



# Severe steroid-resistant anti-PD1 T-cell checkpoint inhibitor-induced hepatotoxicity driven by biliary injury

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## ABSTRACT

**Introduction** Hepatotoxicity from T-cell checkpoint blockade is an increasingly common immune-related adverse event, but remains poorly characterised and can be challenging to manage. Such toxicity is generally considered to resemble autoimmune hepatitis, although this assumption is extrapolated from limited clinicopathological reports of anti-cytotoxic T-lymphocyte-associated protein 4-induced hepatotoxicity.

**Methods** Here we report, with full clinicopathological correlation, three cases of T-cell checkpoint inhibitor-induced hepatotoxicity associated with anti-programmed cell death protein 1 agents.

**Results** We find that a major feature of these cases is biliary injury, including a unique case of vanishing bile duct syndrome, and that such toxicity was poorly responsive to long-term immunosuppression (corticosteroids and mycophenolate mofetil). Any potential benefits of long-term immunosuppression in these cases were outweighed by therapy-related complications.

**Discussion** We discuss potential aetiologies and risk factors for immune-mediated biliary toxicity in the context of the limited literature in this field, and provide guidance for the investigation and supportive management of affected patients.

## INTRODUCTION

T-cell checkpoint inhibitors, including the anti-cytotoxic T-lymphocyte-associated protein 4 (CTLA4) monoclonal antibody ipilimumab and the anti-programmed cell death protein 1 (PD1) monoclonal antibodies pembrolizumab and nivolumab, have transformed patient outcomes in a growing number of cancer types.<sup>1</sup> However, a high prevalence of immune-related toxicity has been observed in clinical trial and real-world populations treated with these drugs, and management of such toxicities is often difficult,<sup>2–4</sup> partially owing to a poor understanding of their underlying pathogenesis. While the development of immune-related toxicity may correlate positively with long-term clinical outcome,<sup>5</sup> for a growing number of patients toxicity necessitates treatment discontinuation and high doses of immunosuppressive agents are

## Key points

### What is already known about this subject?

Severe toxicities from growing use of T-cell checkpoint inhibitors in cancer treatment are increasingly encountered in clinical practice. However, little is understood about the pathogenesis of immune-related toxicities, owing to infrequent clinicopathological correlation. Hepatotoxicity from these agents can be challenging to manage, particularly when first-line corticosteroids are ineffective in restoring normal liver function. While most hepatotoxicity clinicopathological correlations have been reported for anti-cytotoxic T-lymphocyte-associated protein 4 agents, there has been a report of extrahepatic biliary injury induced by nivolumab.

### What does this study add?

This study reports three cases where corticosteroid-resistant hepatotoxicity was associated with intrahepatic biliary injury, and ran protracted course where complications of long-term immunosuppression (corticosteroids and mycophenolate mofetil) outweighed its benefits. We report a unique case of vanishing bile duct syndrome after a single infusion of pembrolizumab.

### How might this impact on clinical practice?

We hope this study will lead to early histopathological investigation of T-cell checkpoint inhibitor-induced hepatotoxicity, particularly in cases that do not respond promptly to first-line corticosteroids. We hope that our guidance will limit unnecessary toxicity from long-term immunosuppression in such cases, and aid with their supportive management.

generally indicated, the relative benefits and harms of which are not yet fully defined.

Hepatotoxicity from anti-CTLA4, anti-PD1 or anti-programmed death ligand 1 (PDL1) T-cell checkpoint inhibitors is one such clinically important immune-related toxicity,<sup>6</sup> occurring in approximately 5%–10% of patients,<sup>4</sup> although published rates of hepatotoxicity vary markedly by agent and study. A recent meta-analysis of 17 trials reported ORs for the development of all-grade

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hepatotoxicity of 5.01 (95% CI 4.06 to 6.2) and 1.94 (95% CI 1.28 to 2.94) for anti-CTLA4 and anti-PD1 inhibitors, respectively.<sup>6</sup> The respective ORs for high-grade hepatotoxicity were 4.67 (95% CI 3.42 to 6.39) and 1.58 (95% CI 0.66 to 3.78). T-cell checkpoint inhibitors appear synergistic in combination with regard to hepatotoxicity. For example, in the registration trial of the combination of ipilimumab and nivolumab in advanced melanoma, the rates of grade 3/4 elevations in aspartate aminotransferase (AST) and alanine transaminase (ALT) were 6.1% and 8.3%, respectively, in the combination arm, 0.6% and 1.6% in the ipilimumab arm, and 3.8% and 3.8% in the nivolumab arm.<sup>7</sup> Most current clinical guidelines dictate that grade 2 toxicity requires treatment to be held until improvement, with grade 3 or 4 toxicity mandating checkpoint inhibitor discontinuation, limiting further treatment options (see for example the European Society for Medical Oncology (ESMO) guidelines in ref<sup>4</sup>). Typically, management of presumed T-cell inhibitor-induced hepatotoxicity requires exclusion of other causes such as metastatic infiltration, biliary obstruction or viral causes, before initiating a course of corticosteroids, should the liver function derangement be severe or not spontaneously improve.

