# WILEY
## Online Proofing System Instructions

The Wiley Online Proofing System allows authors and proof reviewers to review PDF proofs, mark corrections, respond to queries, upload replacement figures, and submit these changes directly from the PDF proof from the locally saved file or while viewing it in your web browser.

**1.** For the best experience reviewing your proof in the Wiley Online Proofing System please ensure you are connected to the internet. This will allow the PDF proof to connect to the central Wiley Online Proofing System server. If you are connected to the Wiley Online Proofing System server you should see the icon with a green check mark above in the yellow banner.


Connected


Disconnected

**2.** Please review the article proof on the following pages and mark any corrections, changes, and query responses using the Annotation Tools outlined on the next 2 pages.


**3.** To save your proof corrections, click the "Publish Comments" button appearing above in the yellow banner. Publishing your comments saves your corrections to the Wiley Online Proofing System server. Corrections don't have to be marked in one sitting, you can publish corrections and log back in at a later time to add more before you click the "Complete Proof Review" button below.


**4.** If you need to supply additional or replacement files bigger than 5 Megabytes (MB) do not attach them directly to the PDF Proof, please click the "Upload Files" button to upload files:


Click Here

**5.** When your proof review is complete and you are ready to submit corrections to the publisher, please click the "Complete Proof Review" button below:


Click Here

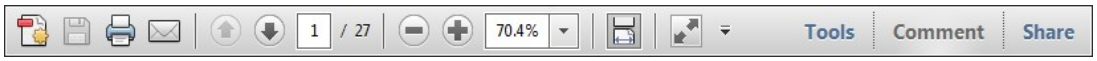
**IMPORTANT:** Do not click the "Complete Proof Review" button without replying to all author queries found on the last page of your proof. Incomplete proof reviews will cause a delay in publication.

**IMPORTANT: Once you click "Complete Proof Review" you will not be able to publish further corrections.**

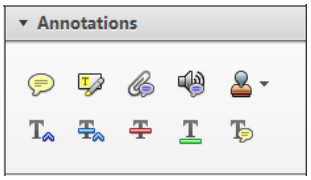
# USING e-ANNOTATION TOOLS FOR ELECTRONIC PROOF CORRECTION

WILEY

Once you have Acrobat Reader open on your computer, click on the Comment tab at the right of the toolbar:




This will open up a panel down the right side of the document. The majority of tools you will use for annotating your proof will be in the Annotations section, pictured opposite. We've picked out some of these tools below:

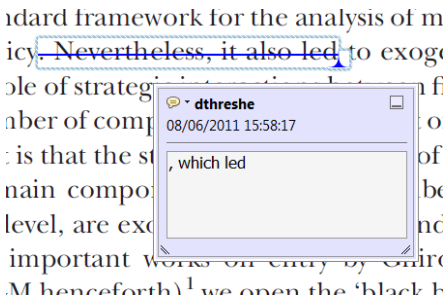



### 1. Replace (Ins) Tool – for replacing text.

Strikes a line through text and opens up a text box where replacement text can be entered.

**How to use it**

- Highlight a word or sentence.
- Click on the Replace (Ins) icon in the Annotations section.
- Type the replacement text into the blue box that appears.

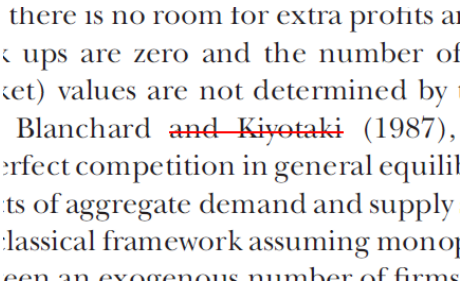



### 2. Strikethrough (Del) Tool – for deleting text.

Strikes a red line through text that is to be deleted.

**How to use it**

- Highlight a word or sentence.
- Click on the Strikethrough (Del) icon in the Annotations section.

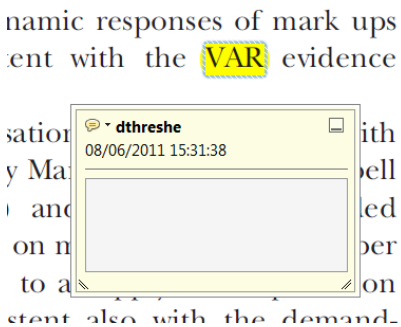


### 3. Add note to text Tool – for highlighting a section to be changed to bold or italic.

Highlights text in yellow and opens up a text box where comments can be entered.

