

Liver toxicity as a limiting factor to the increasing use of immune checkpoint inhibitors



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Summary

Immune checkpoint inhibitors (ICIs) improve clinical outcomes in patients suffering from different types of cancer. Liver toxicity is one of the immune-related adverse events associated with immunotherapy; although not common, its management is challenging as it is extremely heterogeneous in terms of presentation and severity. Differences in the development and evolution of ICI-related toxicity in healthy or cirrhotic livers have not yet been elucidated. Assessing causality is key to diagnosing ICI-induced liver toxicity; liver biopsies can assist not only in the differential diagnosis but also in assessing the severity of histological liver damage. The current classification of severity overestimates the grade of liver injury and needs to be revised to reflect the views of hepatologists. Spontaneous improvements in ICI-related liver toxicity have been reported, so corticosteroid therapy should probably be individualised not systematic. The reintroduction of ICIs in a patient with previous immune-mediated hepatitis may be possible, but the risk/benefit ratio should be considered, as the risk factors for hepatitis recurrence are currently unclear. The management of these patients, requiring a balance between efficacy, toxicity and specific treatments, necessitates multidisciplinary collaboration. The incidence of immune-related liver toxicity will continue to rise based on the increasing use of ICIs for most cancers, mandating improved understanding and management of this complication.

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Introduction

The introduction of immune checkpoint inhibitors (ICIs) has dramatically changed the landscape of cancer therapy and significantly improved survival in several cancer types, including metastatic melanoma and lung cancers. Inhibitory receptors on the T cell membrane preserve self-tolerance and prevent immune-driven diseases. Their sustained expression leads to T cell exhaustion, which can be observed in cancer and chronic infections. Cancer immunotherapy involves blocking these inhibitory receptors with monoclonal antibodies in order to reverse T cell exhaustion and restore T cell activity against tumour-specific antigens.¹ The considerable efficacy of ICIs has been associated with the onset of more or less severe immune-related adverse events (irAEs), affecting several organs due to a loss of self-tolerance.^{2–4}

While ICIs offer an excellent therapeutic option for patients with advanced cancer, the development of toxicities is a factor that limits the prolonged use of this potentially lifesaving treatment. The aim of this paper is to provide a comprehensive review of ICI-induced liver toxicity and highlight several clinically relevant issues that need to be addressed in the future (Table 1).

Immune checkpoint inhibitors: a recent and exponential increase in use

ICIs are becoming increasingly widely used in onco-haematology. Their safety profile, at least as monotherapy, is generally better than that of cytotoxic chemotherapy,⁵ and the quality of the antitumor response is generally more prolonged and lasting.⁶ Long-term follow-up – now more than 5 years – has indicated that if a complete response is obtained and sustained after 2 years of treatment, patients with metastatic melanoma can almost be considered cured.⁷

Since 2015, ICIs targeting programmed cell death 1 (PD-1) and its ligand (PD-L1) have displayed remarkable efficacy in the treatment of non-operable locally advanced or metastatic cancers of many types, including lung cancer, melanoma, kidney cancer, head and neck cancer, tumours with micro-satellite instability, Merkel cell carcinoma, epidermoid skin cancer, Hodgkin's lymphoma, B cell primary mediastinal lymphoma and more recently hepatocellular carcinoma.^{8–10}

It is likely that the use of ICIs will continue to grow in the coming years. Indeed, therapeutic strategies including ICI are improving and developing in the context of adjuvant or neoadjuvant treatments.⁸

Keywords: Immunotherapy; Immune-mediated hepatitis; Liver biopsy; Corticosteroid therapy

Received 6 June 2020;
received in revised form 23
July 2020; accepted 30 July
2020 Available online 11
August 2020

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Finally, evidence is accumulating on the role of the anti-PD-1 and anti-cytotoxic T lymphocyte-associated protein 4 (CTLA-4) antibody combination in patients with hard-to-treat tumours such as metastatic melanoma, kidney cancer, mesothelioma and lung cancer.⁸ Because of this increasing use, the cumulative number of irAEs is expected to rise exponentially,¹¹ and the frequency of autoimmune events related to immunotherapy is likely to exceed the incidence of classic autoimmune diseases unrelated to immunotherapies.¹²

General mechanism of action underlying ICI toxicity

Several mechanisms have been explored to explain ICI-related toxicity. The first is direct immune toxicity. PD-1 and PD-L1 are expressed on cells in healthy tissue, suggesting that direct cytotoxicity after immunotherapy is possible, for example by means of complement activation.^{11,13} CTLA-4 is strongly expressed in the anterior pituitary gland and immune toxicity correlates with this finding as hypophysitis is principally seen with ipilimumab but not with PD-1 or PD-L1 inhibitors.¹⁴ In addition, myocardial PD-L1 is mainly localised on the endothelium and is critical in controlling immune-mediated ICI-induced cardiac injury.¹⁵ Secondly, B cells also play a role in the development of ICI-related toxicity, as early B cell changes can be observed in the blood of patients treated with ICIs who experience higher rates of grade ≥ 3 irAEs.¹⁶ Such early changes following immunotherapy may give rise to autoreactive B cells and through an immune-mediated reaction probably participate in generating irAEs.¹⁶

Thirdly, the gut microbiota seems to influence the onset of irAEs, and particularly colitis. Chaput *et al.* demonstrated that a specific composition of the baseline gut microbiota was associated with immune-related colitis.¹⁷ Most baseline colitis-associated phylotypes were related to Firmicutes (*e.g.* relatives of *Faecalibacterium prausnitzii* and *Gemmiger formicilis*), whereas none were assigned to Bacteroidetes.¹⁷ Fourthly, cross reactivity between tumour cell antigens and normal tissue clinically translates as paraneoplastic syndromes in some cases.¹⁸ Pre-existing paraneoplastic syndromes have been shown to worsen in 50% of patients treated with anti-PD-1 or anti-PD-L1 immunotherapy.¹⁸ Finally, regulatory T (Treg) cell depletion has been suggested to play a role in the development of irAEs, as Treg cells are essential for maintaining peripheral tolerance.¹⁹ CTLA-4 is constitutively expressed on Treg cells while PD-1 expression is restricted to subpopulations. CTLA-4 blockade can affect Treg cell number and function; reduced Treg cell number and increased effector T cells to Treg cells ratio have been observed in patients treated with ipilimumab. Preclinical models of irAEs have shown a negative correlation between the Treg cell number and irAEs.²⁰ The specific pathogenic mechanisms underlying hepatic irAEs are still unclear.

What are the most relevant biomarkers to predict immune-related toxicity in clinical practice?

Data suggest that detecting pre-existing antibodies could be useful to predict the risk of certain irAEs. It has been reported that patients with pre-existing thyroglobulin antibody in baseline serum are at a high risk of thyroid dysfunction during immunotherapy.²¹ Similarly, more patients developed irAEs if they had pre-existing autoantibodies and rheumatoid factor and were being treated with immunotherapy.²²

Key points

- Liver toxicity is a rare complication of immune checkpoint inhibitors with a heterogeneous presentation and prognosis.
- It is more common and severe under combination therapy with anti-PD-1 and anti-CTLA-4 drugs.
- Diagnosis is initially based on excluding all classic causes of acute hepatitis.
- Liver histology is important to identify the characteristic histological features of ICI-induced toxicity and to assess the severity of liver tissue damage.
- Classification of the severity of liver toxicity needs to be revised to consider the clinical expertise of hepatologists.
- The administration of corticosteroids should be tailored to individuals and might not be systematic, even in patients with grade ≥ 3 hepatitis.
- We currently lack predictive clinical or biological factors for a recurrence of ICI-related hepatitis or other immune toxicities affecting other organs after ICI reintroduction.
- The management of liver toxicity due to immune checkpoint inhibitors requires multidisciplinary discussions.

As previously discussed, some changes to B cell lymphocytes may be predictive biomarkers of immune-related toxicity.¹⁶ The deep phenotyping of immune cells showed that immune toxicities elicited by CTLA-4 blockade in cancer patients were found to be associated with an early diversification of the T cell repertoire in peripheral lymphocytes.²³ Changes to the neutrophil-to-lymphocyte ratio (NLR) in peripheral blood could be a powerful tool to predict the efficacy and toxicity of immunotherapy.²⁴ Low NLR values at baseline were significantly associated with the development of irAEs.²⁵ Thus, NLR may represent a simple biomarker that can easily be applied for the management of patients receiving immunotherapy in daily practice.