The aetiology of T-cell checkpoint inhibitor-induced hepatotoxicity is poorly understood, and is generally considered to be secondary to an autoimmune-type acute hepatitis, resembling the sporadic, autoantibody-associated disease,<sup>4,8</sup> but without positive serology for common antibodies associated with this or related autoantibody-associated conditions. Although there has been no definitive demonstration of an immunological overlap between T-cell checkpoint inhibitor-induced hepatotoxicity and sporadic diseases, unmasking of a tendency to autoimmunity has plausible biological rationale, given the mechanism of action of these agents in the promotion of diverse T-cell populations, some of which will be autoreactive, and is therefore likely to be an on-target effect. However, there is clearly significant heterogeneity in pathological reports to date.<sup>9</sup> Given the numbers of patients affected, there are surprisingly few reports concerning investigation, management strategies and long-term outcomes for patients with T-cell checkpoint-induced hepatotoxicity.<sup>10</sup> Steroids appear to be effective in reversing hepatotoxicity in the majority of patients with the condition (with a median time to resolution of 7 weeks in the registration trial of nivolumab and ipilimumab),<sup>7</sup> and reports of responses have also been observed with mycophenolate mofetil (MMF; see for example ref<sup>11</sup>). However, there have been infrequent clinicopathological reports of T-cell checkpoint inhibitor-induced hepatotoxicity, presumably due to the clinical presumption of an autoimmune-type hepatitis as this has been reported as an important mechanism of liver injury after CTLA4 blockade. From published reports, it is likely that a variety of hepatotoxic mechanisms exist, and this may explain the variation in clinical outcomes for patients with this complication.

Since hepatotoxicity associated with T-cell checkpoint inhibitors can be a highly complex immune-related toxicity to manage, it is important that we better understand the biology, heterogeneity and clinical course of such injury, in order to optimise future patient management, particularly for patients who do not respond promptly to corticosteroids. Here we report three cases of steroid-resistant liver injury induced by anti-PD1 agents from a single institution, each with clinicopathological correlation, and discuss the implications of these findings.

## METHODS

The case histories of three patients who had undergone liver biopsies for steroid-resistant T-cell checkpoint inhibitor-induced hepatotoxicity were reviewed retrospectively (including clinical history, results of blood tests, results of imaging tests and results of pathological investigations), along with specialist histopathology review. All biopsies taken were of adequate size for assessment. Presented patient ages are those at the time of presentation with evidence of hepatotoxicity. Cases are presented after clinicopathological correlation.

## RESULTS

### Case 1

A 49-year-old woman with B-rapidly accelerated fibrosarcoma (*BRAF*) mutant metastatic melanoma was treated initially with multiple surgical resections. She subsequently presented with symptomatic, unresectable brain metastases and limited extracranial disease in the subcutaneous tissues and peritoneum. She received oral steroids and whole brain radiotherapy, followed by pembrolizumab (2 mg/kg intravenously, planned for three weekly cycles). A baseline CT scan demonstrated likely hepatic steatosis, but no malignant liver infiltration.

Eight days after the first infusion of pembrolizumab, she presented to the emergency department with jaundice. Liver function tests were markedly deranged (bilirubin 90 µmol/L, alkaline phosphatase (ALP) 237 U/L, gamma-glutamyl transpeptidase 2094 U/L, AST 961 U/L and ALT 1536 U/L, with a normal prothrombin time). An ultrasound scan suggested hepatic steatosis, but did not demonstrate any focal lesions, biliary dilatations or vascular abnormality. A full liver screen excluded infectious and metabolic aetiologies (including hepatitis B, hepatitis C, cytomegalovirus (CMV), Epstein-Barr virus and adenovirus infection,  $\alpha$ 1-antitrypsin deficiency, Wilson's disease and haemochromatosis) and an autoantibody screen (including antinuclear, antimitochondrial, antismooth muscle, and anti-liver kidney microsomal antibodies) was unremarkable. Serum immunoglobulins revealed mild hypogammaglobulinaemia (5.59 g/L), with normal IgA and IgM levels. There were no recent medication changes except for the introduction of pembrolizumab.

Despite immediate treatment with prednisolone (~1 mg/kg once daily orally) for presumed

autoimmune-type hepatitis secondary to pembrolizumab, the patient's serum bilirubin and ALP continued to worsen (see [figure 1A](#) for liver function tests over time, and a summary of treatment types and their durations) and she proceeded to a diagnostic liver biopsy. This demonstrated diffuse steatosis with steatohepatitis and only a single small bile duct evident on H&E staining despite portal tracts being well represented and the sample measuring 14 mm in length ([figure 1B](#)). Cytokeratin 7 (CK7) immunohistochemistry also failed to demonstrate bile ducts and showed only a very minimal and focal intermediate hepatobiliary phenotype ([figure 1C](#)). Copper-associated protein (a feature of prolonged cholestasis) was not identified, and typical autoimmune hepatitis-like features were absent. The findings were felt to be consistent with a relatively acute-onset vanishing bile duct syndrome. The patient was commenced on ursodeoxycholic acid (UDCA) after the diagnosis was made.

