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Heterogeneity of Hepatic Steatosis Definitions and Reporting of Donor Liver Frozen Sections Among Pathologists: A Multi-Center Survey

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Abbreviations:

LD-MAS (large-droplet macrovesicular steatosis), MAS (macrovesicular steatosis), MIS (microvesicular steatosis), SD-MAS (small-droplet macrovesicular steatosis)

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TO THE EDITOR:

The demand for donor livers continues to increase. Strategies to increase the donor liver pool include increasing donor registration, using living donors, and using “extended criteria” for donor livers. One such extended criterion is using livers with hepatic steatosis. With the high prevalence of obesity, the number of potential donors with liver steatosis will increase. The assessment of donor liver steatosis is typically made by intraoperative consultation, including frozen section. Most studies analyzing the accuracy of intraoperative steatosis determination have shown promising results (2, 3, 4, 5).

Current practice generally outright excludes donor livers with severe steatosis (usually defined as >60%) and very judiciously allows for the use of livers with moderate steatosis (usually defined as 30%-60%). Several studies around the world have examined the association of hepatic steatosis in donor livers with post-transplant outcomes (1). Heterogeneity of pathologic evaluation and reporting may contribute to the varying findings in these studies. For example, there are different definitions for steatosis, steatosis types (macrovesicular steatosis [MAS] and microvesicular steatosis [MIS]), and steatosis subtypes (large-droplet macrovesicular steatosis [LD-MAS] and small-droplet macrovesicular steatosis [SD-MAS]). Furthermore, there are variations in biopsy methods, stains used, the approach to assessing steatosis, and how results are reported to the surgeon. Various studies have established different cutoff values for categorizing steatosis.

This study aimed to assess different definitions for hepatic steatosis and reporting patterns in the frozen section setting. In September of 2020, we developed a 27-question questionnaire regarding evaluation of graft hepatic steatosis by intraoperative frozen section and sent it via email to pathologists in 25 academic pathology departments across the United States. Anonymized results were received from 28 pathologists. All respondents practice in an academic setting with 27 of 28 having received subspecialty training in liver pathology and 27 of 28 interpreting donor liver frozen sections.

There was widespread variation in respondents' definitions of steatosis types and subtypes. Most notably, the definitions of SD-MAS and MIS showed weak agreement between respondents. When asked to define SD-MAS, a plurality (50%) of respondents considered SD-MAS to be fat vacuoles larger than the nucleus but smaller than half of the cell with a central nucleus. When asked to define MIS, a majority (64%) percent of responders defined MIS as the accumulation of tiny lipid vesicles in the cytoplasm of hepatocytes with a central nucleus.

This variation in definitions was also seen when respondents were asked to interpret the types of steatosis in four example photomicrographs (**Figure 1**). 100% considered the steatosis in **Figure 1A** to be LD-MAS. 89% considered the steatosis in **Figure 1B** to be SD-MAS, 3.6% said MIS, 3.6% said foamy degeneration, and 3.6% said small droplet steatosis. 50% considered the steatosis in **Figure 1C** to be SD-MAS, 46% said MIS, and 3.6% said MIS with foamy degeneration. 50% considered the steatosis in **Figure**

1D to be foamy degeneration, 36% said MIS, 11% said foamy degeneration/MIS, and 3.6% said MIS (while noting that Oil Red O staining would be required for confirmation).

There was also variation in how graft steatosis is determined and reported at the time of surgery. Specimen types were mixed: wedge (21%), needle (25%), both wedge and needle (54%) biopsies. Responders varied in their use of low-medium power vs. high-medium power. They also varied on whether they calculated the percentage of parenchyma occupied by fat (46%) vs. the number of hepatocytes involved by steatosis (54%). The values reported to the surgeon varied as well. 11% report only LD-MAS (semi-quantitatively); 11% report only LD-MAS (exact percentage); 14% report total MAS (semi-quantitatively); 28% report total MAS (exact percentage); 18% report total MAS, LD-MAS, and SD-MAS (exact percentages); and 18% report LD-MAS and SD-MAS separately (exact percentages). The steatosis cutoff values used by surgical colleagues varied: 64% had different cutoffs depending on the circumstances, 25% had a strict cutoff of 30%, 3.6% had a strict cutoff of 50%, and 7.2% had a strict cutoff of 60%. Finally, 86% of responders expressed that they encountered challenges in evaluating donor livers for steatosis - including freezing artifact (64%), the lack of a uniform definition for MAS (18%), and a lack of education/training on this topic (3.6%).

In summary, this survey-based study demonstrates significant variation in how subspecialty-trained liver pathologists define, assess, calculate, and report donor liver steatosis during intraoperative consultation. These findings call for unified definitions of steatosis types and subtypes and consistent methods for determining and reporting the presence of steatosis in graft livers. This consistency in practice is essential at the time of clinical decision-making and for research purposes. This study has a major impact in graft liver pathology practice considering many community pathologists reading these frozen sections (often in the middle of the night) may not have subspecialty training and may have greater heterogeneity in steatosis interpretation and reporting. Hopefully, the introduction of Banff consensus recommendations for the determination and reporting of “large droplet fat” in a recent publication (6) will reduce the heterogeneity in interpreting and reporting steatosis in donor liver frozen sections to help achieve optimal utilization of steatotic donor livers without compromising post-transplant outcomes.

