

Circulating TREM2 as a non-invasive diagnostic biomarker for NASH in patients with elevated liver stiffness

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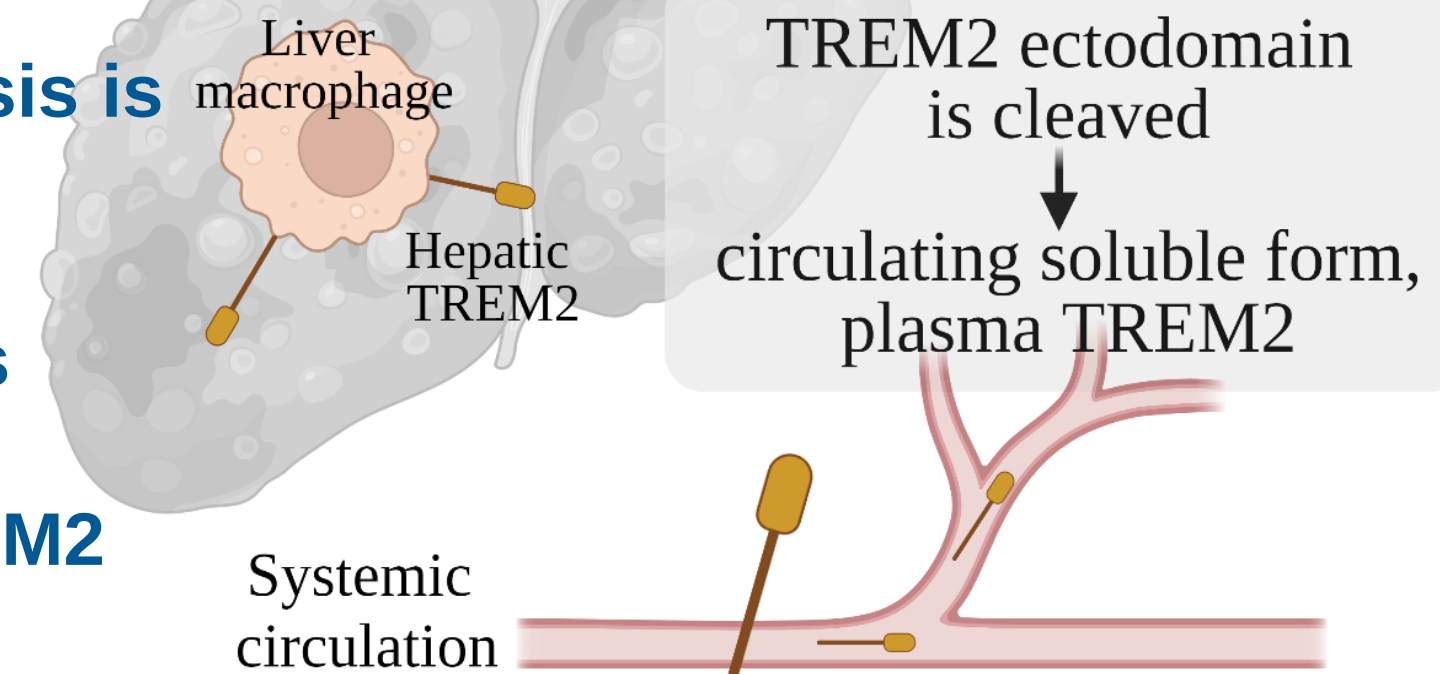
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INTRODUCTION

Reliable non-invasive biomarkers are an unmet clinical need for the diagnosis of NASH. Liver macrophages play a central role in the etiology of NASH and TREM2 is expressed on macrophages in NAFLD and NASH^{1,2}. TREM2 is further reported to be shedded from macrophages³.

Our hypothesis is that liver macrophage expression is reflected in levels of TREM2 in plasma.



AIM

In this study, we explored the potential of plasma TREM2 as a non-invasive biomarker to distinguish NASH patients. **The main outcome was diagnosis of NASH, defined as NAS $\geq$ 4**, in subjects with an elevated LSM $\geq$ 8 kPa. The secondary endpoint was to explore optimal rule-in/rule-out cut-offs for plasma TREM2.

