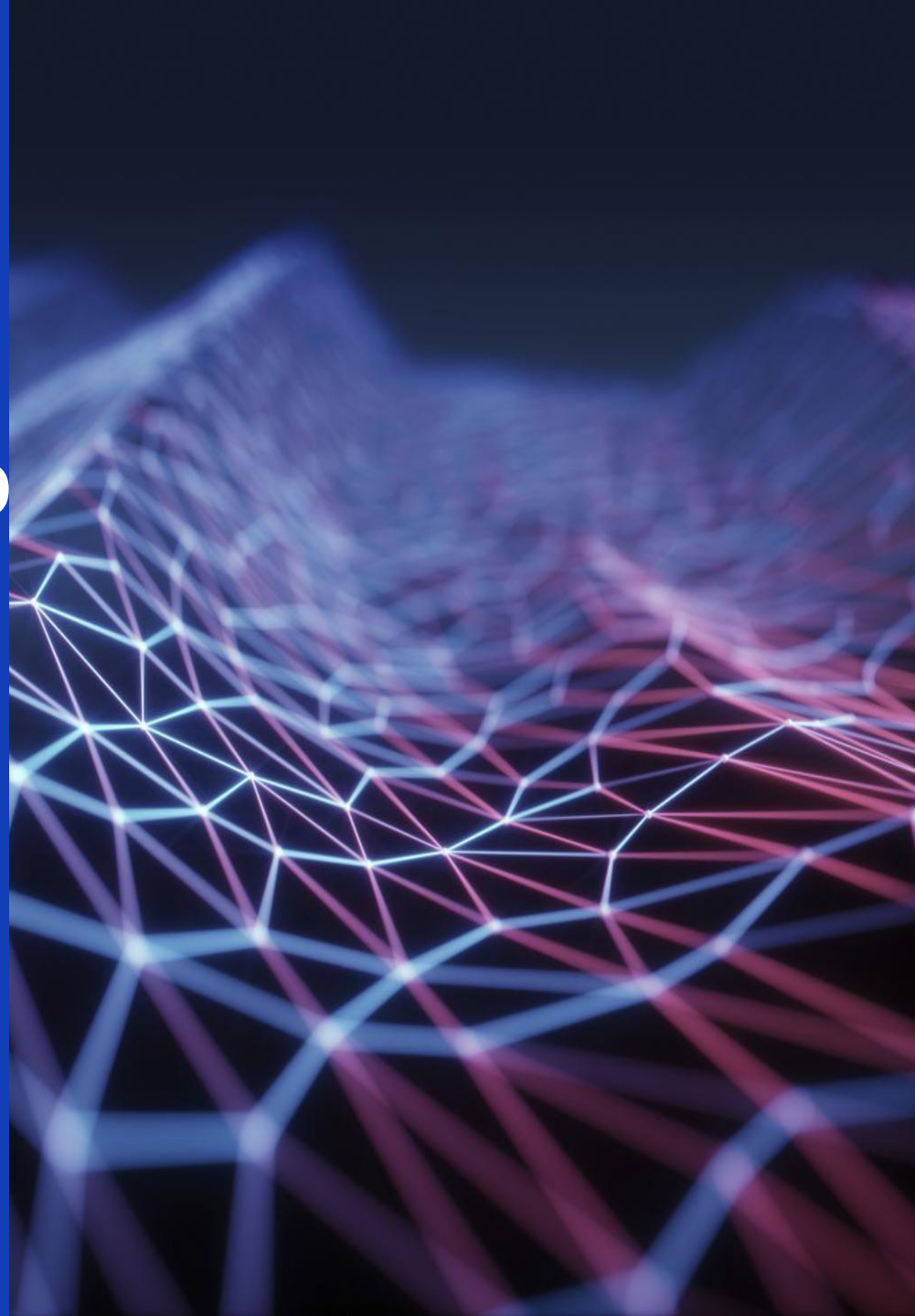
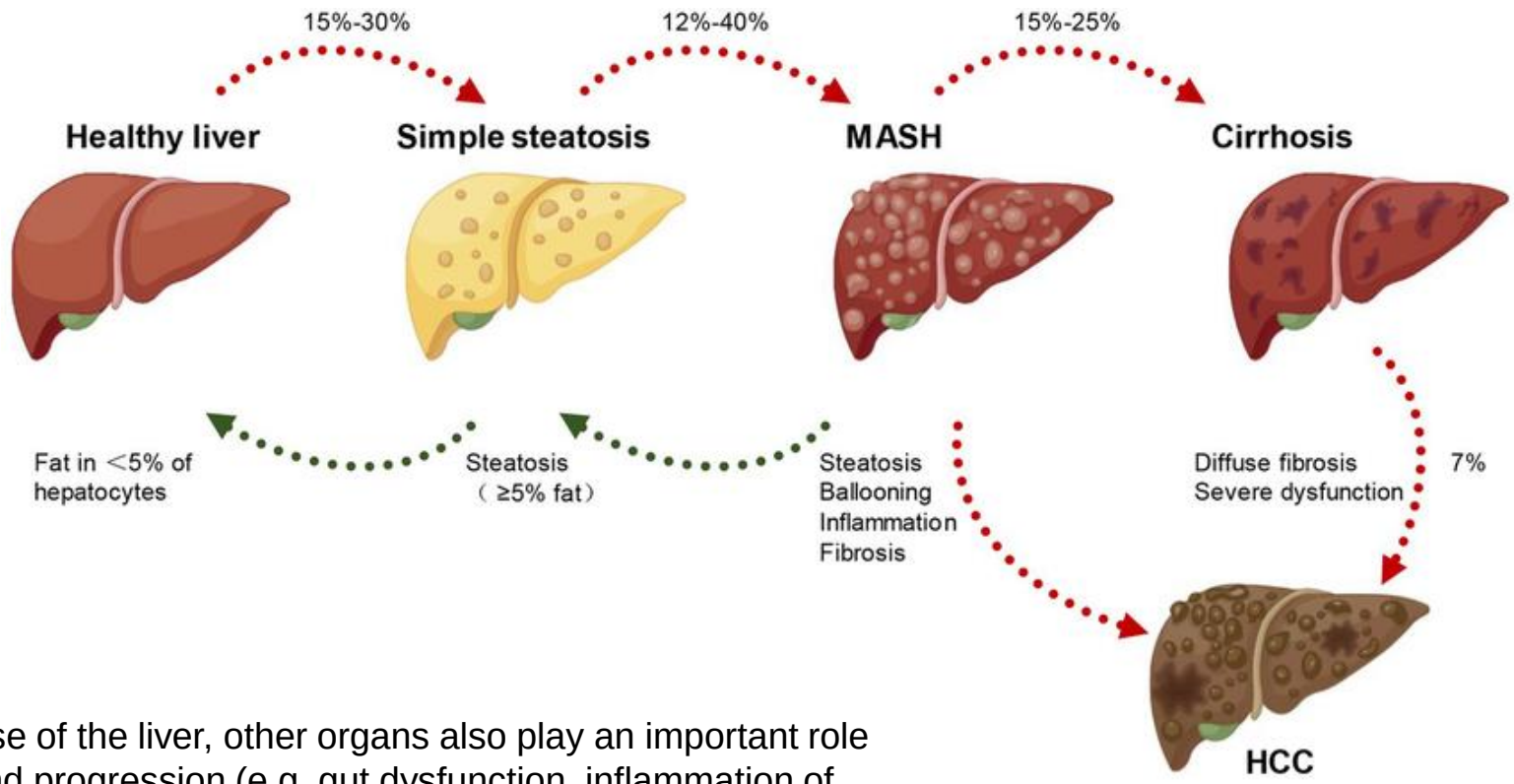
Novel functional biomarker panel (TLM3) for hepatic fibrosis in MASLD