Interleukin (IL)-17 has been found to correlate significantly with the incidence of grade 3 diarrhoea/colitis when measured at baseline in patients receiving immunotherapy.²⁶ IL6 was also found to be associated with the risk of immune toxicity.²⁷ Another study found 11 cytokines that were significantly upregulated in patients with severe immune-related toxicities; they included the pro-inflammatory cytokines IL1a, IL2, and IFN α 2.²⁸ The sum of these data suggests that the profile of cytokines and their expression could be integrated in a score to help with the early detection of severe, potentially life-threatening immune-related toxicity.²⁸

Patients of European descent with immune-related arthritis are more likely to carry the HLA-DRB1*04:05 phenotype²⁹ and other studies have identified an association between HLA-DQB1*03:01 and immune-related colitis.³⁰ The HLA phenotype in some populations could constitute a biomarker to predict immune-related toxicity, but although powerful, its efficiency in all patient populations remains a subject of debate.¹¹ Despite these promising results (Table S1), potential biomarkers to predict toxicity and in particular liver toxicity in clinical practice are still needed.

Incidence of liver toxicity due to immune checkpoint inhibitors

Liver toxicity associated with immune checkpoint inhibitors is characterised by elevated liver parameter values, usually those of aspartate aminotransferase (AST) and alanine aminotransferase (ALT). Drug-induced liver toxicity is classified according to the

Table 1. Open questions, which need to be addressed in order to improve the management of ICI-induced liver toxicity.

Open questions	Impact of answers and future studies goal
What is the pathogenesis of immune-mediated hepatitis in normal liver and cirrhotic liver?	Identify predictors of toxicity and diagnostic markers
What are the best markers to evaluate the severity of ICI-induced liver toxicity?	Avoid overestimation of severity and identify the patients in need of corticosteroids
Do corticosteroids shorten the interval to liver test normalisation compared to spontaneous remission?	Decide on corticosteroid introduction and tapering
Does immunosuppression have an impact on ICI efficacy?	Modulate the dose of immunosuppressive therapy. Define relationship between immunosuppression and type of cancer
Is UDCA enough to treat ICI-induced cholangitis?	Avoid unnecessary corticosteroid therapy
What are the criteria to safely re-introduce an ICI after an episode of immune-mediated hepatitis?	Identify biomarkers of activity resolution and risk factors for recurrence

ICI, immune checkpoint inhibitor; UDCA, ursodeoxycholic acid.

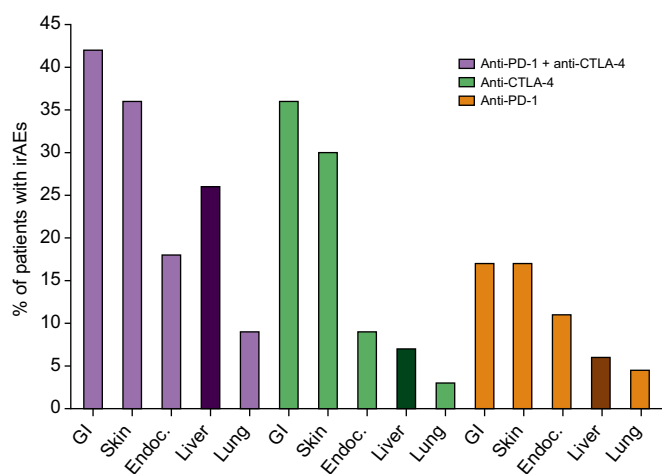


Fig. 1. Distribution of irAEs for organ categories according to treatment in the main clinical trials of ICIs. Patients were treated with anti-PD-1 + anti-CTLA-4,^{4,33,36} anti-CTLA-4^{4,33,104,124} and anti-PD-1.^{33,34,124} The values quoted are mean irAE rates of clinical trials as a whole. CTLA-4, cytotoxic T lymphocyte-associated protein 4; Endoc, endocrine (hypo/hyper-thyroidism); GI, gastrointestinal (diarrhoea); ICI, immune checkpoint inhibitors; irAEs, immune-related adverse events; Liver (increased alanine aminotransferase); PD-1, programmed cell death 1; PD-L1, programmed cell death 1 ligand 1; Skin (rash).

pattern of elevation of liver enzymes based on the first set of laboratory tests available in relation to the clinical event. This pattern is defined by the increase of ALT or alkaline phosphatase (ALP) alone above a specific threshold or by the ratio of serum ALT to ALP levels ($R \text{ value} = [\text{ALT}/\text{upper limit of normal (ULN)}] / [\text{ALP}/\text{ULN}]$) and can be categorised as hepatocellular (ALT ≥ 5 -fold above ULN or $R > 5$), mixed ($R > 2$ to < 5), or cholestatic (ALP ≥ 2 -fold above ULN or $R < 2$).³¹ The pattern of ICI-induced liver toxicity is heterogeneous; it may be cytolytic, mixed or cholestatic, although the incidence of cholestasis seems to be lower. Liver toxicity depends on the type of immunotherapy, the dose and baseline liver status. Overall, the incidence of liver toxicity is higher in patients who receive combination therapy than in those under monotherapy, but it remains lower compared to other organ toxicities (Fig. 1). Moreover, elevations of AST and ALT were shown to be more frequent among patients treated for HCC with chronic hepatitis or cirrhosis compared with those treated for non-liver cancers such as melanoma or non-small cell lung cancer, although these elevations did not require patients to withdraw from clinical trials or stop therapy.³²

Patients treated for non-liver cancers

Liver toxicity can occur in patients receiving ICIs for non-hepatic malignancies. For example, in patients treated with anti-PD-1 monotherapy for advanced melanoma, elevated ALT levels of any grade were reported in 4% to 7% of patients, while a grade ≥ 3 elevation of ALT was reported in 1% of patients.^{33,34} In the same study, among patients treated with anti-CTLA-4 monotherapy, ALT elevations of any grade and of grade ≥ 3 were found in 4% and 2% of patients, respectively.³³ The incidence of liver toxicity increases in patients who receive combination therapies. In patients treated with anti-PD-1 in combination with a tyrosine kinase inhibitor (TKI) for advanced renal cell cancer, ALT elevations of any grade were reported in 27% of patients while ALT grade ≥ 3 elevations were found in 13% of patients.³⁵ Finally, in patients receiving a combination of anti-PD-1 and anti-CTLA-4 therapy for metastatic melanoma, ALT elevations of any grade and of grade ≥ 3 were reported in 37% and 16% of patients, respectively³⁶ (Fig. 2A and B).

Patients treated for hepatocellular carcinoma

Among patients who receive ICI therapy for HCC, the incidence of liver toxicity varies as a function of the type of drug and dose received. If we look at the overall population treated with the anti-PD-1 antibody nivolumab (CHECKMATE 040 trial), ALT elevations of any grade and of grade ≥ 3 were found in 15% and 6% of patients, respectively.³⁷ In those receiving another anti-PD-1 antibody pembrolizumab (KEYNOTE-224 trial), ALT elevations of any grade were seen in 9% of patients, while those of grade ≥ 3 were observed in 4%.³⁸ Use of the anti-CTLA-4 antibody tremelimumab was associated with ALT elevations of any grade and of grade ≥ 3 in 19% and 9% of patients, respectively. Nevertheless it is important to remember that immunotherapy was associated with ablation in this study.³⁹ In patients treated with a combination of nivolumab + ipilimumab, the rise in ALT levels of any grade ranged from 8% to 16% as a function of the dose administered, while ALT grade ≥ 3 elevations ranged from 0% to 8%.⁴⁰ These are preliminary data and ongoing trials are evaluating the efficacy and safety of anti-PD-1 and anti-CTLA-4 combination therapy. In our experience this combination is associated with more liver toxicity than that previously described in patients treated for other malignancies. In those receiving the combination of tremelimumab + durvalumab, elevated ALT levels of any grade were seen in 20% of patients, while grade ≥ 3 was seen in 5% of patients.⁴¹ Interestingly, the combination of anti-PDL-1 (atezolizumab) and a vascular endothelial growth factor (bevacizumab), as described in the IMbrave150 trial, was not