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REFERENCES

1. Chu MJ, Dare AJ, Phillips AR, Bartlett AS. Donor Hepatic Steatosis and Outcome After Liver Transplantation: a Systematic Review. J Gastrointest Surg 2015;19:1713-24.

2. Lo IJ, Lefkowitch JH, Feirt N, Alkofer B, Kin C, Samstein B, Guarrera JV, Renz JF. Utility of liver allograft biopsy obtained at procurement. Liver Transpl 2008;14:639-46.

3. Fiorentino M, Vasuri F, Ravaioli M, Ridolfi L, Grigioni WF, Pinna AD, D'Errico-Grigioni A. Predictive value of frozen-section analysis in the histological assessment of steatosis before liver transplantation. Liver Transpl 2009;15:1821-5.

4. Holowko W, Mazurkiewicz M, Grat M, Koperski L, Lewandowski Z, Smoter P, et al. Reliability of frozen section in the assessment of allograft steatosis in liver transplantation. Transplant Proc 2014;46:2755-2757.

5. Flechtenmacher C, Schirmacher P, Schemmer P. Donor liver histology--a valuable tool in graft selection. Langenbecks Arch Surg. 2015;400:551-7.

6. Neil DAH, Minervini M, Smith ML, Hubscher SG, Brunt EM, Demetris AJ. Banff consensus recommendations for steatosis assessment in donor livers. Hepatology 2021;00:1-12.

Figure Legend

Figure 1. Four photomicrographs of hepatic steatosis used in the survey. A. A single fat vacuole pushing the nucleus to the edge of the hepatocyte, which all respondents interpreted as LD-MAS. B. A fat vacuole with a size similar to the centrally-located nucleus, which was interpreted as SD-MAS by 89% of respondents. C. A few fat vacuoles in a hepatocyte with a centrally located nucleus, which was interpreted as SD-MAS by 50% of respondents and as MIS by 46%. D. Numerous tiny vesicles in the hepatocyte, which were interpreted as foamy degeneration by 50% of respondents, as MIS by 36%, and as foamy degeneration/MIS by 11%.

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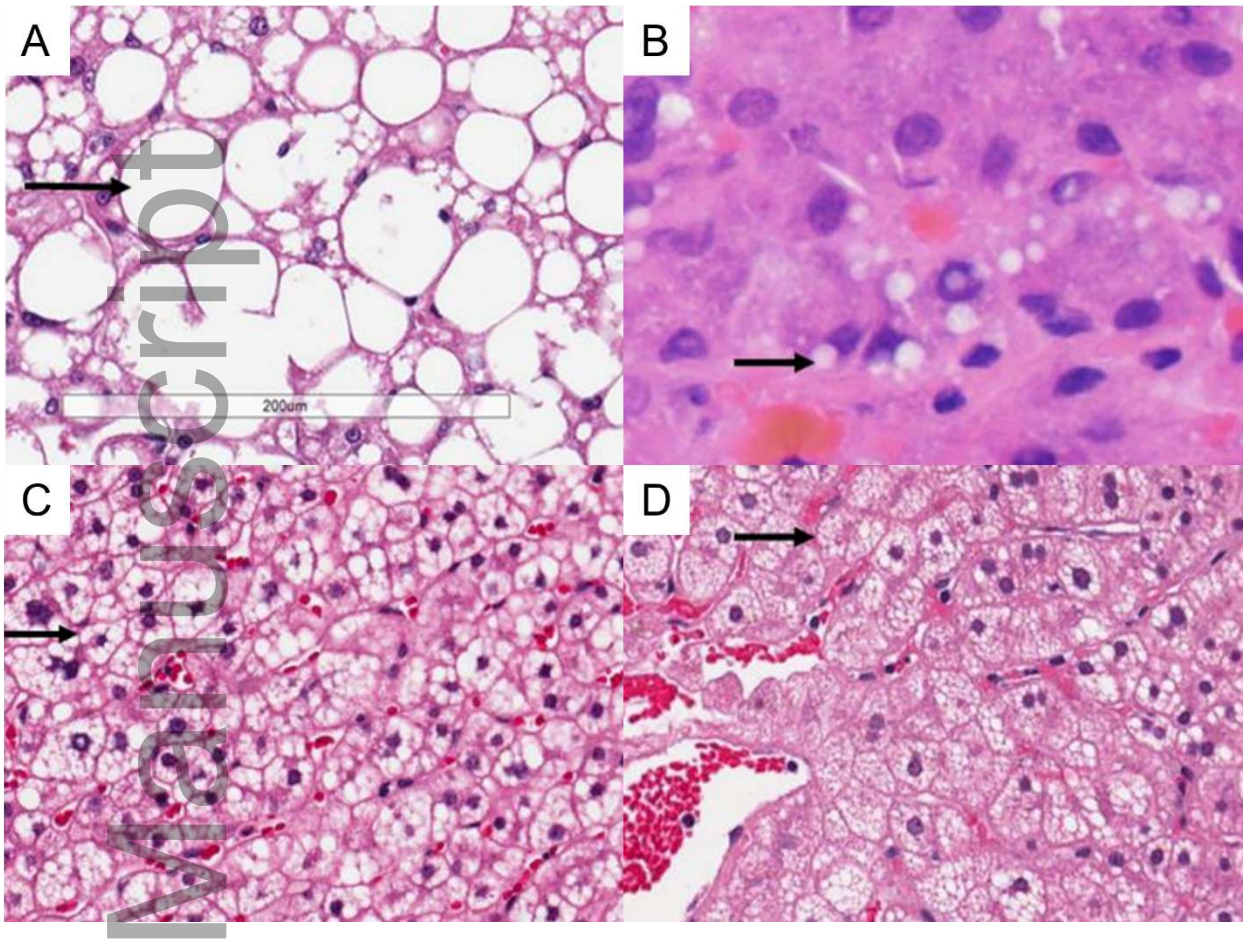