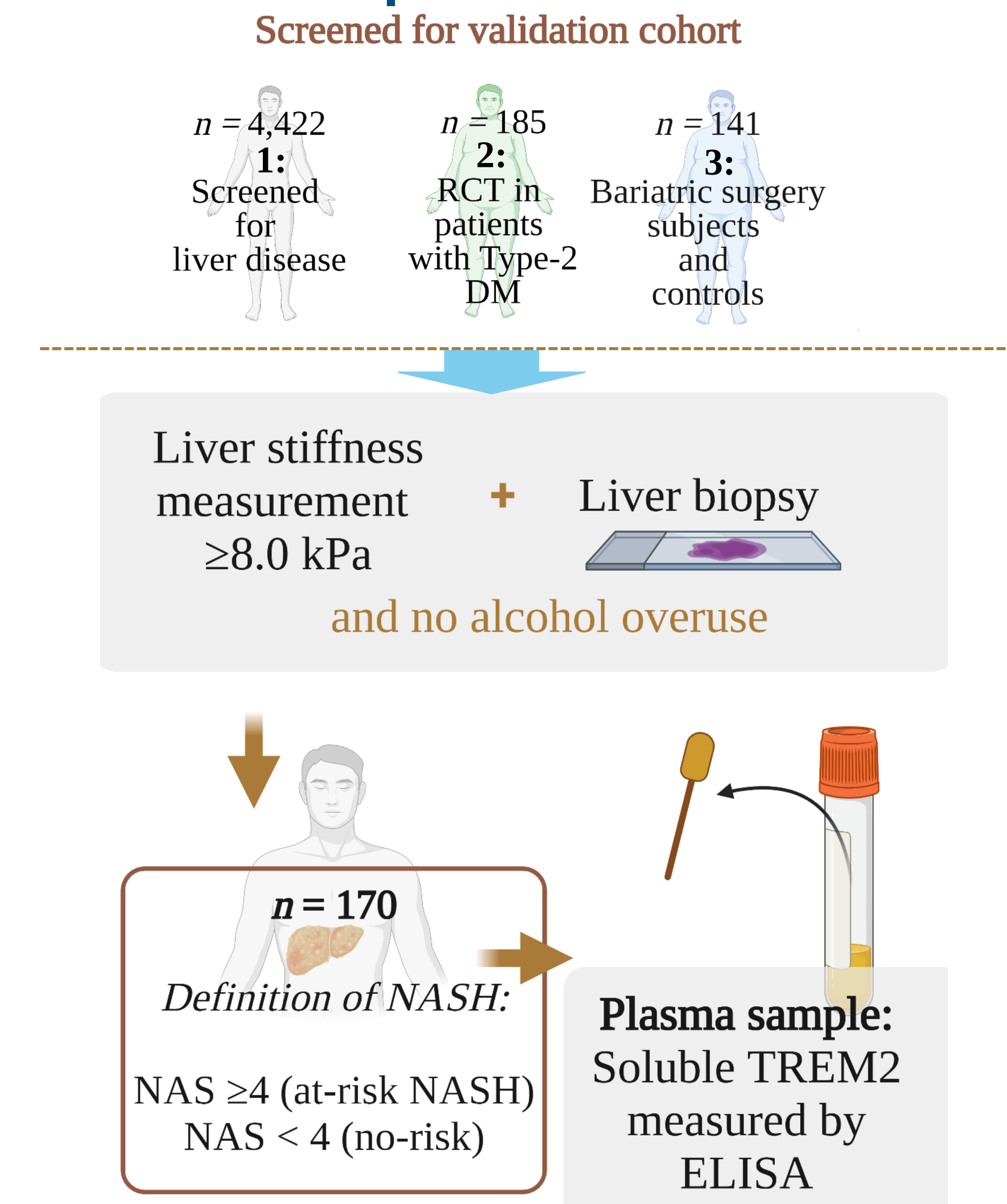
METHOD

Cross-sectional, clinical data including liver biopsies from a derivation (n=48) and a validation cohort (n=170) of **patients with elevated liver stiffness measurement (LSM $\geq$ 8.0 kPa)**.