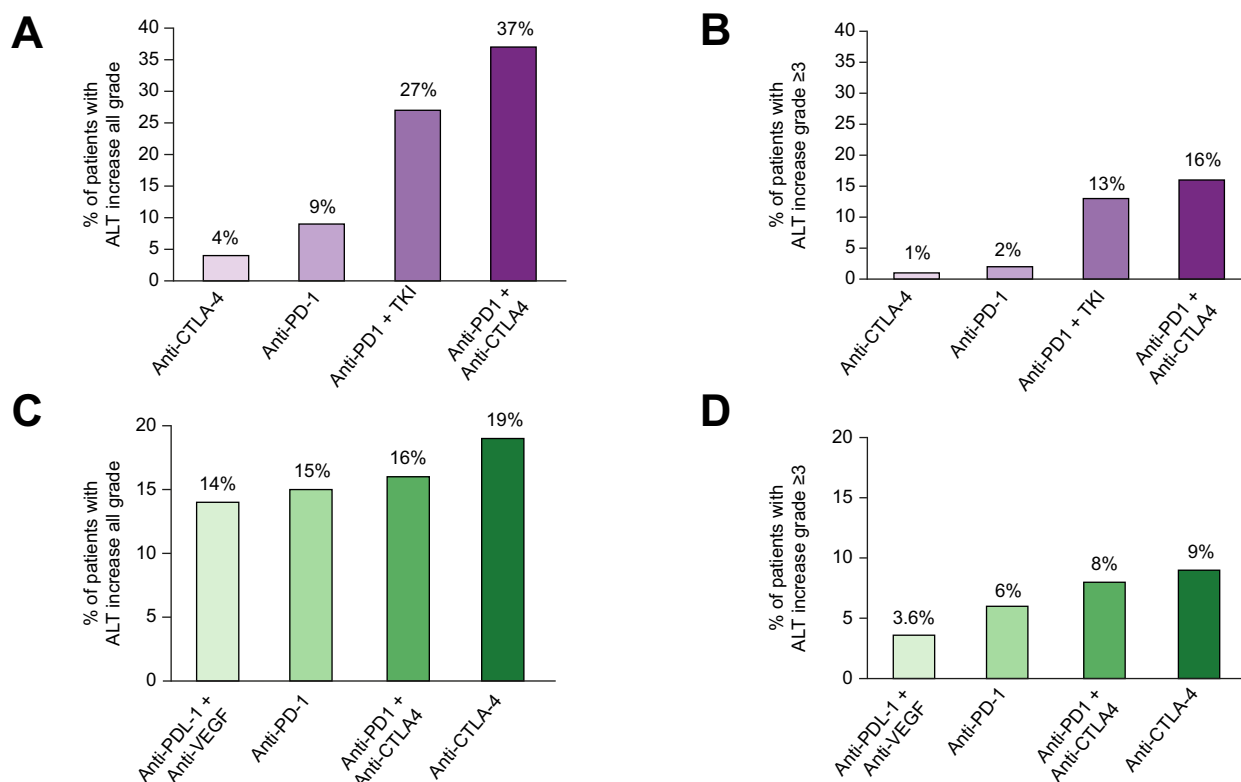


Fig. 2. Distribution of ALT increase in patients treated for non-HCC malignancies and HCC according to treatment in the main clinical trials of ICIs. Patients with non-HCC malignancies were treated with anti-CTLA-4³³ anti-PD-1,³⁴ anti-PD-1 + TKI,³⁵ anti-PD-1 + anti-CTLA-4,³⁶ while patients with HCC were treated with anti-PD-L1 + anti-VEGF,⁹ anti-PD-1,³⁷ anti-PD-1 + anti-CTLA-4,⁴⁰ anti-CTLA-4.³⁹ (A) Distribution of ALT increase of all grades in non-HCC patients; (B) Distribution of ALT increase of grade ≥ 3 in non-HCC patients; (C) Distribution of ALT increase of all grade in HCC patients; (D) Distribution of ALT increase of grade ≥ 3 in HCC patients. ALT, alanine aminotransferase; CTLA-4, cytotoxic T lymphocyte-associated protein 4; HCC, hepatocellular carcinoma; ICI, immune checkpoint inhibitor; PD-1, programmed cell death 1; PD-L1, programmed cell death 1 ligand 1; TKI, tyrosine kinase inhibitors; VEGF, vascular endothelial growth factor.

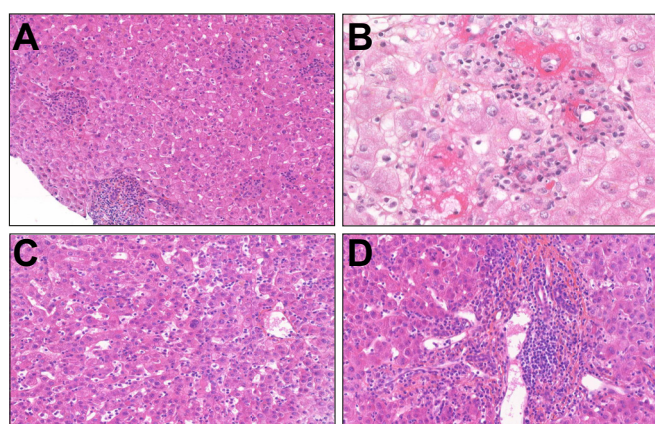


Fig. 3. Liver biopsies of patients treated with anti-CTLA-4 or anti-PD-1 alone or in combination. (A) Microgranulomatous hepatitis (HES $\times 120$); (B) Hepatitis with fibrin-ring granulomas (HES $\times 370$); (C) Non-granulomatous lobular hepatitis (HES $\times 190$); (D) Lobular and portal hepatitis with endotheliitis (HES $\times 190$). CTLA-4, cytotoxic T lymphocyte-associated protein 4; PD-1, programmed cell death 1.

associated with increased liver toxicity; ALT elevations of all grades and grade ≥ 3 were seen in 14% and 3.6% of patients, respectively⁹ (Fig. 2C and D).

Patients with pre-existing chronic liver disease

Before starting an ICI, a complete work-up should be performed to rule out misdiagnosed liver disease (Table S2).

Viral disease

Most of the clinical trials that have explored the safety and efficacy of ICI therapy for cancer excluded patients with HBV, HCV and HIV infections. Only studies testing immunotherapy for HCC included patients with viral hepatitis. Most patients with HBV hepatitis were receiving effective antiviral therapy with nucleos(t)ide analogues and had a viral load < 100 IU/L at screening, and no HBV reactivation was observed.^{37,39,42} In contrast, a reduction in viral load was documented in some cases. Outside clinical trials, only 3 case reports have described HBV reactivation in patients not receiving HBV treatment, all of whom were treated successfully with antiviral therapy.^{43–45} During HCC clinical trials^{37–39,42,46} and in a few case reports,^{47,48} HCV RNA-positive patients displayed a reduction in viral load during immunotherapy. Therefore, patients with viral hepatitis are not at a higher risk of liver toxicity than those without viral hepatitis. All patients need to be screened for HBV and HCV infection before ICI administration, but the presence of HBV or HCV is not a contraindication to mono- or even combination therapy using ICIs. Serological tests for HBV should be carried out using a complete diagnostic panel (HBsAg, HBs antibody, HBe antibody, and HBV DNA, when appropriate). Patients with active HBV

infection (HBsAg positive) with both positive and negative viral load, independently of HBeAg status, should receive effective antiviral therapy with a nucleos(t)ide analogue in order to avoid viral reactivation. It is difficult to determine the duration of prophylaxis as ICI activity following withdrawal may be variable, but we agree with Lombardi *et al.* that it should be maintained for at least 6 months after the end of ICI treatment.⁴⁹ For patients with a resolved HBV infection (anti-HBc positive) strict monitoring can be suggested. By contrast, patients with HCV infection do not require antiviral therapy but need to be monitored regularly for HCV replication. For patients with HIV, a recent review showed that there was no increase in hepatic side effects, so ICIs can also be considered a therapeutic option for these patients.⁵⁰ Indeed, a few ongoing trials are now including HIV-infected patients.