Discovery, Validation and Applications

Lars Verschuren, PhD



Metabolic dysfunction-Associated Steatotic Liver Disease MASLD



MASLD is not just a disease of the liver, other organs also play an important role in disease development and progression (e.g. gut dysfunction, inflammation of adipose tissue).

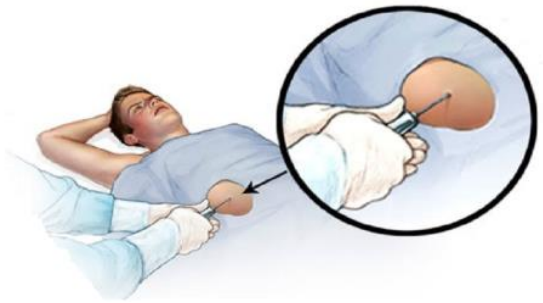
MASLD progression is considered a ‘silent disease’.

Rao et al. Front Med (Lausanne). 2023; 10: 1294267.

High demand for biomarkers for MASH diagnosis

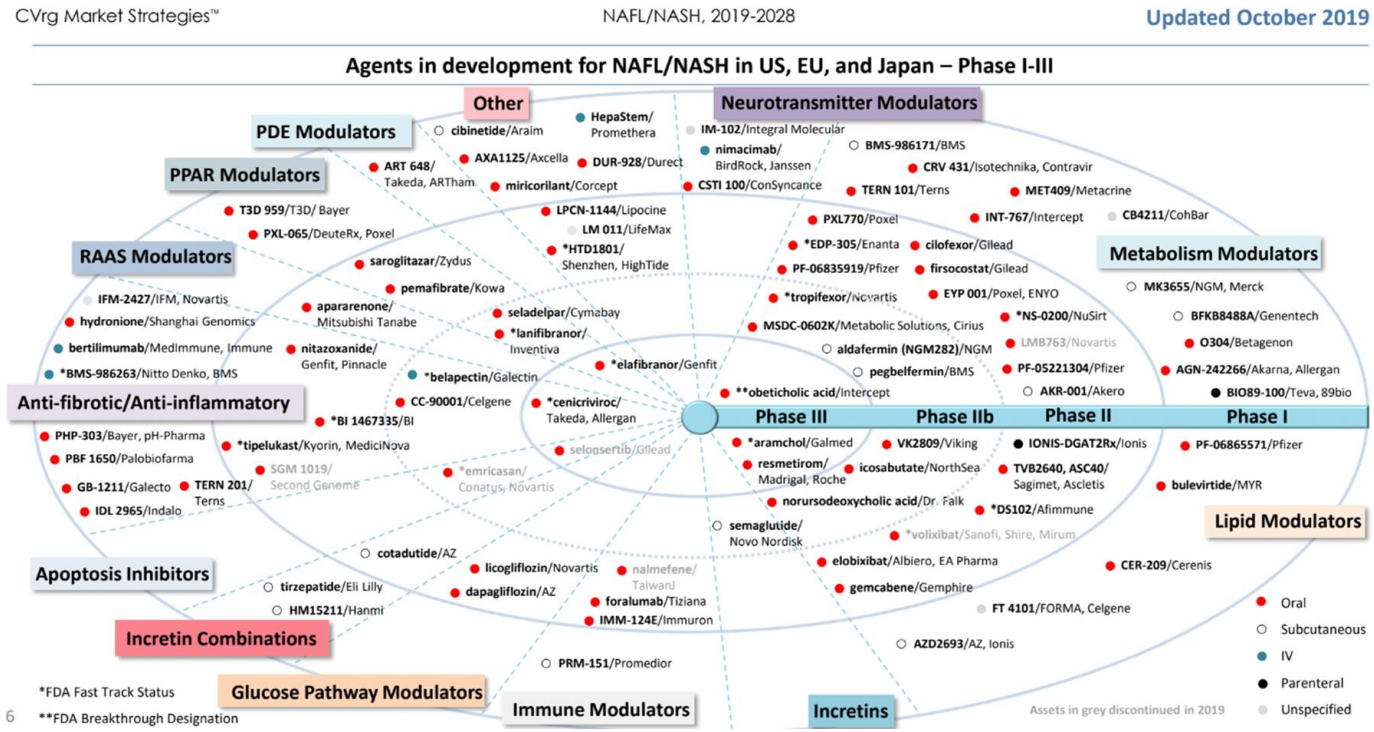
Liver fibrosis
Fast increasing prevalence “epidemic”
– late stage diagnosis -

Current diagnosis invasive (liver biopsy)



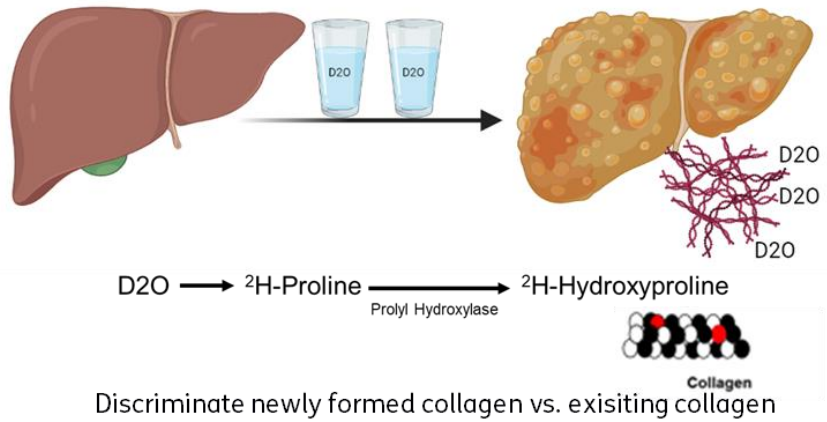
An invasive procedure and is not without risk.