Given no improvement in serum ALP or bilirubin with corticosteroids and UDCA, MMF was added at a dose of 1g twice daily orally. Although ESMO guidelines<sup>4</sup> recommend alternative escalation to 2mg/kg dose of (methyl) prednisolone for immune-related hepatitis, this was not felt appropriate given the appearances of the liver biopsy. After 56 days, MMF was then stopped owing to a lack of improvement in liver function and profound neutropaenia. The patient also developed insulin-dependent diabetes mellitus, likely steroid induced, as well as a remarkably high serum cholesterol (40.7 mmol/L) that was unresponsive to statin treatment. Fat-soluble vitamin depletion was observed with an undetectable serum vitamin D level (presenting with profound symptomatic hypocalcaemia with a corrected serum calcium of 1.4 mmol/L), a serum vitamin K level of 0.09 µg/L and a protein induced by vitamin K absence/antagonist-II of >10 AU/mL (accompanied by a rising international normalised ratio (INR)). The low serum calcium and the elevated INR were considered to be a consequence of cholestasis and responded completely to oral vitamin D and K supplementation. The patient suffered a wedge fracture at T11, thought to be a consequence of prolonged steroid use and/or vitamin D depletion.

Eight weeks after receipt of the single pembrolizumab infusion, the patient was offered *BRAF*-targeted treatment. Since both dabrafenib and trametinib are metabolised by the liver and excreted in the bile, dabrafenib was initially introduced at 75 mg once daily (25% standard dose), and increased after demonstration of tolerance to 75 mg twice daily after 2 weeks. A partial response in the extracranial disease, but a mixed response in the intracranial disease was observed. Trametinib was then added at a dose of 1 mg once daily, with further partial response in the extracranial disease, and mixed response in the intracranial disease. Both drugs were escalated to full treatment doses (dabrafenib 150 mg twice daily and trametinib 2 mg once daily). These doses were well tolerated, and the hepatic dysfunction slowly began to improve with time and ongoing steroid/UDCA treatment. The

patient ultimately died from progressive intracranial disease almost 8 months after presenting with vanishing bile duct syndrome, and 6 months after starting *BRAF*-targeted treatment.