Autoimmune disease

No data have been reported concerning patients with pre-existing autoimmune liver diseases treated with ICI. Some reports have focused on patients with previous autoimmune diseases, but the incidence of adverse events in this subgroup cannot be evaluated precisely because the studies are all retrospective. A study based on the REISAMIC registry (Institut Gustave Roussy, France) identified 45 patients with 54 known autoimmune or inflammatory diseases (AIDs) treated with an anti-PD-1; the most frequent AIDs were vitiligo, psoriasis, thyroiditis, Sjögren syndrome and rheumatoid arthritis. As expected, the study revealed that patients with a pre-existing AID had a significantly higher risk of irAEs (44%), but anti-PD-1 treatment in this group of patients was as effective as in AID-free patients.⁵¹ In another report on 41 patients with 44 pre-existing AIDs treated with ipilimumab, 12 (29%) experienced a flare-up of the AID and an additional irAE occurred in 12 patients (29%). The response rate was comparable to that seen in previous trials.⁵² A systematic review evaluated the outcomes of 123 patients with pre-existing AIDs and found that an exacerbation of the AID, the onset of *de novo* irAEs or both occurred in 75% of patients. Most of the adverse events were managed successfully with corticosteroids and only 16% of patients required an alternative immunosuppressive drug; the death rate due to an adverse event was 2.4%.⁵³ These data were also confirmed by a French multicentre study that included 112 patients; 71% of them experienced AID flare-ups or *de novo* irAEs that were generally manageable without discontinuing ICI. Interestingly, ongoing immunosuppressive therapy at the initiation of ICI was associated with a poorer outcome.⁵⁴ Therefore the presence of a pre-existing AID should not be considered as a contraindication to ICI therapy, but patients should be monitored closely as they are at high risk of a flare-up of the previously known autoimmune disease and/or of developing *de novo* irAEs. Whether these general findings can be transposed to patients with autoimmune liver diseases still needs to be clarified.

Liver transplant recipients

ICI therapy can be indicated after liver transplantation (LT) for the treatment of HCC recurrence or *de novo* malignancies. LT recipients were excluded from clinical trials based on the association of ICI with acute rejection and the risk of graft loss.^{55,56} A total of 11 patients who were treated with ICIs after LT were reported in the literature, among them 4 (36%) patients developed acute rejection with or without graft loss.⁵⁷ Of note, this

case series is extremely heterogeneous concerning the interval between LT and the introduction of immunotherapy, the immunosuppression protocol and the type of ICI used. In 5 (45%) patients, immunotherapy was administered to treat HCC recurrence after LT. In a recently published review of the literature, the incidence of acute rejection in LT recipients treated with ICIs was reported to be 39%.⁵⁸ PDL-1 expression in the allograft seems to be correlated with rejection,^{59,60} although a panel of validated risk factors is lacking. However, safe employment of immunotherapy after LT has also been described.^{61–63} Although the risk of rejection remains elevated it seems inconsistent to deny this treatment to patients without other therapeutic options in the absence of evidence-based trials.

Another issue that needs to be addressed is the identification of the optimal time between ICI therapy and LT in patients treated for HCC. LT was successfully performed in a patient who underwent HCC downstaging with nivolumab.⁶⁴ However, another study reported the fatal liver necrosis of a patient who was treated with nivolumab for HCC and underwent LT 8 days after ICI discontinuation, indicating that this interval was probably too short.⁶⁵ As the duration of ICI activity, which is longer than the half-life of the agent, is not known, the time elapsing between treatment discontinuation and a safe transplant needs to be defined.

Diagnosis of liver toxicity

Patient characteristics

Patients with liver toxicity on ICIs are usually asymptomatic and the symptoms, when present, are particularly non-specific; they may present with fever, skin rash and, in rare cases, jaundice.^{66,67} Symptoms deriving from concomitant toxicity affecting another organ, such as colitis, hypophysitis or pneumonitis, may also occur.⁶⁸

No male or female preponderance has been described and age does not appear to constitute a risk factor for the development of liver toxicity.^{66,67}

The interval elapsing between the initiation of therapy and the onset of liver toxicity varies considerably, and toxicity may even occur after treatment discontinuation. In patients treated with ipilimumab as adjuvant therapy for stage III melanoma, the median time to the onset of hepatic irAE was 8.9 weeks (ranging from 1.9 weeks to 145.4 weeks).⁶⁹ It appears that patients treated with anti-CTLA-4 (with or without anti-PD-1) experience a shorter interval between immunotherapy initiation and hepatic irAE development compared to those receiving anti-PD-1 or anti-PD-L1, with a median interval of 3 (1–7) weeks vs. 14 (2–49) weeks ($p = 0.019$).⁶⁶ This was confirmed in the study by Gauci *et al.* who showed a median time to the onset of hepatitis of 9.9 weeks (2.9–19.7); 9.9 (6.1–14.7) weeks for patients treated with anti-CTLA-4 and 14.1 (9.4–19.7) weeks for patients under anti-PD-1.⁷⁰ More recently, a study from the VigiBase, <http://www.vigiaccess.org/>, the World Health Organization database for individual safety case reports, also found a significantly earlier onset of hepatitis in patients treated with anti-CTLA-4 antibodies (34 [25–46.5] days) compared to those receiving anti-PD-1/PD-L1 antibodies (48 [27–118] days) ($p = 0.04$).⁷¹ In the paper by Riveiro-Barciela, the interval between ICI treatment initiation and liver toxicity was shorter in patients on anti-CTLA-4 than in those receiving anti-PD-1/PD-L1 antibodies, although the difference was not statistically significant.⁶⁷ The combination of anti-CTLA-4 and anti-PD-1 was reported not to be associated

with an earlier onset of grade ≥ 3 liver toxicity in a study by Szol et al., who reported a median interval of 8.4 (2.1–48) weeks,⁷² while it was significantly shorter (median 2.9 weeks) with the combination therapy in the study by Gauci et al.⁷⁰

In most patients, the profile of liver injury is usually hepatocellular, but cases of cholestatic presentation have been described.^{66,73,74} Seven patients with nivolumab-induced cholangitis with predominant ALP and GGT elevations have been reported. In these cases, no increase in IgG4 was found.^{75–79} The presence of non-specific anti-tissue antibodies was described in 30% to 50% of the patients but at a low titre (1:80), while IgG levels were usually normal.^{66,67} It should be pointed out that the immune-mediated hepatitis induced by ICI is an entity that is entirely different from autoimmune hepatitis (AIH). This was clearly shown in a recent study where patients with AIH were younger (median of 55 vs. 63 years, $p = 0.02$), presented more frequently with previous autoimmune disorders, and more frequently had cirrhosis, a lower platelet count, higher bilirubin levels and higher gamma globulin levels than patients with liver toxicity. Patients with a diagnosis of AIH were also more numerous in needing a second immunosuppressive drug and their liver test values took longer to normalise than those with liver toxicity.⁶⁷ Moreover, liver histological features were also completely different between the 2 groups, as described below.

Cases of acute liver failure with hepatic encephalopathy remain rare.^{80–82} Of note are data from the Vigilyze-VigiBase, the World Health Organization pharmacological database, which reported a 0.4% incidence of fulminant hepatitis.⁸³ In a multi-centre study that included 3,545 patients, fulminant hepatitis was reported in 0.14% of them.⁸³ Interestingly, in trials involving HCC, none of the patients receiving ICIs experienced severe acute liver failure.