**How to use it**

- Highlight the relevant section of text.
- Click on the Add note to text icon in the Annotations section.
- Type instruction on what should be changed regarding the text into the yellow box that appears.

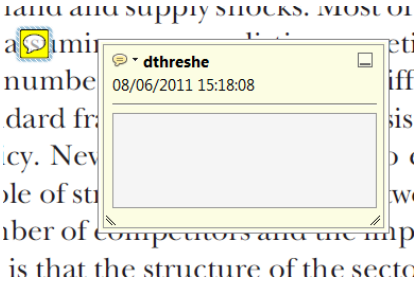



### 4. Add sticky note Tool – for making notes at specific points in the text.

Marks a point in the proof where a comment needs to be highlighted.

**How to use it**

- Click on the Add sticky note icon in the Annotations section.
- Click at the point in the proof where the comment should be inserted.
- Type the comment into the yellow box that appears.

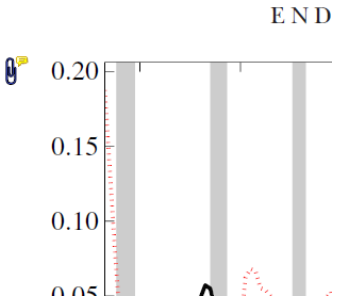


## 5. Attach File Tool – for inserting large amounts of text or replacement figures.

Inserts an icon linking to the attached file in the appropriate place in the text.

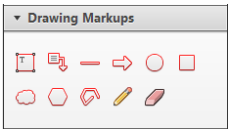
**How to use it**

- Click on the Attach File icon in the Annotations section.
- Click on the proof to where you'd like the attached file to be linked.
- Select the file to be attached from your computer or network.
- Select the colour and type of icon that will appear in the proof. Click OK.



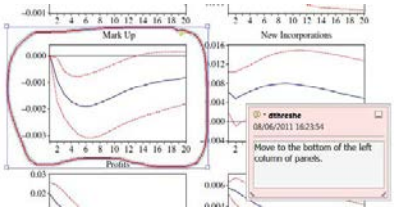

## 6. Drawing Markups Tools – for drawing shapes, lines and freeform annotations on proofs and commenting on these marks.

Allows shapes, lines and freeform annotations to be drawn on proofs and for comment to be made on these marks.




**How to use it**

- Click on one of the shapes in the Drawing Markups section.
- Click on the proof at the relevant point and draw the selected shape with the cursor.
- To add a comment to the drawn shape, move the cursor over the shape until an arrowhead appears.
- Double click on the shape and type any text in the red box that appears.



Received: 8 March 2017 | Accepted: 13 March 2017

DOI: 10.1111/liv.13421

DEBATES

WILEY Liver INTERNATIONAL

# Hepatocellular carcinoma and direct-acting antivirals: A never ending story?

Vincenza Calvaruso | Antonio Craxì

Sezione di Gastroenterologia & Epatologia, DIBIMIS, Università di Palermo, Palermo, Italy

**Correspondence**
Vincenza Calvaruso, MD, PhD, Gastroenterologia & Epatologia, DIBIMIS, University of Palermo, Palermo, Italy.
Email: vincenza.calvaruso@unipa.it

Handling Editor: Mario Mondelli

## Abstract

Xxxxx

**KEYWORDS**

direct antiviral agents, hepatitis C virus, hepatocellular carcinoma

A large body of evidence accumulated over the two decades of the "interferon era" shows conclusively that HCV eradication (sustained virological response, SVR) in patients with cirrhosis reduces both liver and non-liver-related deaths.[1,2] When cirrhosis is subclassified according to the stage of portal hypertension, it is apparent that the benefit of SVR is higher in patients without clinically significant portal hypertension.[3] In all cohorts studied the risk of developing hepatocellular carcinoma (HCC) for patients with IFN-induced SVR, albeit reduced in comparison to those with persistent HCV infection, is not cancelled altogether.[4,5] Hence, continuing HCC surveillance is recommended in patients with HCV cirrhosis after SVR.[5]

The introduction of direct-acting antivirals (DAAs) made possible to eradicate HCV effectively at all stages of liver disease and has led to the rapid accumulation of large cohorts of patients with cirrhosis in whom treatment has been initiated regardless of the stage of the disease, including decompensated cirrhosis, in order to obtain an improvement of liver function and thus ameliorate the short and long-term outcome.

