THU-333

Digital pathology using stain-free imaging indices as a tool for fibrosis quantification in patients with congestive hepatopathy

Matthew Yeh[1], Hong-Wen Tsai[2], Che-Wei Hsu[2], Cheng-Yi Chen[2], Yayun Ren[3], Kutbuddin Akbary[3], Elaine Chng[3], Dean Tai[3]

[1]University of Washington, Department of Pathology, United States, *[2]National Cheng Kung University Hospital, Taiwan*, *[3]HistoIndex Pte Ltd, Singapore*

Email: yehmliver@gmail.com

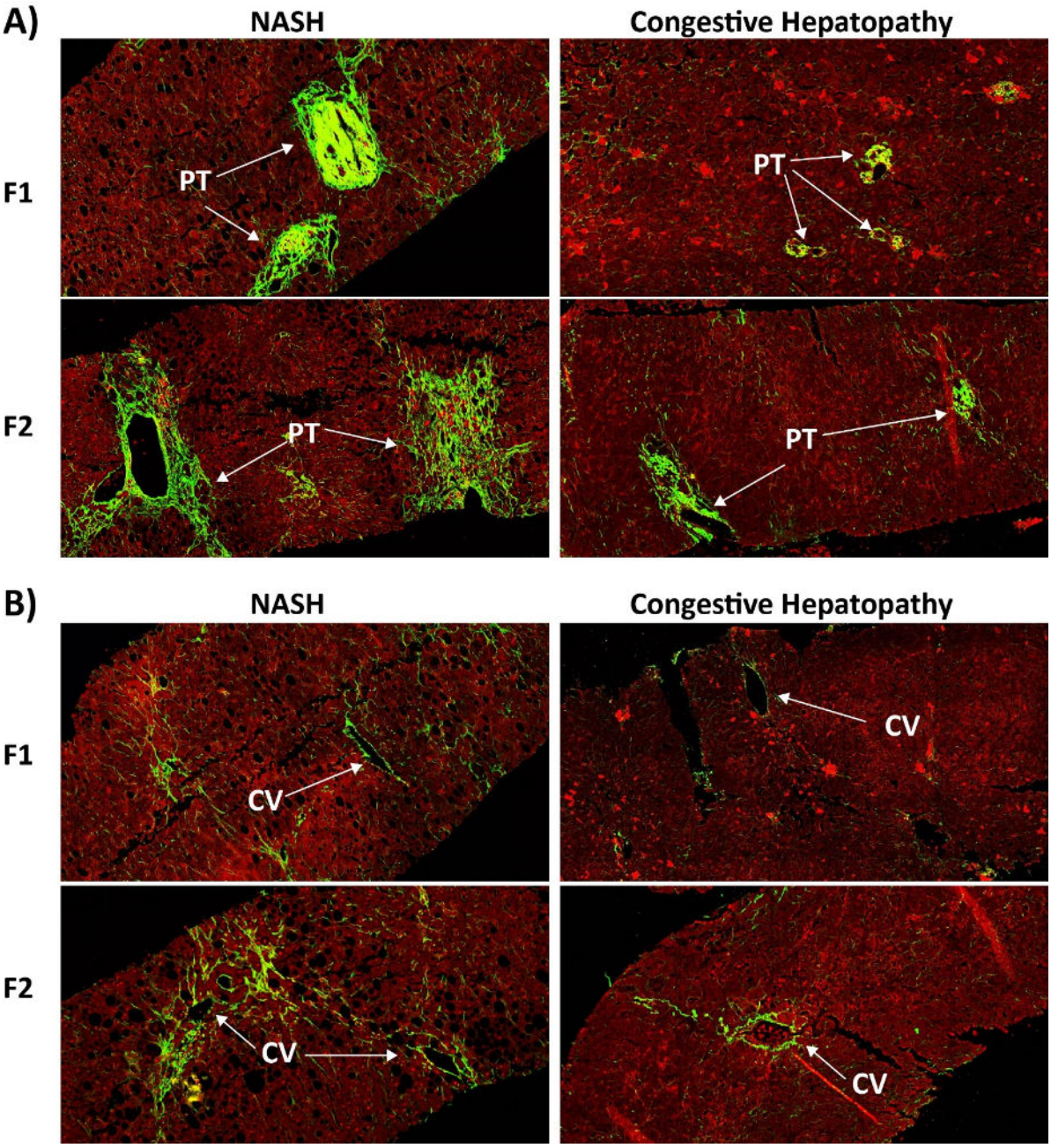
Background and Aims: Congestive hepatopathy (CH) is the result of right heart failure from myriads of heart diseases. It causes centrilobular fibrosis and can progress to portal and bridging fibrosis and even cirrhosis. While histological scoring system for fibrosis in congestive hepatopathy exists, the scores are categorical but not continuous. There are also intra- and inter-observer variation among pathologists. Second harmonic generation/two-photon excitation fluorescence (SHG/TPEF) microscopy has been demonstrated to provide accurate and reproducible fibrosis quantification in preclinical and clinical liver specimens, including biopsies from patents with viral hepatitis and NAFLD. We aim to test the feasibility using SHG/TPEF microscopy in assessing liver fibrosis in congestive hepatopathy.