Causality assessment

Because there are no specific biomarkers to distinguish liver toxicity induced by ICIs, it is essential to exclude all classic causes of hepatitis. All patients with grade ≥ 3 hepatitis should be evaluated by a hepatologist and undergo a comprehensive work-up to investigate different causes of acute hepatitis and misdiagnosed chronic hepatitis. (Table S3) The causality between the drug and the onset of hepatitis needs to be evaluated using a scale such as the Roussel-UCLAF Causality Assessment Methods (RUCAM) scale to assess the likelihood of an association between a drug and liver toxicity.^{84,85}

Imaging

Imaging forms part of the diagnostic work-up for ICI-induced liver toxicity. The aim of imaging is to exclude the presence of hepatic metastases, vascular thrombosis, biliary obstruction and features of chronic liver disease.⁸⁶ Despite the limited number of patients studied, unspecific abnormalities, such as steatosis, hepatomegaly, periportal oedema, gallbladder oedema and lymphadenopathy, have been described using ultrasound in patients treated with anti-PD-1 agents.⁸⁷ These findings were reported in patients treated with an anti-CTLA-4 antibody.⁸⁸ In rare cases, patients with a cholestatic profile may display macroscopic involvement of the bile ducts.⁷⁹ Early biliary MRI plus elastometry data are also essential and currently lacking.

Histology

A liver biopsy may confirm a suspected diagnosis of immune-mediated hepatitis and can be used to evaluate the features

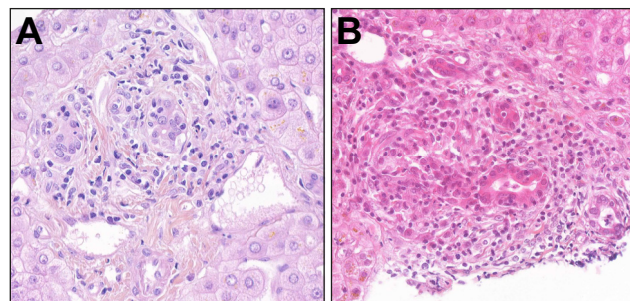


Fig. 4. Liver biopsies of patients treated with anti-PD-L1 and with combination of anti-PD-1 and anti-CTLA-4. (A) Destructing cholangitis (HES $\times 400$); (B) Granulomatous cholangitis HES $\times 300$. CTLA-4, cytotoxic T lymphocyte-associated protein 4; PD-1, programmed cell death 1; PD-L1, programmed cell death 1 ligand 1.

and severity of liver tissue damage, as well as to rule out any underlying misdiagnosed chronic liver disease. The data concerning the pathological features of ICI-induced hepatitis are scarce. The most common patterns include acute hepatitis with spotty or confluent necrosis that predominates in the centrilobular zone, and granulomatous hepatitis.^{66,73,89,90} One hallmark that characterises patients under anti-CTLA-4 treatment (as monotherapy or in combination with another ICI) is the presence of poorly defined granulomas, including fibrin-ring granulomas^{66,73} (Fig. 3A and B). Others have also reported the presence of such fibrin-ring granulomas in patients treated with the combination of ipilimumab and nivolumab.⁹¹ Interestingly, fibrin-ring granuloma was also found in a patient who received nivolumab as monotherapy, but whether he had previously been exposed to ipilimumab was not clear.⁹² In the event of treatment with anti-PD-1 or PD-L1, there are no specific histological features. Lobular hepatitis with polymorphic infiltration and the clarification of hepatocytes have been described (Fig. 3C and D). Macrovesicular steatosis and steatohepatitis have also been reported but these features are difficult to attribute to ICI.⁹³

Bile duct injury has been described in some cases. Features of acute cholangitis with mixed inflammatory infiltrate (rich in polynuclear neutrophils) have been reported,^{79,89} as well as lymphocytic cholangitis with bile duct dystrophy and bile duct proliferation^{66,75,88} (Fig. 4A and B). Immunohistochemical characterisation revealed a predominance of inflammatory T lymphocyte infiltration with a prevalence of CD8+ cytotoxic elements, particularly in patients treated with anti-CTLA-4 antibodies, while those receiving anti-PD-1/anti-PD-L1 antibodies had a more mixed CD8+/CD4+ inflammatory infiltrate.^{79,89,90,94}

A case report found a nodular regenerative hyperplasia in the biopsy of a patient treated with pembrolizumab for advanced melanoma.⁹⁵ The patient presented with diffuse anasarca and ascites requiring paracentesis twice a week. Portal pressure gradient was 16 mmHg. Reticulin staining revealed nodular transformation with compressed and atrophic hepatocytes between nodules and portal vein stenosis. Immunohistochemical staining for CD34 showed aberrant capillarisation in periportal sinusoids. A transjugular intrahepatic portosystemic shunt was placed, leading to clinical improvement.

Importantly, ICI-induced liver toxicity does not display the usual features of AIH (*i.e.* no plasma cell infiltration in liver biopsies from 5 patients treated with ipilimumab).⁷³ Zen et al.

compared the histological features of patients with drug-induced liver injury (DILI), AIH and immune-mediated hepatitis due to ICI and found significant differences regarding the amounts of confluent necrosis (which were lower in patients with ICI-induced hepatitis), eosinophilic infiltration (higher in patients with DILI) and plasmacytosis (higher in patients with AIH).⁹⁴ The shift of CD3+/CD20+ and CD4+/CD8+ ratios in favour of CD8+ cytotoxic T lymphocytes could be useful in the event of irAEs and helpful to discriminate them from other conditions such as AIH.⁹⁶ Differences between ICI-induced liver toxicity and AIH are depicted in Table 2.

Whether immune-mediated hepatitis results in the development of fibrosis is still unknown. Kleiner and colleagues found some degree of fibrosis in 2 out of 5 patients treated with ICI: one patient displayed perisinusoidal and periportal fibrosis, and the other post-necrotic fibrosis (which sometimes follows acute hepatitis).⁷³ In our series of 16 patients, we observed only one case where the patient was affected by grade 4, difficult-to-treat hepatitis and developed portal fibrosis. The course of this disease was followed by performing 3 serial liver biopsies.⁹⁰

No histological findings have been reported to date in patients with liver toxicity developing in a cirrhotic liver. In our very preliminary experience, we found similar histological features in cirrhotic liver as observed in non-cirrhotic liver.

Should a liver biopsy form part of the work-up? Indeed, histology can offer much information regarding the diagnosis of ICI-related hepatitis and the severity of liver tissue injury; together with clinical and biological features, a biopsy can guide the decision on whether or not to introduce corticosteroids. That said, at present, a liver biopsy should be reserved for patients with more severe liver toxicity (grade ≥ 3) or an uncertain diagnosis. While liver toxicity under ICI remains poorly understood, a biopsy may be useful to improve our knowledge and thus optimise management.

Evaluation of severity

Evaluating the severity of clinical liver toxicity is of primary importance as it will guide patient management. The method used to grade toxicity, applied during all clinical trials of ICI and used by oncologists in clinical practice, is the Common Terminology Criteria for Adverse Events (CTCAE) developed by the National Cancer Institute (Table S4).

However, this classification may not always be adequate to evaluate liver toxicity as it may overestimate the severity of the disease. This is particularly true when it is compared to the drug-induced liver injury network (DILIN) classification.⁹⁷ For example, under the CTCAE classification, an elevation of transaminases to more than 20x the upper limit of normal without a rise in bilirubin levels or an impairment of coagulation is considered as grade 4 toxicity, which corresponds to a life-threatening event. From a hepatological standpoint, acute hepatitis is considered severe if the INR is ≥ 1.5 , and fulminant if impaired coagulation is accompanied by hepatic encephalopathy.⁹⁸ Histological findings do not always reflect the clinical severity of ICI-related toxicity when compared to other chronic liver diseases.

Management

According to the guidelines of the Society for Immunotherapy of Cancer (SITC) and the European Society of Medical Oncology (ESMO), liver toxicity should be treated by pausing

immunotherapy and administering corticosteroids from grade 2 liver injury.^{99,100} The corticosteroid dose may rise in proportion to the grade of hepatitis up to a maximum of 2 mg/kg/day. Immunotherapy should be withdrawn temporarily in the case of grade 2 and 3, but permanently discontinued in the event of grade 4 hepatitis. This approach was confirmed by more recent guidelines from the American Society of Clinical Oncology (ASCO).¹⁰¹ However, several teams have demonstrated that this approach is not supported by clinical evidence and liver test findings may rapidly improve when immunotherapy is stopped, without the addition of corticosteroids.^{66,70} This is particularly important in view of the increased infection risk in patients receiving ICI.¹⁰² Treatment with budesonide, a drug with 90% hepatic clearance and metabolism, has not been evaluated.