This "all-comers" approach has included patients with HCC whose tumour had been successfully treated. Also in this setting, DAAs obtain exceedingly high SVR rates thus likely prolonging the life span of these patients and creating a further period of time for HCC recurrence. After the first report by Reig et al.,[6] who raised a first warning about an unexpected high rate of recurrence of HCC in these patients, various groups have reported their experience on the recurrence and/or occurrence of HCC in cirrhotic patients treated with DAAs, mostly with negative results. The "Debates" section of this issue of Liver International hosts a thoughtful review by Alberti and Piovesan focusing on this highly controversial issue.[7]

When discussing recurrence, we should bear in mind that the estimated likelihood of HCC recurrence after its presumed cure in patients with HCV cirrhosis untreated with antivirals approximates 10% at 6 months, 20% at 12 months and 50% after 24 months since HCC treatment.[8] Hence, against such a high background of reappearance of cancer in viraemic patients, a healthy dose of caution should be exercised before concluding that DAA-induced SVR is associated with an unexpected rate of HCC recurrence. An extensive body of data not confirming the Spanish hypothesis comes from the pooling of DAA-treated patients who underwent curative HCC therapies enrolled in three French prospective multicenter ANRS cohorts. These data, encompassing over 500 subjects with sufficient follow-up, do not show an increased risk of HCC recurrence after DAA-induced SVR and report instead a comparable rate of reappearance of cancer among DAA-treated and untreated patients.[9] In support of the French findings, data from the large cohort from the UK show a reduction in HCC rates in DAA-treated patients with advenced cirrhosis.[10] Last but not least, when the interval between complete tumour eradication and antiviral therapy is quite long as in the cohort reported by Zavaglia et al.,[11] the recurrence rate is actually low suggesting that the longer the interval, the lower the risk that residual cancer is still present at the start of DAA therapy.

We recently evaluated in the setting of a regional database (RESIST-HCV), 185 cirrhotic patients (84% Child A) with complete response after curative treatment of HCC treated with DAAs.[12] Over a mean follow-up of 24 weeks (range 8-60) since starting treatment, 24

**Abbreviations:** DAAs, direct-acting antivirals; HCC, hepatocellular carcinoma; HCV, Hepatitis C virus; IFN, interferon; RBV, ribavirin; SVR, sustained virological response.

CON commentary for "Increased incidence of liver cancer after successful DAA treatment of chronic hepatitis C: fact or fiction?" *Liver Int.* 2017;XX:XXX-XXX.



Journal Name LIV
Manuscript No. 13421
WILEY
No. of pages: 3 Dispatch: 30-3-2017
PE: Rathi T.S. CE: Lenard S

patients had a recurrence of HCC with a crude rate of 13%. The 6 and 12 months. HCC recurrence rates were 7.9% and 16.3% respectively. One patient died during follow-up. The pattern of HCC recurrence was nodular in 83% patients (20/24) and infiltrative in 17% (4/24).

Many concerns about this side of the controversy can be linked to:

- High heterogeneity of the groups of patients with HCC in terms of clinical features (stage of cirrhosis; morphology of HCC)
- Different treatments, from palliative (transarterial chemoembolisation) to potentially curative (ablation and surgery)
- Time elapsed between presumed cure of HCC and treatment with DAAs.

In fact, some cases of cancer relapse within a few days or weeks after starting DAAs, which have been counted as HCC recurrence, are most likely instances of incompletetly cured HCC already present at the time of initiation of DAA use, not detected by imaging.