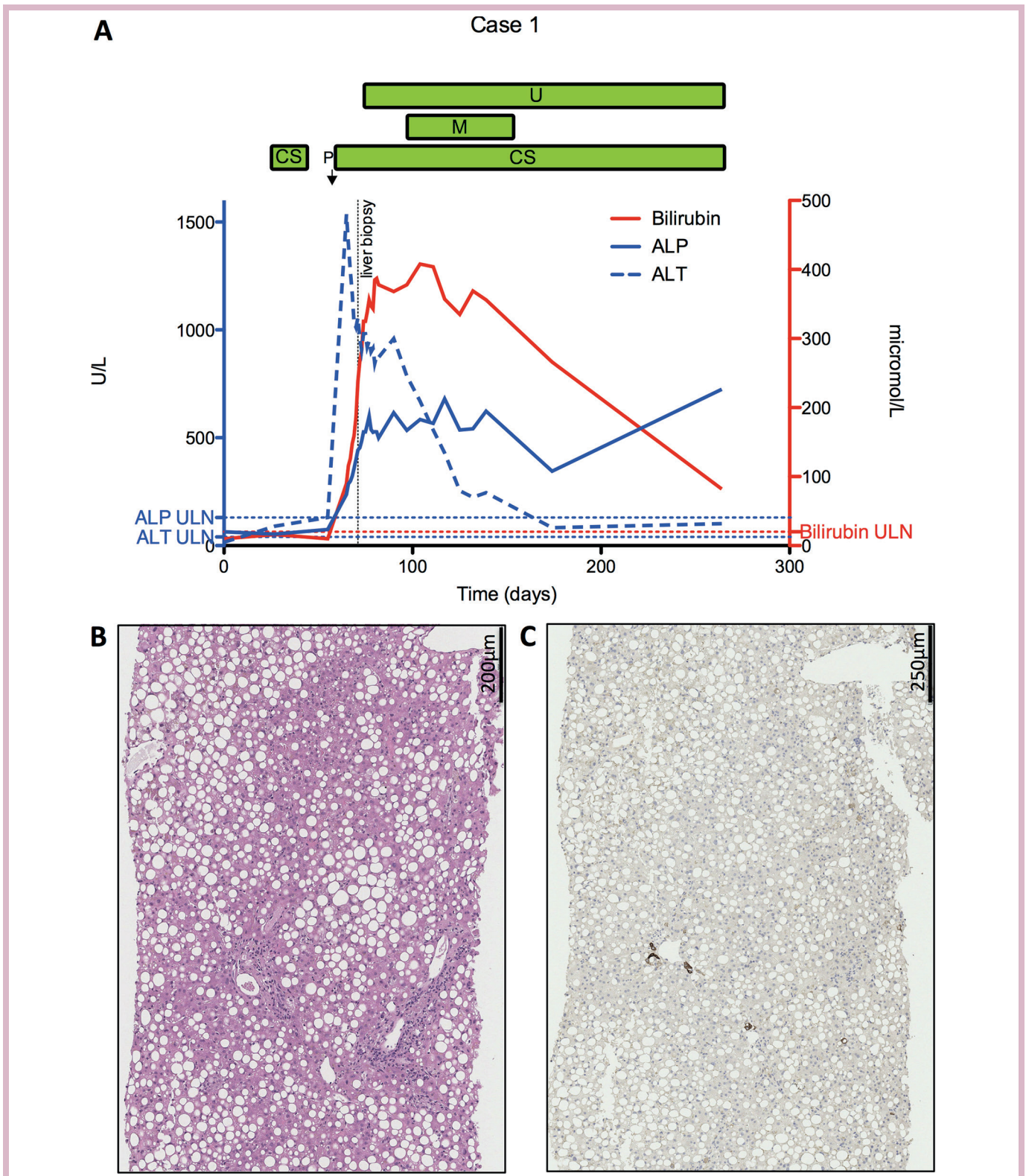
## Case 2

A 59-year-old woman with *BRAF* wild-type metastatic melanoma was initially treated with multiple surgical resections. After further disease progression in the left adrenal gland and retroperitoneum, the patient commenced treatment on the CHECKMATE 067 trial,<sup>7</sup> but confirmed to have received nivolumab on subsequent unblinding. After three cycles of treatment, reimaging confirmed a complete radiological response to treatment. However, liver function tests began to worsen, a liver screen (as per case 1) was unremarkable, and there were no recent changes to medications, aside from nivolumab. The patient was commenced on prednisolone ~1mg/kg once daily orally, initially with some improvement, but her liver function tests then worsened and she underwent a diagnostic liver biopsy ([figure 2B–D](#)). This showed a spectrum of focally severe, degenerative duct injury with ductular reaction, focal periductal fibrosis and slight inflammation of a partially sampled large duct. Patchy periportal copper-associated protein accumulation was also noted along with evidence of possible focal ductopaenia and focal concentric periductal fibrosis, all in keeping with a subacute (or chronic) pattern of biliary injury. In addition, there was evidence of parenchymal necroinflammation, including a histiocytic and eosinophilic component. The patient was started on UDCA after pathology review, with subsequent improvement in liver function tests. The patient developed pelvic and spinal insufficiency fractures, thought to be due to long-term steroid use, and follow-up imaging revealed definitive disease progression. In the absence of other systemic treatment options at the time, she proceeded to three cycles of dacarbazine chemotherapy, with a mixed response. Given the deterioration in her clinical status, the patient was discharged to community palliative care. Two years and three months later, after gradual clinical improvement, she re-presented to our service. Imaging revealed no evidence of metastatic disease, and liver function tests had normalised. She was asymptomatic, aside from ongoing back pain from spinal fractures. The patient remains alive at the time of writing, 5.5 years after stopping nivolumab treatment, and continues on surveillance.