Patients with NAFLD activity scores (NAS) $\geq$ 4 were defined as having NASH. Results presented in poster are from validation study. Histologic assessments according to NASH Clinical Research Network classification and hepatic fibrosis was assessed using Kleiner fibrosis stages F0-F4⁴.

The methods and results of this study were reported in accordance with the **Liver-FibroSTARD** standards.

Selection of patients for Validation



RESULTS

Study	1: Screened for liver disease n=75	2: Type 2 diabetes patients n=36	3 : Bariatric surgery patients n=59
Age (mean, SD)	57 (10)	56 (10)	45 (13)
Sex (Male, n %)	46 (61.3)	16 (44.4)	27 (45.8)
BMI (kg/m ²)	35.0 (10.3)	38.0 (12.1)	42.7 (8.6)
T2DM	32 (42.7)	36 (100.0)	20 (33.9)
Dyslipidemia	31 (41.3)	17 (47.2)	19 (32.2)
Hypertension	44 (58.7)	22 (61.1)	34 (57.6)
ALT (U/L)	44.0 (27.0)	37.0 (26.8)	53.0 (42.0)
AST (U/L)	33.0 (24.5)	27.0 (16.8)	36.5 (28.3)
TREM2 in NAS<4	30.4 (17.4)	46.5 (20.2)	33.0 (27.2)
TREM2 in NAS $\geq$ 4	63.6 (39.8)	74.4 (51.7)	86.5 (81.9)
TE (kPa)	11.9 (8.0)	11.0 (3.9)	11.8 (10.6)
NAS group (%) 0-1/2-3/4-5/ $\geq$ 6	13/47/31/9	8/56/33/3	14/39/37/10
SAF score (%) no-NAFLD/ NAFLD/ NASH	12/55/33	8/67/25	10/56/34
Fibrosis stage (%) 0/1/2/3/4	3/41/29/20/7	3/33/55/6/3	9/52/25/9/5

