagnosis evolved as more information became available.

Conflicts of interest and sources of funding: The authors state that there are no conflicts of interest to disclose.

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DOI: https://doi.org/10.1016/j.pathol.2018.07.007

An insight into *in vitro* susceptibility of non-*albicans* *Candida* species from bloodstream infections to echinocandins in a Singapore cohort

Sir,

Initial therapy of candidaemic patients with echinocandins has been demonstrated to be a significant predictor of survival.[1] The echinocandins have been promulgated as frontline agents for *Candida* bloodstream infection (BSI) in all patients, neutropenic or non-neutropenic.[2] In recent years, incidence of BSIs due to non-*albicans* species has surpassed those due to *Candida albicans* across many centres.[3] *Candida albicans* is deemed typically susceptible to the commonly used antifungals while acquired echinocandin resistance is an emerging problem among non-*albicans* species like *Candida glabrata*.[4] At the same time, other species, like *Candida parapsilosis* and *Meyerozyma guilliermondii*, are characterised by inherently raised minimum inhibitory concentrations (MICs) to echinocandins. Against the rising spectre of multidrug resistance, the Infectious Disease Society of America, in its recent guidelines on candidiasis, recommends susceptibility testing to echinocandins among those who have infections with *C. glabrata* or *C. parapsilosis* (strong recommendation; low-quality evidence).[2] Few studies in Singapore have evaluated resistance among non-*albicans* species to this class of drug. It was previously reported in 2016 that resistance to the echinocandins does not pose a major challenge across the Asia-Pacific region.[5]

At our 1600 bed tertiary care hospital, antifungal susceptibility testing is performed by the Sensititre YeastOne (TREK Diagnostic Systems, USA) on index isolates of *Candida* species from positive blood culture broths (BACTEC FX; Becton Dickinson, USA). The Laboratory Information System was used to interrogate the susceptibility profiles for all bloodstream isolates over a period of 3.5 years (February 2014–August 2017). The MICs were interpreted as per the recent Clinical and Laboratory Standards Institute species-specific clinical breakpoints.[6]

A total of 163 episodes of candidaemia were identified within this time period. Strains isolated from expired and discharged patients [*C. glabrata* ($n = 20$), *C. tropicalis* ($n = 11$), *Candida dubliniensis* ($n = 4$), *Candida orthopsilosis* ($n = 3$), *C. parapsilosis* ($n = 1$), *Candida duobushaemulonii* ($n = 1$)] were not subjected to susceptibility testing as per the laboratory policy and were excluded from the analysis.

Our hospital mainly deals with elderly patients. Patients in our study cohort had a median age of 69 years (interquartile range 61–79 years). In geriatric populations, infections due to *C. glabrata* typically eclipse those due to other non-*albicans* species.[7] Not surprisingly, this species comprised 50.4% ($n = 62$) of all non-*albicans* ($n = 123$) isolates subjected to a susceptibility test at our centre (Table 1). Of note, *C. glabrata*, a species prone to echinocandin resistance due to its plastic and haploid genome, tested almost uniformly susceptible. Only one isolate tested non-susceptible with an anidulafungin MIC of 0.25 mg L^{-1}. This patient had been on 11 days of intravenous anidulafungin (100 mg once a day) prior to collection of blood cultures.

The near absence of resistance bodes well for our elderly patient population where the presence of co-morbidities would significantly curtail therapeutic options. Amphotericin B would be a likely candidate for this species given its innately high MICs to the azoles. However, nephrotoxic agents like polyenes may not find favour in a geriatric centre such as ours, with liposomal formulations being several-fold more expensive than conventional options. Further, widely preferred liposomal formulations share similar side effect profiles, albeit with a lower frequency. These adverse effects are pertinent in this geriatric cohort given their overall compromised renal function status.

Echinocandin resistance was absent in *C. tropicalis*, a species for which non-susceptibility to the triazoles