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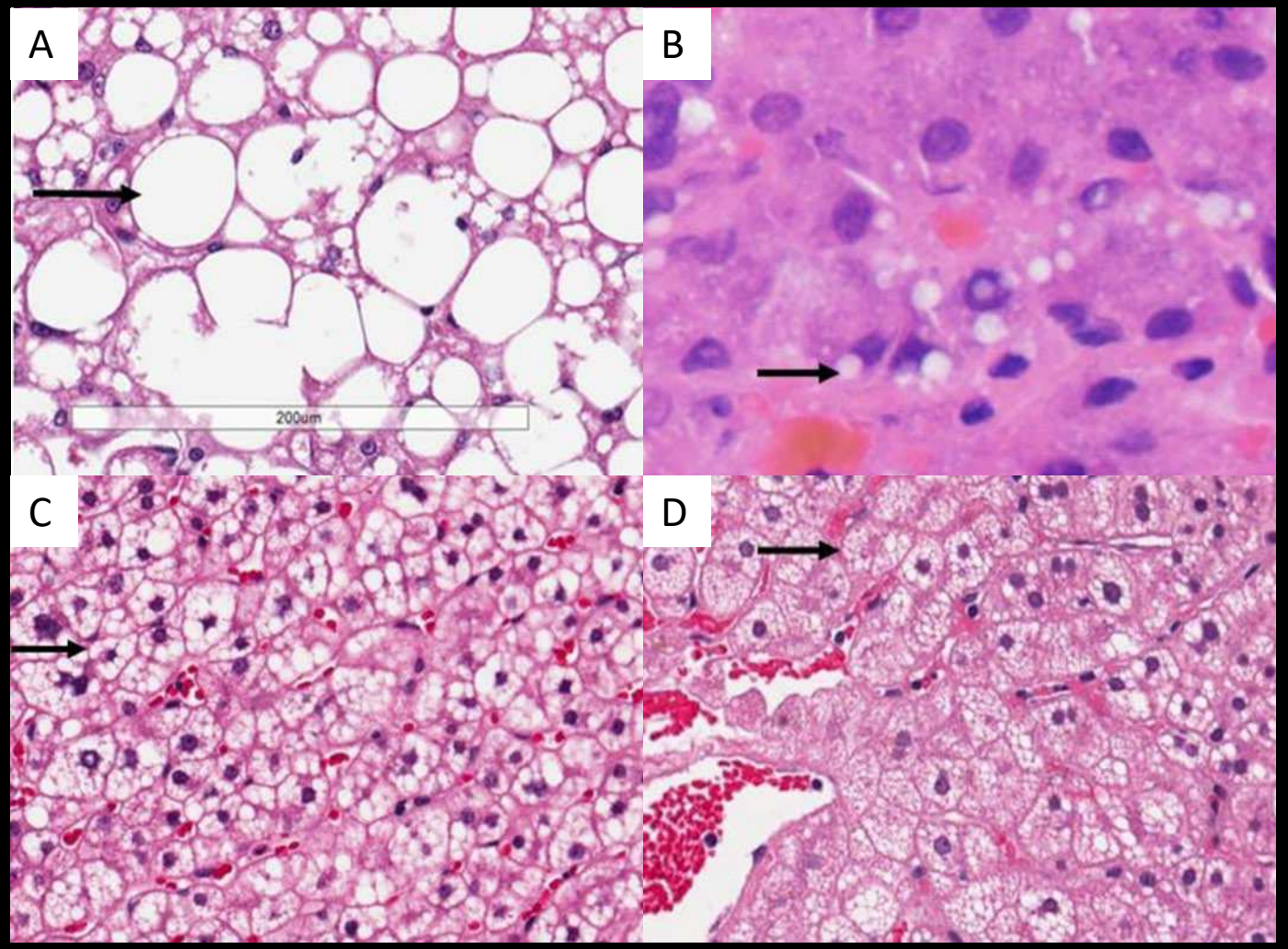
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