Method: Unstained sections from 10 congestive hepatopathy cases with Dai scheme stages 0, 1, 2 and 3 were imaged using SHG/TPEF microscopy. Changes in overall liver fibrosis and in five zonal regions of liver lobules were quantitatively assessed by qFibrosis – a cumulative index based on measuring 184 collagen features on a continuous scale. Using sequential feature selection, 3 parameters were chosen out of 184 fibrosis parameters and a linear regression method was used to construct a congestive hepatopathy index.

Results: 3 parameters chosen for CH fibrosis are all from central vein regions, whereas in NASH, 15 parameters chosen were from total tissue area and portal tract regions. This finding indicates the fibrosis progression patterns is quantitatively different in portal tract and central vein regions. This difference can be better visualized in the figures, where SHG/TPEF images shows more fibrosis in NASH comparing to CH. Note that the staging systems used are NASH-CRN and Dai for NASH and CH patients respectively.

Conclusion: This is the first series liver fibrosis in congestive hepatopathy is shown to be assessed using SHG/TPEF microscopy. These data will be expanded and validated in an additional larger number of congestive hepatopathy cohort correlating with clinical and outcome data, and compared with other liver diseases (e.g., NASH and viral hepatitis), which may provide insight evaluating disease severity that aid clinical management plan algorithm and decision-making.

Figure: Image examples of (A) portal fibrosis and (B) central fibrosis for NASH and CH patients


A)
NASH
Congestive Hepatopathy
F1
F2
PT
PT
PT
PT
B)
NASH
Congestive Hepatopathy
F1
F2
CV
CV
CV
CV

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[1]University of Washington, Department of Laboratory Medicine and Pathology, Seattle, WA, USA
[2]National Cheng-Kung University, Department of Pathology, Tainan, Taiwan
[3]Histoindex Pte. Ltd., Singapore

INTRODUCTION

- Congestive hepatopathy (CH) is the result of right heart failure from a myriad of heart diseases. It causes centrilobular fibrosis and can progress to portal and bridging fibrosis, and even cirrhosis.
- While a histological scoring system for fibrosis in congestive hepatopathy exists, the scores are categorical and not continuous.
- There is also intra- and inter-observer variation among pathologists.[1]
- Second harmonic generation/two-photon excitation fluorescence (SHG/TPEF) microscopy has been demonstrated to provide accurate and reproducible fibrosis quantification in preclinical and clinical liver specimens, including biopsies from patents with viral hepatitis and NAFLD.

AIM

- The aim of this exploratory study was to test the feasibility of using SHG/TPEF microscopy in assessing liver fibrosis in congestive hepatopathy.

METHOD

- Unstained sections from 10 congestive hepatopathy cases with Dai scheme[2,3] stages 0, 1, 2 and 3 were imaged using SHG/TPEF microscopy.
- Changes in overall liver fibrosis and in five zonal regions of liver lobules were quantitatively assessed by qFibrosis – a cumulative index based on measuring collagen features on a continuous scale.
- Using sequential feature selection, 3 parameters were chosen out of 184 fibrosis parameters and a linear regression method was used to construct a congestive hepatopathy index.

RESULTS

Figure 1. Masson's trichrome stained liver sections showing congestive hepatopathy fibrosis. (From left to right): Dai stages 0 to 3.

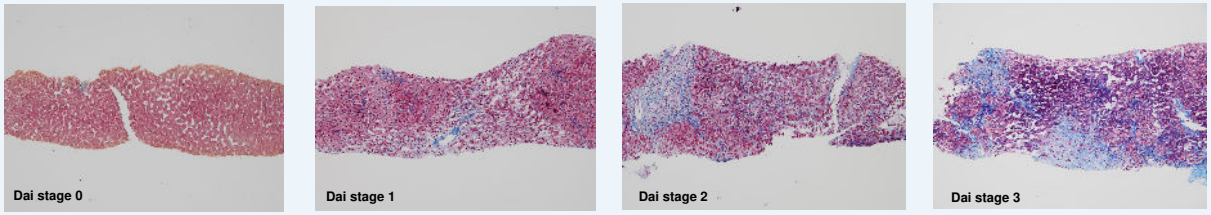
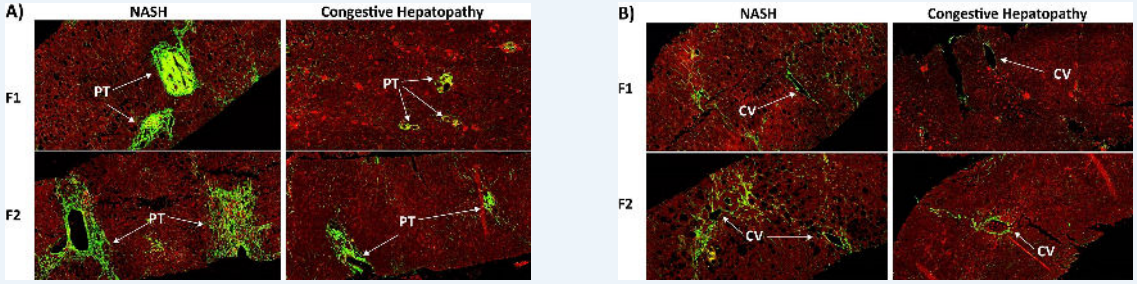