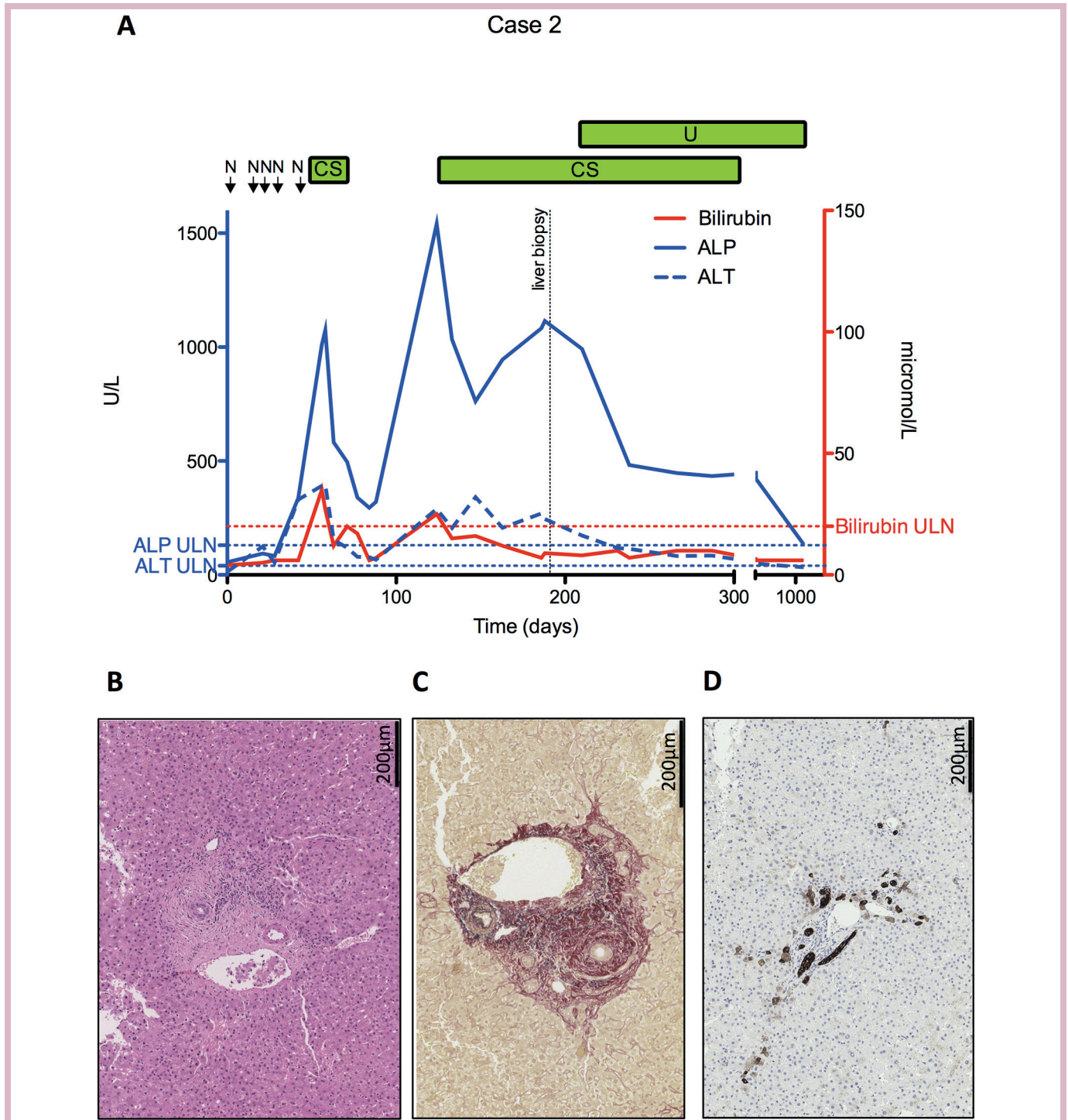
## Case 3

A 76-year-old man with epithelioid mesothelioma was initially treated with an extended pleurectomy/decortication. He progressed with peritoneal metastatic disease, and was offered pembrolizumab off-label at another institution. After a single infusion of pembrolizumab, he presented 24 days later to the emergency department with jaundice and deranged liver function tests. An ultrasound scan of the upper abdomen revealed only mild hepatomegaly, with concomitant mild splenomegaly, and a liver





**Figure 1** (A) Graph showing the changes in liver function tests over time for case 1, with a summary of systemic treatments given over time. The right y-axis refers to bilirubin levels, the left y-axis refers to ALP and ALT levels. (B) Photomicrograph of a representative H&E section from a liver biopsy in case 1. This section shows severe hepatic steatosis, and includes portal tracts without discernible bile ducts. (C) Photomicrograph showing cytokeratin 7 immunohistochemistry of a representative section from a liver biopsy in case 1. This confirms the absence of bile ducts, and demonstrates slight and focal intermediate hepatobiliary phenotype. ALP, alkaline phosphatase; ALT, alanine aminotransferase; CS, corticosteroids; M, mycophenolate mofetil; U, ursodeoxycholic acid; ULN, upper limit of normal.