The time to resolution of non-hepatic irAEs in patients under corticosteroids is generally around 2 weeks.¹⁰³ The time to resolution of hepatic irAEs varies considerably; it has been reported as ranging from 3 to 104 days in different case reports and series.^{68,70,74,104–108} Corticosteroids at doses higher than 60 mg/day appear to have no benefit regarding the time to the resolution of hepatitis when compared to 1 mg/kg/day or higher.⁷⁴ Unexpectedly, Gauci *et al.* reported a longer time to resolution in patients who received corticosteroids than in those who did not (median 8.6 vs. 4.7 days), probably because the hepatitis was more severe in the former group.⁷⁰

Patients with cirrhosis present a different scenario because their liver function may be impaired before ICIs are initiated. A management algorithm for these patients has recently been proposed.¹⁰⁹ Even in the setting of cirrhosis, corticosteroid therapy is not always recommended. Liver tests are usually abnormal in patients with cirrhosis before the introduction of ICIs, so the severity of toxicity and the decision regarding whether to introduce corticosteroids is based on the gradual rate of transaminase elevation or the presence of features of hepatic failure.

Patients with hepatitis refractory to corticosteroids

Some patients whose liver function tests worsen despite adequate corticosteroid therapy are considered refractory to this treatment. In these cases, a second immunosuppressive drug can be added, the most widely used being mycophenolate mofetil (MMF).^{66,74,105,107,108,110,111} Azathioprine has also been used with success.^{68,106} In a few clinical observations, the use of calcineurin inhibitors (cyclosporine and tacrolimus) has also been described as effective.⁶⁸ The use of thymoglobulin combined with methylprednisolone and MMF for refractory hepatitis has been reported in 4 patients.^{108,112,113}

Infliximab therapy was used successfully in 2 patients,⁷⁴ but it is not the best second-line immunosuppressive option in view of potential liver toxicity and increased risk of infection in the setting of prolonged corticosteroid therapy.

Proposed management algorithm

In 2018, we proposed a management protocol for patients who experience severe liver toxicity due to ICI administered for non-liver malignancies.⁶⁶ Based on our experience and on the recent literature, this algorithm has since evolved and now integrates recommendations regarding the reintroduction of immunotherapy (Fig. 5 and Table S5).

The decision to start corticosteroids is based on: worsening of liver tests, histological confirmation of hepatic irAEs and severity, elevation of bilirubin ≥ 2.5 mg/dl and impaired coagulopathy proven by an INR ≥ 1.5 , which is the cut-off defining the severity

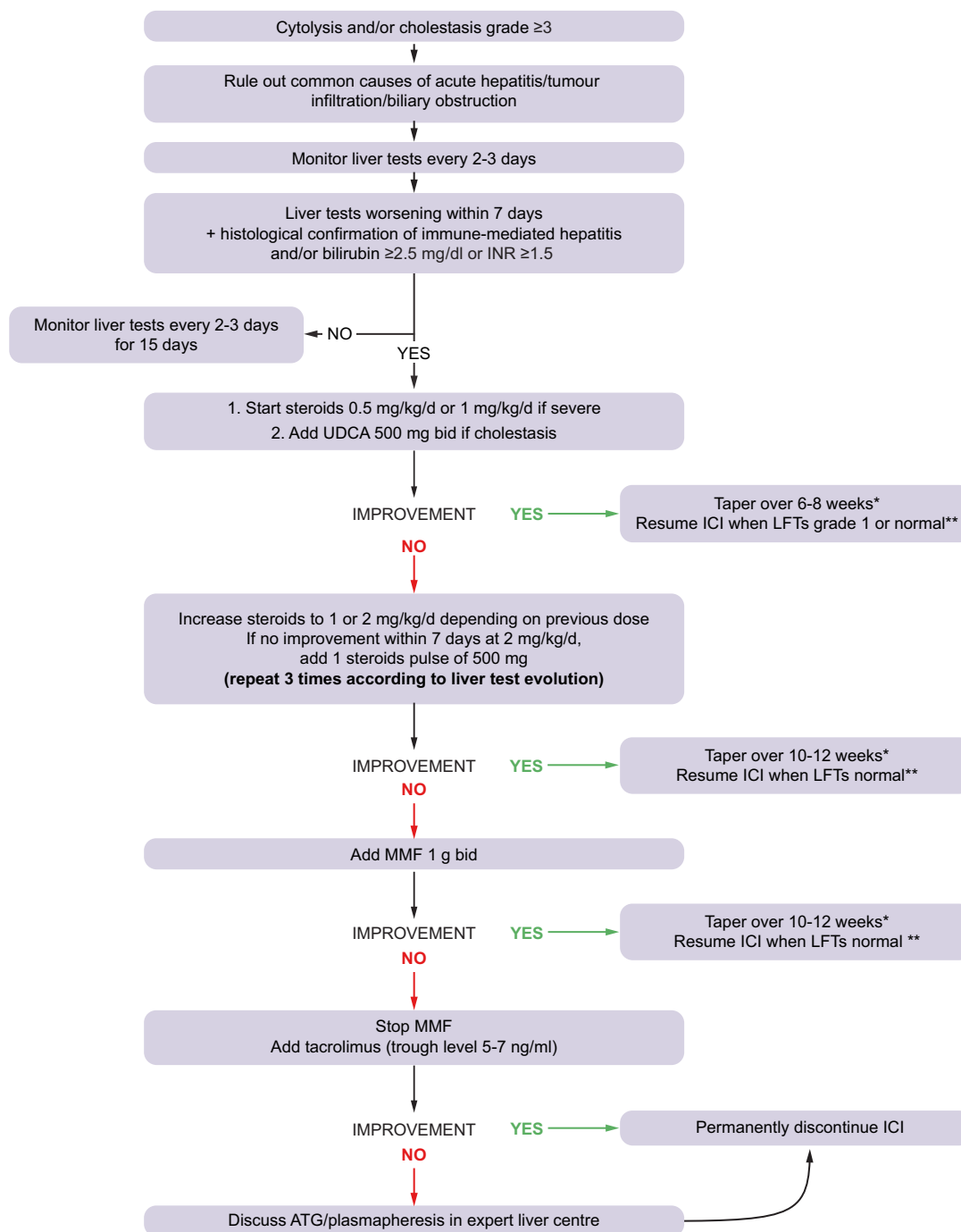


Fig. 5. Proposal for the management of patients who experience ICI-related grade ≥ 3 hepatitis. *Steroid tapering proposal see Table S2. **Wait until MMF and steroid discontinuation. Resume anti-PD-1 or anti-PD-L1 alone; permanently discontinue anti-CTLA-4; if UDCA administered resume ICI under UDCA. CTLA-4, cytotoxic T lymphocyte-associated protein 4; ICI, immune checkpoint inhibitor; LFTs, liver function tests; MMF, mycophenolate mofetil; PD-1, programmed cell death 1; PD-L1, programmed cell death 1 ligand 1; UDCA, ursodeoxycholic acid.

of acute hepatitis. Corticosteroid therapy is clearly indicated in these patients. Ursodeoxycholic acid (UDCA) may be added in those with biological features of cholestasis. In patients whose liver tests do not improve under corticosteroids despite increasing doses, MMF should be introduced. In patients not

responding to MMF, tacrolimus with a trough level of between 5–7 ng/dl can be used. In exceptional cases of hepatic encephalopathy, plasmapheresis or anti-thymoglobulin can be discussed during multidisciplinary meetings and in expert centres. Patients with a predominant cholestasis, with minimal/no elevation in

Table 2. Comparison of characteristics between ICI-induced liver toxicity and autoimmune hepatitis.