Moving to occurrence, ie, de novo HCC in patients with cirrhosis treated with DAAs, it must be stressed that extrapolation of data from patients treated with interferon-based regimens to those who received the all-oral regimens is inappropriate. In fact, the strong selection effect due to the low tolerability of IFN has restricted antiviral therapy, until the availability of DAAs, to patients with lesser stages of cirrhosis, ie, to those who are intrinsically less likely to develop HCC.[3,13] By the way, Kozbial et al.[14] in a small series of patients with cirrhosis observed a 6.6% overall (13/195) and 5.2% (10/192 in patients with SVR) of rate of HCC after DAAs and compared them with the estimated 1% per year frequency of HCC in patients with SVR treated with IFN/RBV.[3,15] As expected patients treated with IFN were younger and all had compensated cirrhosis, while those on DAAs could also have decompensated cirrhosis or signIficant comorbidity. As a further confirmation, when the incidence of HCC during and after DAAs is assessed after stratifyng for stage of cirrhosis, patients with Child A cirrhosis develop HCC at a ratecomparable to historical cohorts of patients treated with IFN based therapies. Indeed, two recently reported large Italian prospective cohorts[16,17] demonstrate that the rate of occurrence of HCC in patients with Child A HCV cirrhosis and no or mild portal hypertension does not exceed 2% at 1 year of follow, while patients with Child B HCV cirrhosis have a significantly higher rate of occurrence of HCC (more than 3% despite SVR). For the latter, there is no historical benchmark of comparison, given the impossibility to treat the patients with IFN.

Further evidence supporting a positive effect of DAAs on the overall reduction in HCC comes from these cohorts[16,17] by the finding of a higher incidence of cancer, and of all liver-related disease events, in patients who fail to obtain SVR on DAAs as compared to those with SVR.

Whether the morphological pattern of expression and the clinical behaviour of HCC in eradicated patients will be more aggressive than its usual course, as suggested by the NAVIGATORE experience,[16] will need careful confirmation by prospective studies with careful characterisation of the growth pattern and molecular characteristics of the tumour.

In summary, while cirrhotic patients treated with DAAs seem, in our opinion, to have a reduction in the overall risk of HCC, there might be a subset of subjects in whom the imbalance induced by SVR in the inflammatory response and in the tumour microenvironment could originate and/or favour the growth of HCCs with an "aggressive" clinical phenotype. Hence, the challenge for the future is to identify features which allow the profiling of patients to evaluate the risk of HCC in the individual patient.

## CONFLICT OF INTEREST

Vincenza Calvaruso: Travel Grant, Speaking, and Participation to Advisory Boards for: AbbVie, BMS, Gilead Sciences and Intercept. Grant and research support: MSD. Antonio Craxi: Research grants, lecturing fees, advisory boards, scientific consultancy for Novartis, Abbvie, Gilead Sciences, BMS, Achillion, Janssen Cilag, Abbott Diagnostics, Intercept.

## REFERENCES


1. Nahon P, Bourcier V, Layese R, et al. Eradication of hepatitis C virus infection in patients with cirrhosis reduces risk of liver and non-liver complications. *Gastroenterology*. 2017;152:142-156.
2. Tada T, Kumada T, Toyoda H, et al. Viral eradication reduces all-cause mortality in patients with chronic hepatitis C virus infection: a propensity score analysis. *Liver Int*. 2016;36:817-826.
3. Di Marco V, Calvaruso V, Ferraro D, et al. Effects of eradicating hepatitis C virus infection in patients with cirrhosis differ with stage of portal hypertension. *Gastroenterology*. 2016;151:130-139.
4. Bruno S, Di Marco V, Iavarone M, et al. Survival of patients with HCV cirrhosis and sustained virologic response is similar to the general population. *J Hepatol*. 2016;64:1217-1223.
5. Morgan RL, Baack B, Smith BD, et al. Eradication of hepatitis C virus infection and the development of hepatocellular carcinoma: a meta-analysis of observational studies. *Ann Intern Med*. 2013;158:329-337.
6. Reig M, Marino Z, Perello C, et al. Unexpected high rate of early tumor recurrence in patients with HCV-related HCC undergoing interferon-free therapy. *J Hepatol*. 2016;65:719-726.
7. Alberti A, Piovesan S. Increased incidence of liver cancer after successful DAA treatment of chronic hepatitis C: fact or fiction? *Liver Int*. 2017;???:???-???. 10
8. Cabibbo G, Petta S, Barbàra M, et al. A meta-analysis of single HCV-untreated arm of studies evaluating outcomes after curative treatments of HCV-related hepatocellular carcinoma. *Liver Int*. 2017;???:???-???. 11
9. ANRS Collaborative Study Group on Hepatocellular Carcinoma (ANRS CO22 HEPATHER, CO12 CirVir and CO23 CUPILT cohorts). Lack of evidence of an effect of direct-acting antivirals on the recurrence of hepatocellular carcinoma: data from three ANRS cohorts. *J Hepatol*. 2016;65:734-740.
10. Cheung MC, Walker AJ, Hudson BE, et al. Outcomes after successful direct-acting antiviral therapy for patients with chronic hepatitis C and decompensated cirrhosis. *J Hepatol*. 2016;65:741-747.
11. Zavaglia C, Okolicsanyi S, Cesarini L, et al. Is the risk of neoplastic recurrence increased after prescribing direct-acting antivirals for HCV patients whose HCC was previously cured? *J Hepatol*. 2017;66:236-237.
12. Cabibbo G, Cacciola I, Cannavò C, et al. Risk of hepatocellular carcinoma (HCC) recurrence in HCV cirrhotic patients treated with direct acting antivirals (DAAs). *Dig Liver Dis*. 2017;49:e59.
13. Fattovich G, Stroffolini T, Zagni I, Donato F. Hepatocellular carcinoma in cirrhosis: incidence and risk factors. *Gastroenterology*. 2004;127:S35-S50.
14. Kozbial K, Moser S, Schwarzer R, et al. Unexpected high incidence of hepatocellular carcinoma in cirrhotic patients with sustained virologic