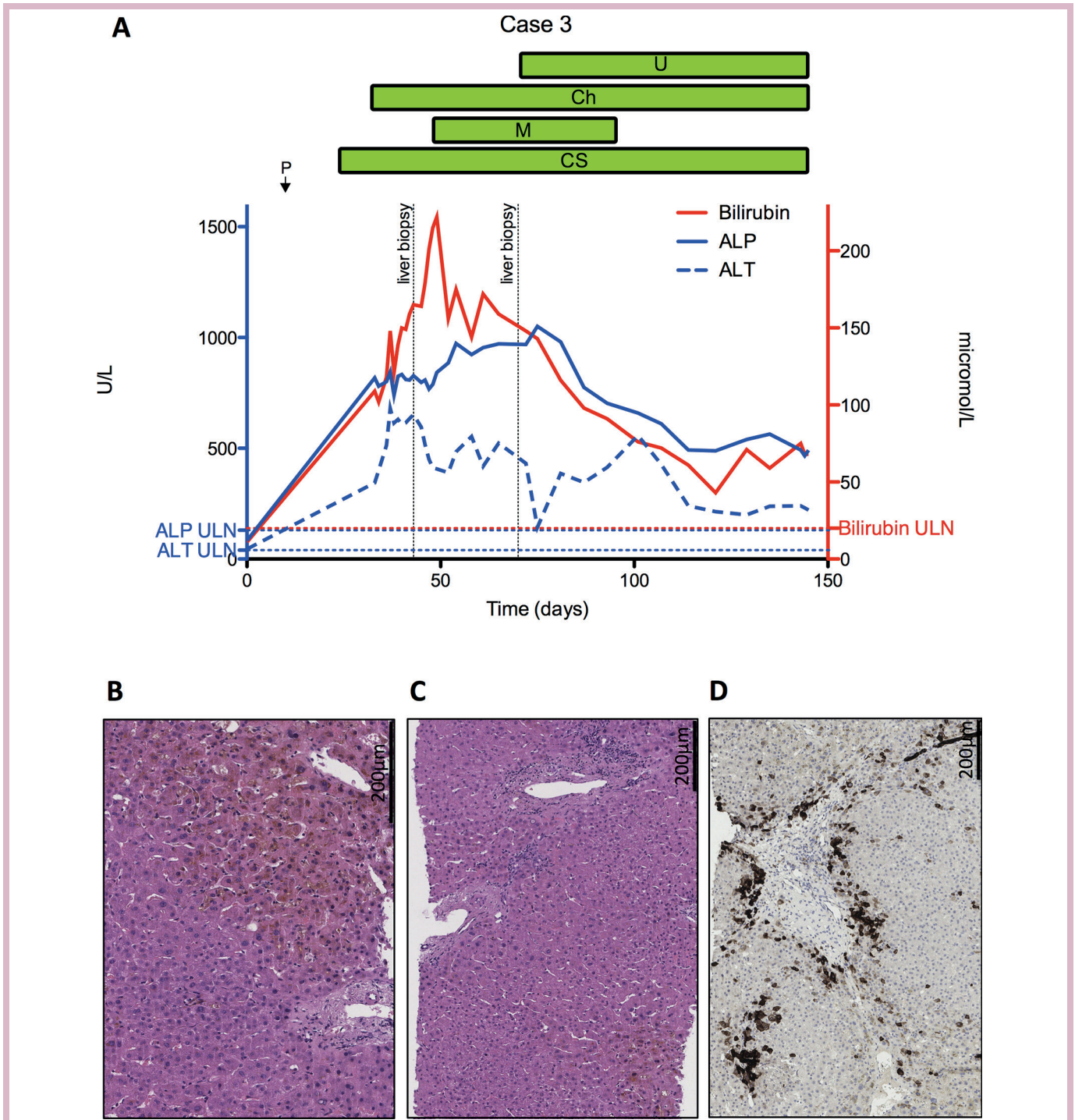


**Figure 2** (A) Graph showing the changes in liver function tests over time for case 2, with a summary of systemic treatments given over time. The right y-axis refers to bilirubin levels, the left y-axis refers to ALP and ALT levels. (B) Photomicrograph of a representative H&E section from a liver biopsy in case 2. This section shows degenerative duct atypia and periductal fibrosis, similar to that observed in chronic biliary disease. (C) Photomicrograph of the same portal tract in B, with elastic picro sirius red staining, demonstrating fibrosis (collagen in red). (D) Photomicrograph showing cytokeratin 7 immunohistochemistry of a representative section from a liver biopsy in case 2. This shows patchy periportal intermediate hepatobiliary phenotype, suggesting a cholangiopathic process. ALP, alkaline phosphatase; ALT, alanine aminotransferase; CS, corticosteroids; U, ursodeoxycholic acid; ULN, upper limit of normal.

screen (as per case 1) was unremarkable. There were no recent medication changes except for the introduction of pembrolizumab.

This patient was commenced promptly on intravenous methylprednisolone ~2 mg/kg daily, but with no associated improvement in liver function ([figure 3A](#)). This was





**Figure 3** (A) Graph showing the changes in liver function tests over time for case 3, with a summary of systemic treatments given over time. The right y-axis refers to bilirubin levels, the left y-axis refers to ALP and ALT levels. (B) Photomicrograph of a representative H&E section from the first liver biopsy in case 3. This section shows a portal tract with an attenuated bile ductule at the periphery of a tract, along with a naked arteriole. The parenchyma shows severe cellular and canalicular cholestasis. (C) Photomicrograph of a representative H&E section from the first liver biopsy in case 3, further demonstrating the marked degree of duct irregularity and degenerative change with focal ductular reaction and cholestasis towards the edge of the image. (D) Photomicrograph showing cytokeratin 7 immunohistochemistry of a representative section from the first liver biopsy in case 3. This shows prominent intermediate hepatobiliary phenotype consistent with the degree of duct injury and ductopaenia. ALP, alkaline phosphatase; ALT, alanine aminotransferase; Ch, cholestyramine; CS, corticosteroids; M, mycophenolate mofetil; U, ursodeoxycholic acid; ULN, upper limit of normal.

switched to oral prednisolone ~1 mg/kg once daily. The patient was also commenced on MMF 500 mg twice daily orally and he proceeded to a diagnostic liver biopsy. This showed severe cholestasis and duct injury with evidence of parenchymal loss and regeneration. Prominent intermediate hepatobiliary phenotype was seen in both periportal and perivenular locations using CK7 immunohistochemistry, with bile ducts absent from 6 of 22 tracts, and copper-associated protein deposition was not evident (figure 3B–D).

Despite corticosteroids and MMF, there was no significant improvement in liver function, so a second liver biopsy was performed. In comparison to the previous biopsy, this showed marked improvement in the degree of cholestasis and inflammation, and although there was significant ongoing duct damage, there did not appear to be progression of ductopaenia. UDCA was introduced at this point, and was associated with some minor improvement in liver function. MMF was discontinued owing to marked lymphopaenia. In the absence of further suitable treatment options, this patient died from progressive disease 140 days after the infusion of pembrolizumab.

## DISCUSSION

We have described three histopathologically distinct cases of steroid-resistant, anti-PD1 T-cell checkpoint inhibitor-induced hepatotoxicity with overlapping clinicopathological characteristics. In each case, although there was some improvement in liver function tests over months after commencing corticosteroids, the clinical course of hepatotoxicity was prolonged and severe, with marked heterogeneity in the patterns of ALT/ALP/bilirubin derangements and recovery. In these cases, the biliary tract was a major target of injury, revealing variable degrees of duct damage and ductopaenia, including vanishing bile duct syndrome. In case 2, some of the changes were reminiscent of those seen in chronic biliary disease with accumulation of copper-associated protein and periductal fibrosis, despite the absence of known pre-existing biliary disease. None of the cases showed classical features of autoimmune hepatitis, and there was no significant lymphocytic infiltrate evident at the time of liver biopsy. In case 3, where two liver biopsies were taken, there was an improvement in the appearances of the second biopsy after the patient was on both prolonged corticosteroids and MMF, but we cannot rule out the possibility of intrinsic repair which could have occurred in the absence of intervention.

There is limited literature on the outcomes for ipilimumab-induced hepatotoxicity, although most reports suggest a good outcome with immunosuppression.<sup>4 7 12</sup> Much less information is available about the natural course and response to corticosteroids for severe cases, and for anti-PD1 antibody-induced hepatotoxicity. None of the cases reported here showed swift improvement in liver function with corticosteroids or MMF, although cases 2 and 3 did show some possible improvement with UDCA.