Plasma TREM2 concentration

Plots that show plasma conc. TREM2 and separate histologic features

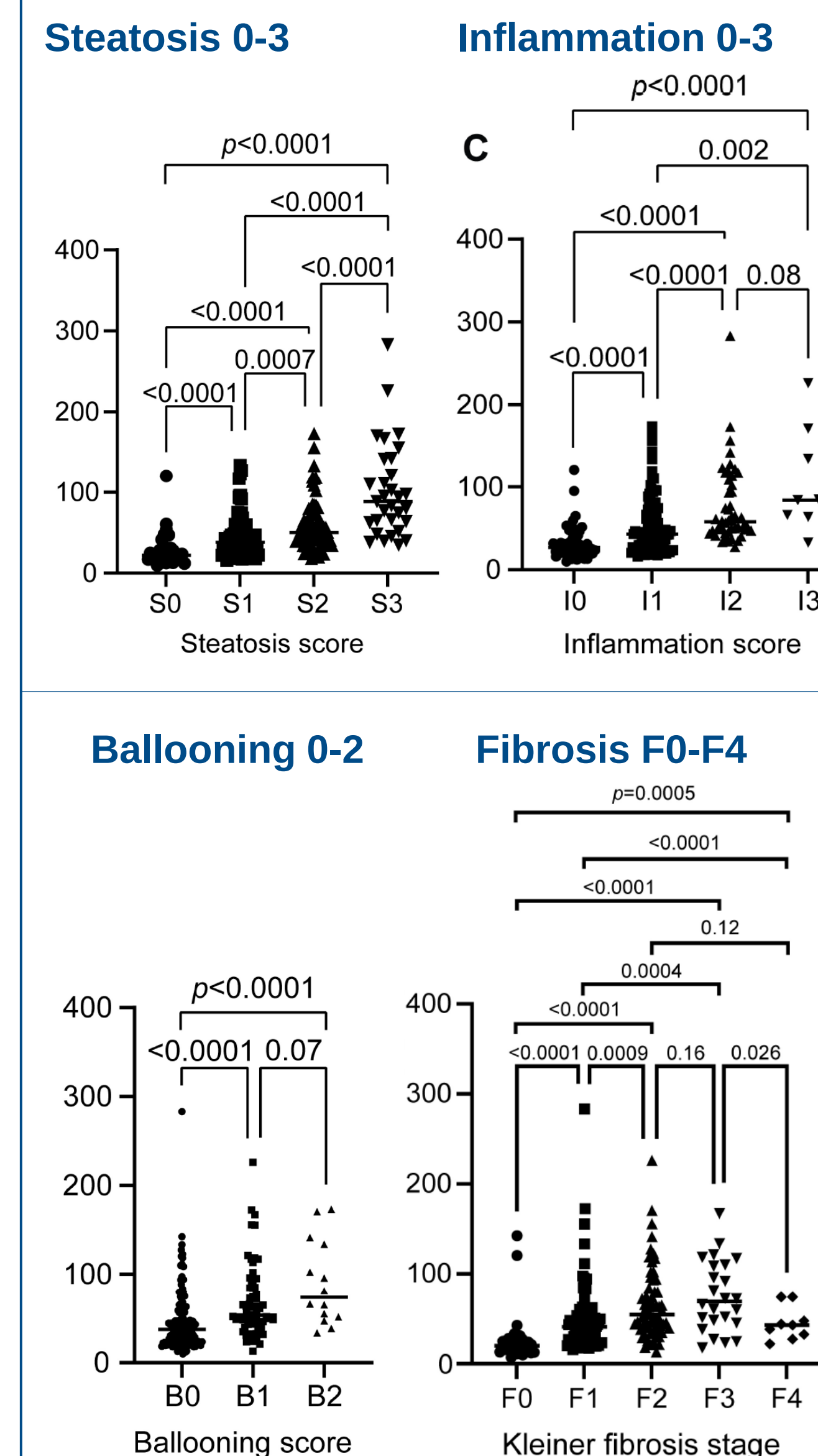
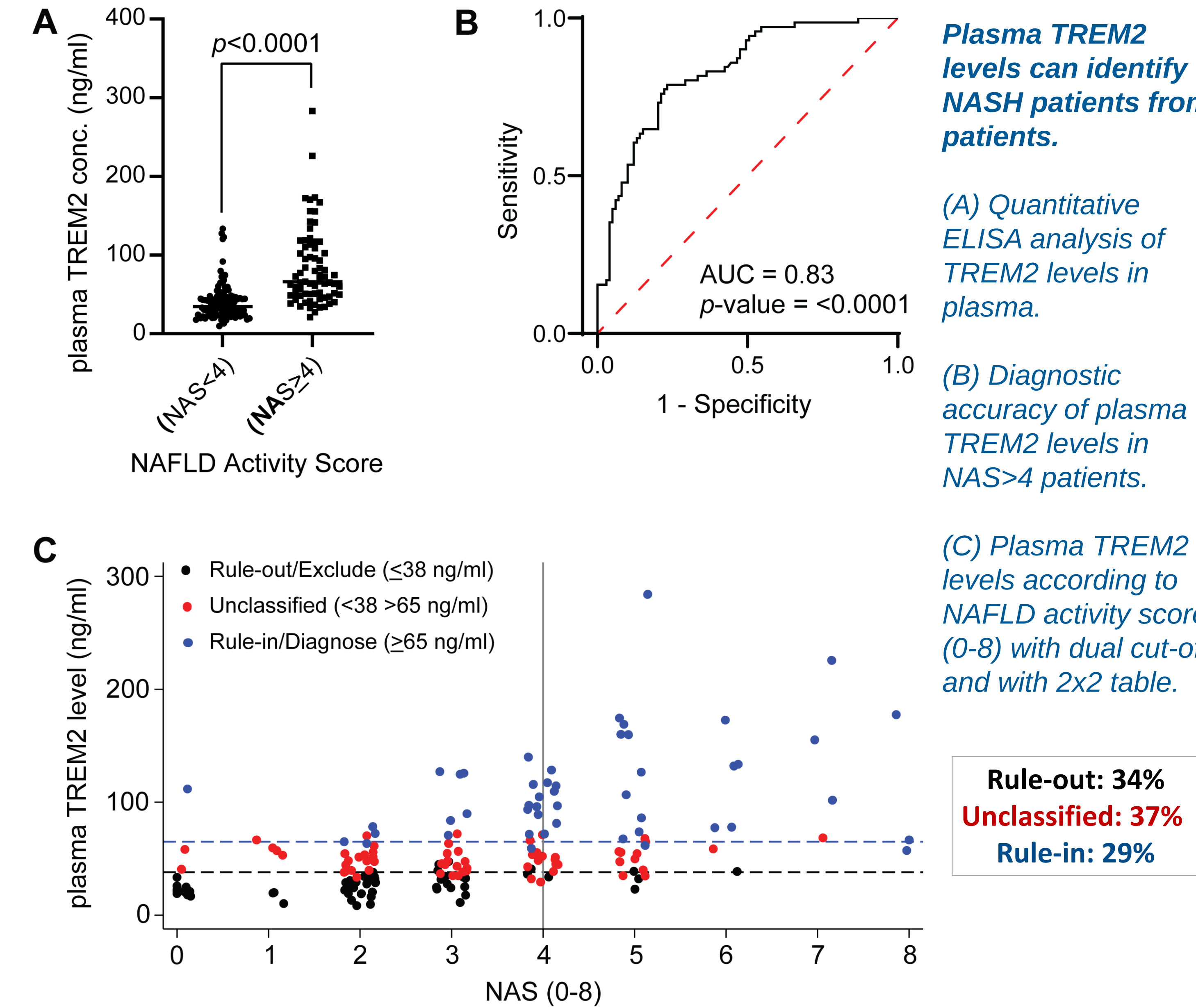