There is a high need for blood-based biomarkers.

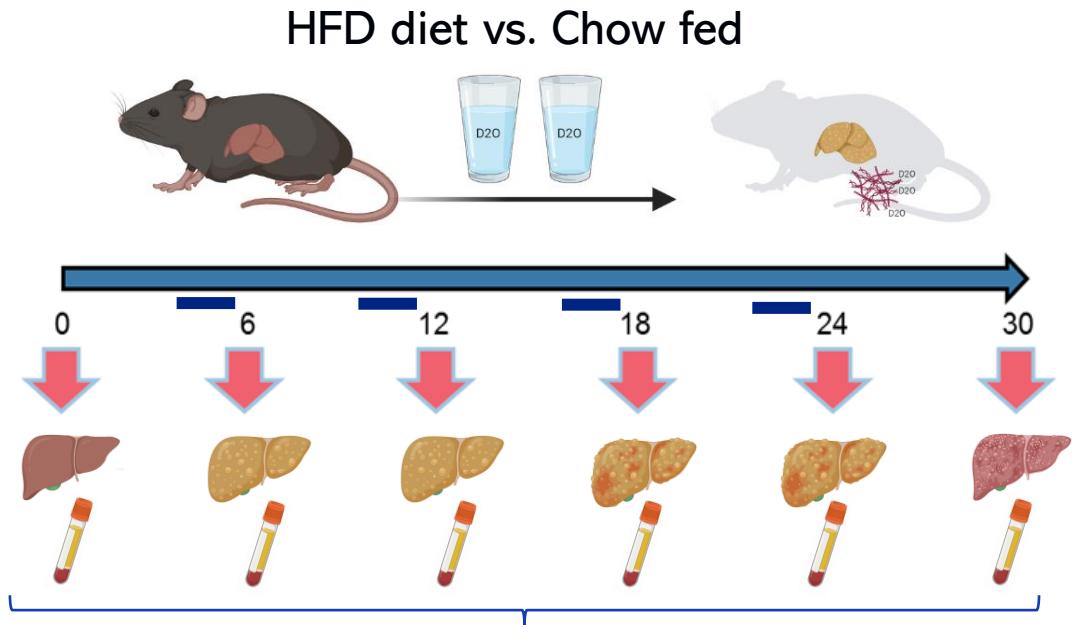


Pre-clinical identification of candidates

- Disease mechanisms are important
- Dynamics cannot be studied in human
- TNO experience in pre-clinical models (translational)
- Study disease dynamics in mice