The timescale of improvement despite prolonged immunosuppression (many months) suggests that improvement was due to a slow, intrinsic repair process, rather than as a result of our interventions. Prolonged immunosuppression resulted in very significant treatment-related complications in cases 1 and 2, and we would now urge caution with prolonged use of high doses of immunosuppressive drugs, especially where there is little evidence of improvement with a trial of treatment. We recommend a dose of 1 mg/kg (methyl)prednisolone and a trial period of 2 weeks' treatment duration, with careful dose tapering of 5 mg increments on a weekly basis should a liver biopsy not reveal any significant lymphocytic/neutrophilic infiltrate—if this is present, escalation to 2 mg/kg is appropriate, although it is unclear if such higher doses are justified. UDCA is well tolerated, has biological rationale for treating these patients and should be used more widely in these patients. We also suggest early involvement of hepatologists in the care of these patients, and, where feasible, pathological determination of the pattern of hepatobiliary injury by liver biopsy to help guide further management as above. The anti-tumour necrosis factor monoclonal antibody infliximab, while useful for the management of severe T-cell checkpoint inhibitor-induced colitis, has not yet been reported as a possible management option for hepatotoxicity, perhaps owing to fears of worsening liver function, given that it is itself associated with hepatotoxicity. Other immunosuppressants with distinct mechanisms of action could be considered for severe or non-resolving cases, for example, the calcineurin inhibitor tacrolimus, or antithymocyte globulin, which was successfully used in one case of corticosteroid and MMF-resistant, severe hepatotoxicity after ipilimumab and nivolumab administration.<sup>13</sup>

The striking presentation of vanishing bile duct syndrome in case 1 is the first such report of this very rare and serious condition in association with T-cell checkpoint inhibitors. This syndrome refers to a condition with diverse aetiologies where intrahepatic bile ducts are progressively destroyed, leading to cholestasis, with limited outcome data available.<sup>14 15</sup> Case reports and small case series suggest a generally poor prognosis, especially for patients with more complete ductal loss, where limited recovery is seen and the condition is often progressive. The pathogenesis is poorly understood, and aetiological precipitants include medications (including many classes of antibiotics), infections (such as HIV/CMV) and lymphomas, and there is an overlap with various autoimmune conditions such as primary biliary cirrhosis, primary sclerosing cholangitis, sarcoidosis and graft versus host disease (see the excellent NIH LiverTox summary at [https://livertox.nih.gov/phenotypes\\_vbds.html](https://livertox.nih.gov/phenotypes_vbds.html)). These latter precipitants, coupled together with our report of the syndrome secondary to T-cell checkpoint inhibition, lend support to the theory that this is an immunological condition. However, given that there is no definitive evidence that corticosteroids are beneficial, and given the rapidity of onset in our case, it is

possible that an initial immunological event causes sufficient damage such that subsequent immunosuppression comes too late to effectively treat the condition. Immunosuppression appears to be particularly ineffective in severe injury, and a marker of severe injury appears to be biliary involvement. Other possible mechanisms include a metabolic insult, perhaps through systemic release of cytokines. In case 1, there was pre-existing steatosis, which may be a risk factor for the development of the syndrome. Moderate to severe steatosis has also been described in two cases of presumed ipilimumab-induced hepatotoxicity.<sup>9</sup> The pathogenic mechanisms leading to biliary injury in cases 2–3 may be similar to (but milder than) those in case 1, or may be distinct. As well as auto-antigen recognition on biliary epithelium, other plausible explanations include an unmasked recognition of bacterial epitopes in a chronically colonised biliary tract. This may be stimulated by systemic interferon- $\gamma$ , since this promotes antigen presentation by biliary epithelial cells, and biliary epithelial cell-mediated T-cell activation is partially PD1 ligand dependent.<sup>16–18</sup> These mechanisms require further research in both animal models and with patient biopsy specimens.

An early case series of ipilimumab-induced hepatotoxicity reported three liver biopsies: two of these showed panlobular hepatitis, while one showed mild portal mononuclear infiltrate.<sup>8</sup> A further series of 11 such patients with liver biopsies revealed active hepatitis in six cases, zone 3 hepatitis in three cases, one case with features suggestive of non-alcoholic steatohepatitis, as well as a further case with portal inflammation and cholestasis.<sup>9</sup> Recently, treatment with nivolumab was associated with the development of extrahepatic biliary injury in three patients.<sup>19</sup> Two of these had liver biopsies that showed T-cell infiltration around the Glisson's capsule. There was a poor or slow response to steroid treatment in the two cases thus treated. While our cases did not show radiological evidence of extrahepatic biliary injury, taken together, all of these observations suggest that biliary injury in general may be a class effect of anti-PD1 antibodies, that steroids and other immunosuppressants may have a limited role in treating these particular complications, and that UDCA, cholestyramine and fat-soluble vitamin supplementation should be instituted when cholangitis is observed on imaging, when a pathological diagnosis of bile duct injury is made or when fat-soluble vitamin depletion is present.