Figure 2. SHG/TPEF images of (A) portal fibrosis patterns and (B) central vein fibrosis patterns in F1 and F2 untreated NASH and CH patients. Note that the staging systems used are NASH-CRN and Dai for NASH and CH patients, respectively.

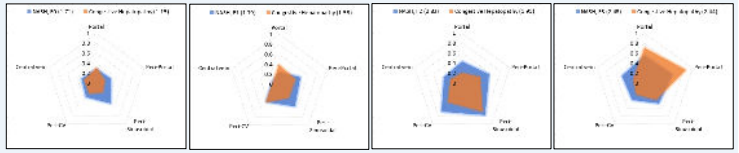


- SHG/TPEF detects fibrosis pattern of congestive hepatopathy observed using Masson trichrome as depicted in Figure 1. The collagen fibers in congestive hepatopathy and NASH distribute variously in central and portal regions using SHG/TPEF, as shown in figures 2, which reflect disease progression in these diseases.
- The 3 parameters chosen using sequential feature selection method to construct a congestive hepatopathy fibrosis index are central vein-related parameters. This contrasts with NASH fibrosis index, whose parameters consist of total tissue area including portal tract-related parameters. This difference may be attributed to uneven distribution of samples for each stage of the small CH cohort.
- This finding indicates the fibrosis is quantitatively different in portal tract and central vein regions. This difference can be better visualized in Figure 2 above, where SHG/TPEF images show more fibrosis in NASH compared to CH. Again, this result may be attributed to the small number of CH cases in this exploratory analysis.

Table 1. Quantitative fibrosis measurements in each region for NASH and CH patients grouped according to NASH-CRN and Dai staging, respectively.

	NASH F0 vs Dai F0		NASH F1 vs Dai F1		NASH F2 vs Dai F2B		NASH F3 vs Dai F3	
	NASH (n=37)	CH (n=2)	NASH (n=29)	CH (n=3)	NASH (n=7)	CH (n=4)	NASH (n=12)	CH (n=1)
Portal fibrosis	0.282	0.301	0.234	0.384	0.425	0.196	0.548	0.704
Peri-portal fibrosis	0.301	0.166	0.438	0.326	0.561	0.365	0.569	0.857
Peri-sinusoidal fibrosis	0.491	0.210	0.538	0.322	0.780	0.683	0.481	0.394
Peri-central fibrosis	0.335	0.233	0.430	0.428	0.688	0.456	0.406	0.261
Central vein	0.302	0.240	0.145	0.118	0.376	0.234	0.472	0.229
Total Weighted Score	**1.711**	**1.150**	**1.786**	**1.579**	**2.830**	**1.933**	**2.476**	**2.444**

Figure 3. Radar charts of quantitative fibrosis measurements in each region for NASH and CH patients grouped according to NASH-CRN and Dai staging, respectively. The data is based on Table 1 as shown on the left.



- When fibrosis was assessed on a continuous scale, the data suggested that the total weighted score for CH patients were lower than that of NASH, but with no statistical significance ($p>0.05$).

CONCLUSIONS

- This is the first series of applying SHG/TPEF microscopy to evaluate liver fibrosis in congestive hepatopathy.
- The total weighted fibrosis score was lower for CH patients than that of NASH patients, even though there were no significant difference between these two groups at the parameter level.
- These data will be expanded and validated in an additional larger congestive hepatopathy cohort correlating with clinical and outcome data; as well as comparing with other liver diseases (e.g., NASH and viral hepatitis).
- We postulate that this may provide additional insights into evaluating disease severity which can aid in patients' clinical management and clinicians' decision-making.

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REFERENCES

1. Soon GST, Liu F, Leow WQ, Wee A, Wei L, Sanyal AJ. Clin Gastroenterol Hepatol. 2022 Jun 10:S1542-3565(22)00555-9.

2. Dai DF, Swanson PE, Krieger EV, Liou IW, Carithers RL, Yeh MM. Mod Pathol. 2014 Dec;27(12):1552-8.

3. Bosch DE, Koro K, Richards E, Hoch BL, Jalikis F, Koch LK, Swanson PE, Truong CD, Liou I, Yu L, Bhattacharya R, Yeh MM. Am J Surg Pathol. 2019 Jun;43(6):766-772.

CONTACT INFORMATION

yehmliver@gmail.com