response following interferon-free direct-acting antiviral treatment. *J Hepatol*. 2016;65:856-858.
15. van der Meer AJ, Wedemeyer H, Feld JJ, et al. Life expectancy in patients with chronic HCV infection and cirrhosis compared with a general population. *JAMA*. 2014;312:1927-1928.
16. Romano A, Capra F, Piovesan S, et al. Incidence and pattern of "de novo" hepatocellular carcinoma in HCV patients treated with oral DAAs. *Hepatology*. 2016;64S1:10A.
17. Calvaruso V, Cabibbo G, Cacciola I, et al. Early occurrence of hepatocellular carcinoma (HCC) in patients with HCV cirrhosis treated with direct-acting antivirals (DAAs). EASL HCC Summit 2017. Geneva, Switzerland. 2017. *J Hepatol*. 2017;???:???-???.

12

**How to cite this article:** Calvaruso V, Craxì A. Hepatocellular carcinoma and direct-acting antivirals: A never ending story? *Liver Int*. 2017;00:1–3. https://doi.org/10.1111/liv.13421

# Author Query Form

Journal: LIV

Article: 13421

Dear Author,

During the copy-editing of your paper, the following queries arose. Please respond to these by marking up your proofs with the necessary changes/additions. Please write your answers on the query sheet if there is insufficient space on the page proofs. Please write clearly and follow the conventions shown on the attached corrections sheet. If returning the proof by fax do not write too close to the paper's edge. Please remember that illegible mark-ups may delay publication.

Many thanks for your assistance.

| Query reference | Query | Remarks |
|---|---|---|
| 1 | **AUTHOR: Please confirm that given names (red) and surnames/family names (green) have been identified correctly.** |  |
| 2 | **AUTHOR: Please check that author and their affiliation is correct.** |  |
| 3 | **AUTHOR: Please check edit made in the article title.** |  |
| 4 | **AUTHOR: Please supply an unstructured abstract for this paper.** |  |
| 5 | **AUTHOR: The set of keywords is extracted from the supplied PDF source file. Please check if the keywords are okay.** |  |
| 6 | **AUTHOR: Please check that the "article note" has been presented correctly.** |  |
| 7 | **WILEY: Please updeate the volume and page number inplace of "XXX".** | |
| 8 | **AUTHOR: Please provide "key points" section for the article.** |  |
| 9 | **AUTHOR: Please check the text under the heading "Conflict of Interest" section.** |  |
| 10 | **WILEY: Please update the volume number, page range for reference [7].** | |
| 11 | **WILEY: Please update the volume number, page range for reference [8].** | |
| 12 | **AUTHOR: Please provide the volume number, page range for reference [17].** |  |