## CONCLUSIONS

This case series provides insight into the mechanism of injury in a subset of patients who present with anti-PD1 T-cell checkpoint inhibitor-induced hepatotoxicity. A common link between these cases is the presence of biliary ductal injury, and a poor or incomplete response to immunosuppressive measures. These and other reported cases of T-cell checkpoint inhibitor-induced hepatotoxicity point to varied mechanisms of injury, and provide an argument for early investigation of presumed T-cell

checkpoint inhibitor-induced hepatotoxicity with liver biopsies, in order to provide detailed information on the pattern of injury, likely prognosis, and help determine the likely response to toxic immunosuppressive regimens. Further clinicopathological correlation in larger patient populations will likely provide further information to help guide prognostication and management for individual patients. We encourage physicians with patients who have undergone liver biopsies for presumed T-cell checkpoint inhibitor-induced hepatotoxicity to send us fully anonymised details of their cases (to [gd231@cam.ac.uk](mailto:gd231@cam.ac.uk)) in order to help build up a fuller picture of this important treatment complication to provide guidance on optimal patient care.

**Contributors** GJD and PGC devised the concept for the manuscript. GJD wrote the manuscript and prepared the figures. AMD prepared the histopathological slides for publication. AMD, SED and RB provided the specialist histopathological review of the biopsy specimens. CAP and SVH provided specialist oncological input into patient management. GFM provided specialist hepatological input into patient management. All authors read and edited the manuscript to produce the final version.

**Competing interests** None declared.

**Patient consent** Detail has been removed from this case description/these case descriptions to ensure anonymity. The editors and reviewers have seen the detailed information available and are satisfied that the information backs up the case the authors are making.

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## REFERENCES

- Martin-Liberal J, Ochoa de Olza M, Hierro C, *et al*. The expanding role of immunotherapy. *Cancer Treat Rev* 2017;54:74–86.
- Kumar V, Chaudhary N, Garg M, *et al*. Current Diagnosis and Management of Immune Related Adverse Events (irAEs) Induced by Immune Checkpoint Inhibitor Therapy. *Front Pharmacol* 2017;8:49.
- Friedman CF, Proverbs-Singh TA, Postow MA. Treatment of the immune-related adverse effects of immune checkpoint inhibitors: a review. *JAMA Oncol* 2016;2:1346–53.
- Haanen J, Carbone F, Robert C, *et al*. Management of toxicities from immunotherapy: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. *Ann Oncol* 2017;28:iv119–iv142.
- Mian I, Yang M, Zhao H, *et al*. Immune-related adverse events and survival in elderly patients with melanoma treated with ipilimumab. *J Clin Oncol* 2016;34:3047.
- Wang W, Lie P, Guo M, *et al*. Risk of hepatotoxicity in cancer patients treated with immune checkpoint inhibitors: A systematic review and meta-analysis of published data. *Int J Cancer* 2017;141:1018–28.
- Larkin J, Chiarion-Sileni V, Gonzalez R, *et al*. Combined nivolumab and ipilimumab or monotherapy in untreated melanoma. *N Engl J Med* 2015;373:23–34.
- Kim KW, Ramaiya NH, Krajewski KM, *et al*. Ipilimumab associated hepatitis: imaging and clinicopathologic findings. *Invest New Drugs* 2013;31:1071–7.
- Johncilla M, Misdraji J, Pratt DS, *et al*. Ipilimumab-associated hepatitis: clinicopathologic characterization in a series of 11 cases. *Am J Surg Pathol* 2015;39:1075–84.
- Abdel-Wahab N, Shah M, Suarez-Almazor ME. Adverse events associated with immune checkpoint blockade in patients



- with cancer: a systematic review of case reports. *PLoS One* 2016;11:e0160221.
11. Tanaka R, Fujisawa Y, Sae I, *et al.* Severe hepatitis arising from ipilimumab administration, following melanoma treatment with nivolumab. *Jpn J Clin Oncol* 2017;47:175–8.
  12. Della Vittoria Scarpati G, Fusciello C, Perri F, *et al.* Ipilimumab in the treatment of metastatic melanoma: management of adverse events. *Onco Targets Ther* 2014;7:203.
  13. Spänkuch I, Gassenmaier M, Tampouri I, *et al.* Severe hepatitis under combined immunotherapy: resolution under corticosteroids plus anti-thymocyte immunoglobulins. *Eur J Cancer* 2017;81:203–5.
  14. Nakanuma Y, Tsuneyama K, Harada K. Pathology and pathogenesis of intrahepatic bile duct loss. *J Hepatobiliary Pancreat Surg* 2001;8:303–15.
  15. Reau NS, Jensen DM. Vanishing bile duct syndrome. *Clin Liver Dis* 2008;12:203–17.
  16. Kamihira T, Shimoda S, Nakamura M, *et al.* Biliary epithelial cells regulate autoreactive T cells: implications for biliary-specific diseases. *Hepatology* 2005;41:151–9.
  17. Ayres RC, Neuberger JM, Shaw J, *et al.* Intercellular adhesion molecule-1 and MHC antigens on human intrahepatic bile duct cells: effect of pro-inflammatory cytokines. *Gut* 1993;34:1245–9.
  18. Cruickshank SM, Southgate J, Selby PJ, *et al.* Expression and cytokine regulation of immune recognition elements by normal human biliary epithelial and established liver cell lines in vitro. *J Hepatol* 1998;29:550–8.
  19. Kawakami H, Tanizaki J, Tanaka K, *et al.* Imaging and clinicopathological features of nivolumab-related cholangitis in patients with non-small cell lung cancer. *Invest New Drugs* 2017;35:529–36.