	ICI-induced liver toxicity	Autoimmune hepatitis
Presentation	Heterogeneous	Heterogeneous
Gender prevalence	None	Female
Clinical symptoms	Non-specific Possibly asymptomatic	Non-specific
Biology		
AST/ALT elevation	Present	Present
GGT/ALP elevation	Present	Present at lower level than the cytolysis (look for PBC, PSC overlap)
Bilirubin elevation	Rare	Possible
Immunology		
Anti-nuclear antibodies	Possibly positive (about 50% of patients), speckled	Positive, high titre, homogeneous pattern
Anti-smooth muscles antibodies	Possibly positive (non-anti-F actin)	Positive, high titre, anti-F actin
Anti-LKM 1 antibodies	Negative	Positive (AIH type II)
IgG	Usually normal	Elevated
Histology		
Plasmocytes	Absent or rare	Frequent
Lobular inflammation	Present	Present
Portal tract inflammation	Present	Present
Confluent necrosis	Rare	Present
Granuloma	Often present in patients on anti-CTLA-4	Absent
Cholangitis	Present – cholangitis form	Rarely present (look for PBC, PSC overlap)
Chronic hepatitis/cirrhosis	Absent	Frequently present
CD4+/CD20+	Rare	Present
CD8+	Present	Rare
Therapy		
Corticosteroids	Not always needed	Needed
Long-term therapy	No	Yes
Corticosteroid discontinuation	Yes	Possible in selected patients
	Risk of recurrence: rare	Risk of recurrence: high

ALP, alkaline phosphatase; AIH, autoimmune hepatitis; ALT, alanine aminotransferase; Anti-LKM, anti-liver kidney microsome antibodies; AST, aspartate aminotransferase; CTLA-4, cytotoxic T lymphocyte-associated protein 4; GGT, gamma-glutamyltransferase; ICI, immune checkpoint inhibitor; PBC, primary biliary cholangitis; PSC, primary sclerosing cholangitis.

transaminases and without increased bilirubin, should be treated with UDCA alone as first-line treatment. Corticosteroids should be added if liver tests worsen or do not improve.

We believe that the management of ICI-induced liver toxicity should be guided by multidisciplinary discussions involving different specialists such as oncologists, hepatologists, immunologists, radiologists and pathologists.¹¹⁴

Impact of corticosteroids on the response to immunotherapy

It appears that using systemic corticosteroids or a second-line immunosuppressive drug did not significantly impact overall survival (OS) during several clinical trials.¹¹⁵ It has been reported quite recently that patients who experienced severe irAEs (grade ≥3) may even have an improved overall response rate and longer median time to progression compared to those without grade ≥3 irAEs, despite the use of corticosteroids.¹¹⁶ The development of irAEs was associated with a clinical benefit in patients with gastric cancer,¹¹⁷ non-small cell lung cancer¹¹⁸ and melanoma¹¹⁹ who were receiving nivolumab as monotherapy, but corticosteroids had no impact, although their use was not an end-point in these studies. Meanwhile, poor outcomes have been reported in patients receiving steroids (equivalent of ≥10 mg/day of prednisone) during nivolumab treatment for non-small cell lung cancer.¹²⁰ Studies are necessary to address this specific issue.

Reintroduction of immunotherapy after liver toxicity

According to ESMO guidelines, it is possible to re-introduce ICIs following immune-mediated hepatitis; their use is left at the physician’s discretion for grade 3 hepatitis but banned in grade 4 hepatitis. Several examples of re-challenge have been reported in the literature, with hepatitis recurrence rates ranging from 0 to

60%, differentially distributed as a function of the type of molecule used.^{66,67,70,73,110,121} However, when we pooled all the cases reported in the literature regarding a re-challenge with ICIs after liver toxicity, we found that among 58 retreated patients, 11 (19%) experienced a recurrence of liver toxicity. When we looked solely at patients who had experienced initial toxicity of grade ≥3 (a total of 29 patients) the recurrence rate rose to 40% (Table 3).

Three important questions regarding the resumption of immunotherapy remain: the type of ICI that can be administered, the interval elapsing between the resolution of hepatitis and the reintroduction of ICI, and the benefits of immunosuppressive prophylaxis.

Which ICI should be reintroduced is obviously critical. A re-challenge with the combination of anti-PD-1 and anti-CTLA-4 antibodies after hepatitis or another irAE carries a high risk of recurrent toxicity.¹²² The reintroduction of an anti-CTLA-4 antibody in a patient with previous immune-mediated hepatitis under anti-PD-1 treatment was seen to be associated with the development of fulminant hepatitis, although this thankfully improved with the use of plasmapheresis.⁸² However, re-challenge with an anti-PD-1 antibody in 4 patients out of 21 with previous liver toxicity under anti-CTLA-4 and anti-PD-1 combination therapy did not cause a recurrence of hepatitis.⁷⁴ Because the data on reintroduction are limited, some interesting findings can be discussed in the context of sequential therapies. Interestingly, grade ≥3 ALT and AST elevations were shown to be more frequent in patients who received nivolumab first and then ipilimumab, compared to patients who received ipilimumab followed by nivolumab.¹²³ Similarly, a more frequent onset of irAEs in patients treated with ipilimumab after nivolumab has been reported.¹²⁴ Serra-Bellver *et al.* hypothesised that

Table 3. Re-challenge with ICIs after resolved immune-mediated hepatitis.

Study	Patients retreated with ICI after liver toxicity	ICI first therapy	Grade of first liver toxicity ≥ 3	ICI re-challenge	Time between toxicity and reintroduction Days (median, range)	Liver toxicity recurrence	Other irAEs
Ziemer 2016	2	Anti-PD-1	2	Anti-PD-1	103 and 38	0	0
Spankuch 2017	1	Anti-CTLA-4 + anti-PD-1	1	Anti-PD-1	n.a.	0	0
Spain 2017	2	Anti-CTLA-4 + anti-PD-1	2	Anti-CTLA-4 + anti-PD-1	n.a.	1 (grade 4)	1
Pollack 2018	29	Anti-CTLA-4 + anti-PD-1	19	Anti-PD-1	58 (14-395)	5	n.a.
De Martin 2018	3	Anti-CTLA-4 + anti-PD-1	3	Anti-PD-1	n.a.	1	0
Gauci 2018	5	Anti-CTLA4 or Anti-PD-1	NA	Anti-CTLA-4 or Anti-PD-1	n.a.	0	0
Riveiro-Barciela 2019	1	Anti-PD-1	0	Anti-CTLA-4	n.a.	1 (grade 4)	0
Simonaggio 2019	5			Anti-PD-1 or Anti-PDL-1		3	0
Cheung 2019	4	Anti-CTLA-4 + anti-PD-1	2	Anti-PD-1	n.a.	0	0
Riveiro-Barciela 2020	6	Anti-PD-1 or Anti-PD-L1 or Anti-CTLA-4 (2)	0	Anti-PD-1	n.a.	0	0
Total	58		29 (50%)			11 (19%)	

CTLA-4, cytotoxic T lymphocyte-associated protein 4; ICI, immune checkpoint inhibitors; irAEs, immune-related adverse events; n.a., not available; PD-1, programmed cell death 1; PD-L1, programmed cell death 1 ligand 1.

this was caused by the prolonged activity of the anti-PD-1 drug during the introduction of the anti-CTLA-4 antibody.¹²⁵ Although the half-life of nivolumab is 17–25 days, PD-1 remains on circulating T cells at a mean plateau of 72% for at least 59 days, and PD-1 blockade persists even when serum concentrations of nivolumab are undetectable.¹²⁶ This means that because of the prolonged activity of anti-PD-1 antibodies, if a patient is subsequently receiving anti-CTLA-4 monotherapy, it is as if he or she was on combination therapy. According to the guidelines, a resumption of ICIs can be discussed when liver test values return to grade <1. The aim is to restart immunotherapy with a low risk of ICI-related toxicity. Data on the timing of reintroduction are scarce. The interval elapsing between the final dose of anti-CTLA4 + anti-PD-1 and the resumption of anti-PD-1 was slightly longer in patients without toxicities than in those with toxicities (median 62 vs. 56 days, $p = 0.03$).¹¹⁰ In a study that evaluated resuming the same ICI, the interval between anti-PD-1 or anti-PD-L1 administration and the first irAE was shorter among patients who experienced a recurrence than in those with no recurrence (9 vs. 15 days, $p = 0.04$).¹²¹ Although these observations do not specifically refer to the liver, this finding should be kept in mind when discussing the reintroduction of ICIs.