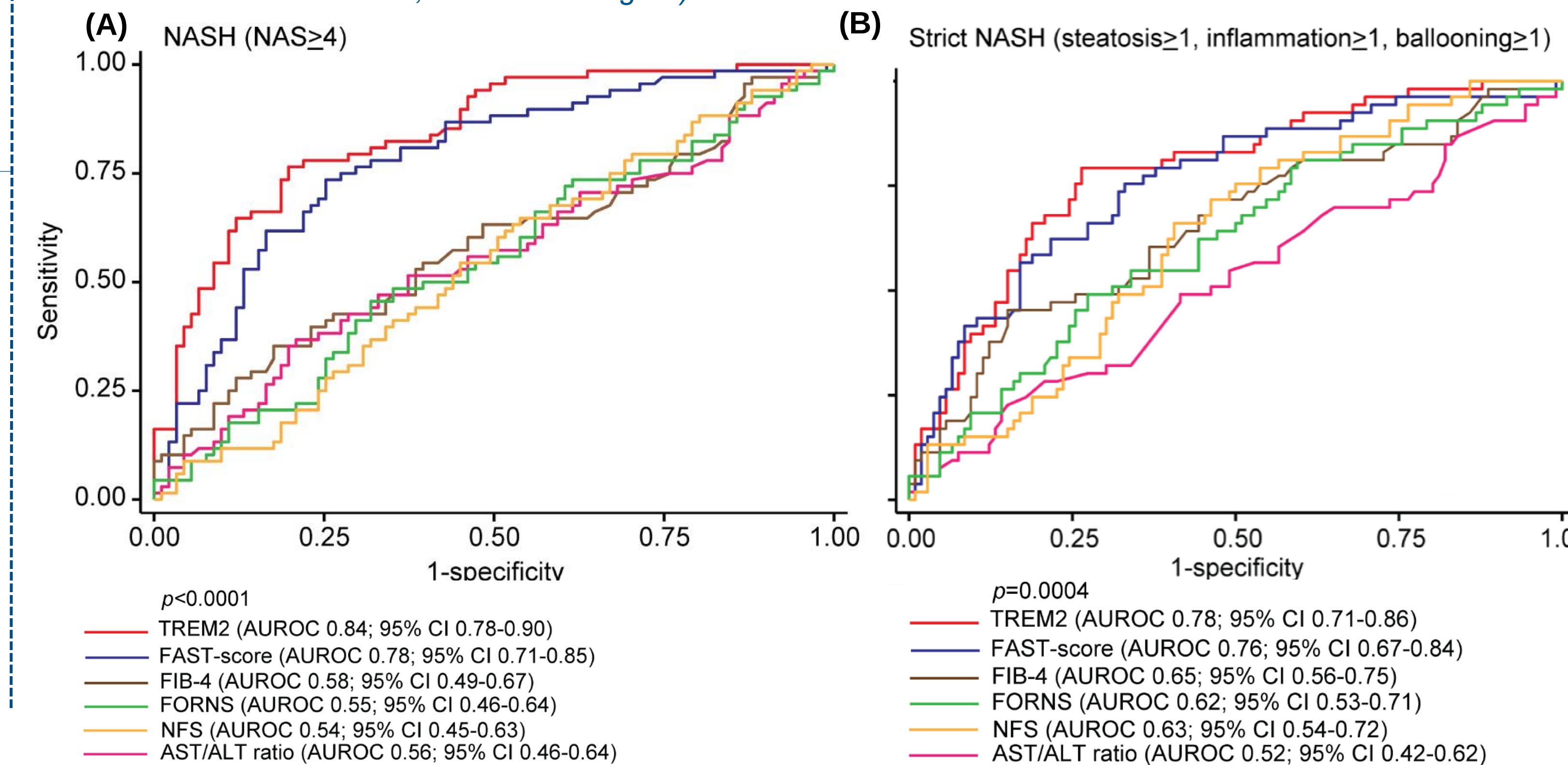
Table with patient characteristics, by cohort. Medians and iqr if or mean and SD.