Whether prophylactic therapy can prevent a recurrence of hepatitis is a matter of debate. Surprisingly, in the series studied by Pollack *et al.*, patients who were on corticosteroids at the resumption of anti-PD-1, following the development of an irAE under anti-PD-1 and anti-CTLA-4 combination therapy, experienced a higher rate of toxicity than those who had discontinued corticosteroids (55% vs. 31%, $p = 0.03$).¹¹⁰

In 2 patients who experienced grade 3 hepatitis under nivolumab and responded to treatment with corticosteroids and UDCA, nivolumab was resumed successfully under budesonide and UDCA prophylaxis.¹²⁷ This finding needs to be confirmed.

Future development of immunotherapy: combination therapies

Treatments for solid tumours have dramatically improved in recent years through the use of ICIs. However, ICIs only benefit a subgroup of patients with an objective response rate (ORR) of 15% to 25%, or less in some refractory tumours such as pancreatic or prostate cancers. Furthermore, abnormal tumour vascularisation in the microenvironment exerts negative effects on the efficacy of anticancer therapies (including immunotherapies) via

different immunosuppressive molecular and cellular mechanisms. Therefore, strategies to induce vascular normalisation (*i.e.* combining ICIs and antiangiogenic agents) may enhance the benefits of immunotherapy and constitute the rationale for most ongoing trials (Table S6) (adapted from Jain *et al.*¹²⁸). Consequently, these benefits may be weighed against the added toxicity of ICIs and of antiangiogenic agents. Moreover, it is still unknown whether the combination of antiangiogenic agent-related toxicity and ICI-related irAEs may reflect a greater benefit in terms of efficacy than that reported for irAEs alone (*i.e.* increased OS and progression-free survival [PFS] for endocrinal and dermatological irAEs, for low grade irAEs and in case of PD-1 inhibitor use).¹²⁹ The toxicity of concurrent ICIs and antiangiogenic monoclonal antibodies (pembrolizumab/nivolumab and bevacizumab) seems to be relatively low, although the data are limited. In contrast, the incidence of severe treatment-related adverse events (trAEs) tended to rise in line with the dose of bevacizumab when combined with ipilimumab, while the incidence of some special interest irAEs (*i.e.* dermatological or gastrointestinal side effects) was not increased.

A meta-analysis that included 1,958 patients from 13 studies recently reported the efficacy and safety of combining ICIs and antiangiogenic agents in the treatment of advanced HCC.¹³⁰ The ORR, disease control rate (DCR) and PFS of combined treatment cohorts were significantly improved vs. those of anti-PD-1/PD-L1 cohorts (ORR $p = 0.016$; DCR $p < 0.001$; PFS $p < 0.001$). In addition, comparable incidence rates of trAEs were seen in the ICI monotherapy and combined treatment cohorts (74.4% vs. 84.3%) but with an increased incidence of grade ≥ 3 trAEs in the latter (18% vs. 32.7%; $p = 0.014$). Finally, patients with PD-L1-positive HCC had a significantly increased ORR when treated with nivolumab (odds ratio 2.32; 95% CI 1.43–3.77; $p < 0.05$).¹³⁰ More recently, in a global, open-label, phase III trial in patients with cirrhosis and unresectable HCC, atezolizumab combined with bevacizumab resulted in better OS and PFS outcomes than sorafenib.⁹ In this trial, the incidence of upper gastrointestinal bleeding observed in the combined treatment arm was 7% vs. 4.5% in the sorafenib arm. In addition, serious adverse events occurred more frequently with atezolizumab-bevacizumab (38%) than with sorafenib alone (30.8%). Concerning the liver, adverse events and more specifically grade ≥ 3 AST/ALT and bilirubin elevations were quite similar between treatment arms. Such an increase in trAEs has also

been observed during other trials using atezolizumab combined with bevacizumab.^{131,132} However, the addition of bevacizumab to atezolizumab did not significantly increase the incidence of irAEs and no unexpected patterns of toxicity were reported with this combination therapy.¹³³ The toxicity results regarding combination anti-PD-1 and TKI therapy were conflicting and some severe trAEs appeared to be largely linked to the TKI rather than being true acquired irAEs. Overall, the incidence of severe trAEs appeared to be slightly higher in ICI + TKI arms than in ICI + monoclonal antibody arms.

Conclusion and future

ICI-induced liver toxicity is a rare complication of cancer immunotherapy which is extremely heterogeneous in its presentation and severity. Although knowledge of this complication

is improving, several questions remain unanswered. Specific diagnostic tools are still required, the classification of toxicity grades must be rethought from a hepatological standpoint and management remains challenging, as it needs to balance the resolution of toxicity with the use of lifesaving treatment; the systematic use of corticosteroids seems to be a simplistic solution that is not always necessary. The use of ICI therapy, increasingly in combination with other therapies, is becoming more widespread. Liver toxicity is not a limiting factor for ICI use, however, it should be considered and better understood. Finding the minimum dose of ICIs with the best efficacy should be considered in the future design of trials to reduce the incidence and severity of hepatic irAEs. Further studies are required to elucidate the pathophysiological mechanisms and risk factors underlying toxicity, as well as to validate predictors of resolution and recurrence (in case immunotherapy is resumed).

Abbreviations

AIH, autoimmune hepatitis; ALT, alanine aminotransferase; ALP, alkaline phosphatase; AMA, anti-mitochondrial antibodies; ANA, anti-nuclear antibodies; anti-LKM, anti-liver-kidney microsomal antibodies; anti-LC1, anti-liver cytosol type-1 antibodies; ASMA, anti-smooth muscles antibodies; anti-SLA, anti-soluble liver antigen antibodies; AST, aspartate aminotransferase; CTLA-4, cytotoxic T lymphocyte-associated protein 4; DCR, disease control rate; DILI, drug-induced liver injury; GGT, gamma-glutamyltransferase; HCC, hepatocellular carcinoma; ICI, immune checkpoint inhibitor; INR, international normalised ratio; irAE, immune-related adverse event; MMF, mycophenolate mofetil; ORR, objective response rate; OS, overall survival; PD-1, programmed cell death 1; PD-L1-2, programmed cell death ligands 1-2; PFS, progression-free survival; TKI, tyrosine kinase inhibitor; trAE, treatment-related adverse event; UDCA, ursodeoxycholic acid; ULN, upper limit of normal.

Financial support

The authors received no financial support to produce this manuscript.

Conflict of interest

EDM: nothing to disclose; JMM: Principal/sub-Investigator of Clinical Trials for : Abbvie, Agios, Amgen, Argen-x, Astex, AstraZeneca, Daiichi Sankyo, Debiopharm, Eisai, Eos, Exelixis, Forma, Genentech, Janssen, Kyowa, Lilly, Lysarc, Lytix Biopharma, Medimmune, Roche, Sanofi, Xencor. PERSONAL FEES (Monies paid to you for services rendered, generally honoraria, royalties or fees for consulting, lectures, speakers bureaus, expert testimony, employment, ad-boards, etc.): Celgene, Bristol-Myers Squibb, AstraZeneca, Janssen. NON-FINANCIAL SUPPORT (Drugs, equipment supplied by the entity, travel paid by the entity, writing assistance, administrative support, etc.): AstraZeneca, Roche, Novartis, Gilead, Celgene, Bristol-Myers Squibb; OR: PERSONAL FEES (Monies paid to you for services rendered, generally honoraria, royalties or fees for consulting, lectures, speakers bureaus, expert testimony, employment, ad-boards, etc.) and Principal/sub-Investigator of Clinical Trials for: Cyrtext, Bayer, Eisai, Roche, BMS; CG: nothing to disclose; DS: nothing to disclose.

Please refer to the accompanying ICMJE disclosure forms for further details.

Authors' contributions

EDM: performed the systemic review of the literature, drafted and revised the manuscript; JMM: performed the systemic review of the literature and drafted the manuscript; OR: performed the systemic review of the literature, drafted and revised the manuscript; CG: provide histological images, revised the manuscript; DS: supervised, drafted and revised the manuscript.

Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.jhepr.2020.100170>.

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Author names in bold designate shared co-first authorship

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