Pooled validation cohort (n=170)		Plasma TREM2 (ng/ml)	Sensitivity (95% CI)	Specificity (95% CI)	PPV (95% CI)	NPV (95% CI)	LR+ (95% CI)
Rul	-risk	38	90% (81–96)	52% (41–62)	57% (47–67)	88 (77–95)	1.86 (1.5–2.31)
Rl	-risk	65	54% (41–66)	89% (81–94)	78% (63–88)	73% (64–80)	4.82 (2.65–8.76)

Comparative ROC curve – NAS $\geq$ 4 and strict NASH

Comparative ROC analysis with plasma TREM2, FAST-score, FIB-4, AST:ALT, Forns-index and NAFLD-score. (A). NASH defined as NAS $\geq$ 4; (B). Sensitivity analysis by strict-NASH by SAF score (Steatosis $\geq$ 1, Lobular inflammation $\geq$ 1, and Ballooning $\geq$ 1)



CONCLUSIONS

In conclusion, we have identified and validated plasma TREM2 as a **reliable non-invasive single biomarker to diagnose NASH in patients with elevated liver stiffness** in a large cohort of patients with histologically characterized NAFLD.

The ability of plasma TREM2 to identify NASH patients, and its strong association with NAFLD severity, makes it a **plausible individual biomarker for NASH**.

The diagnostic accuracy of plasma TREM2 to differentiate advanced fibrosis from lower fibrosis stages was low. Instead, single promising markers like plasma TREM2 should be further tested and externally validated in combination with specific markers of fibrosis such as TE or ProC3.

ACKNOWLEDGEMENTS

Open Patient data Explorative Network (<https://www.sdu.dk/en/forskning/open>).

REFERENCES

- Perugorria MJ, Esparza-Baquero A, Oakley F, Labiano I, Korosec A, Jais A, Mann J, et al. Non-parenchymal TREM-2 protects the liver from immune-mediated hepatocellular damage. Gut 2019;68:533-546.
- Ramachandran P, Dobie R, Wilson-Kanamori JR, Dora EF, Henderson BEP, Luu NT, Portman JR, et al. Resolving the fibrotic niche of human liver cirrhosis at single-cell level. Nature 2019;575:512-518.
- Schleppckow K, Kleinberger G, Fukumori A, Feederle R, Lichtenthaler SF, Steiner H, Haass C. An Alzheimer-associated TREM2 variant occurs at the ADAM cleavage site and affects shedding and phagocytic function. EMBO mol med 2017;9:1356-1365.
- Kleiner DE, Brunt EM, Van Natta M, Behling C, Contos MJ, Cummings OW, Ferrell LD, et al. Design and validation of a histological scoring system for nonalcoholic fatty liver disease. Hepatology 2005;41:1313-1321.

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