Discriminate newly formed collagen vs. exisiting collagen

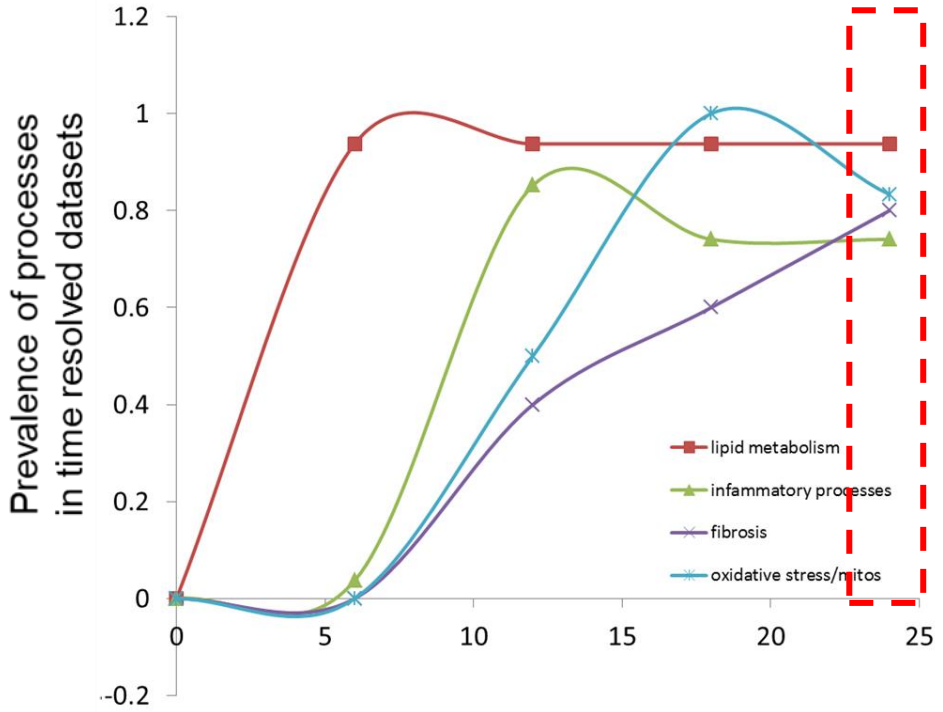


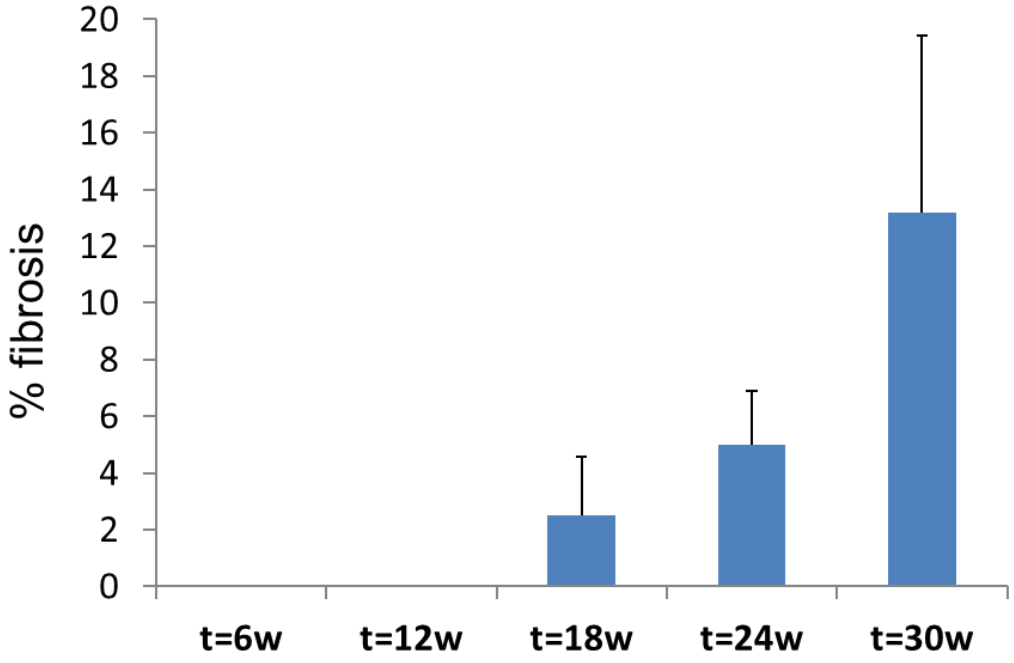
Analyses at different timepoints (every 6 weeks):

- Proteomics analysis of soluble and insoluble matrix proteins
- Histology
- RNAseq
- General metabolic disease parameters (ALT/AST, lipds, etc.)

This approach enables studying of the **dynamics** of ECM deposition in fibrosis development.

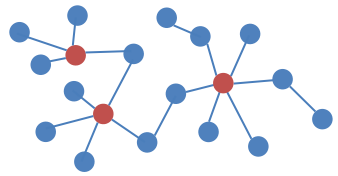
Dynamics of disease pre-clinical study





645 genes

Selected 12 biomarkers for diagnostic use.
Expressed in human liver and verified in serum of patients.



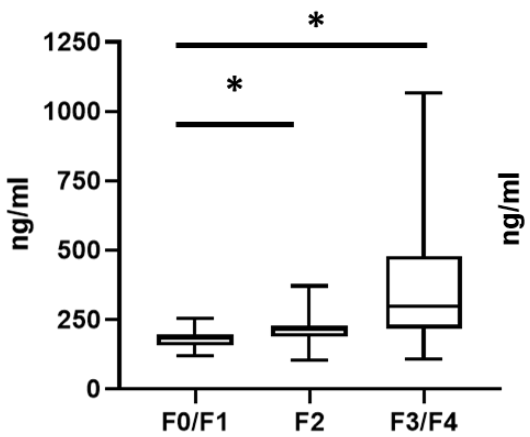
Mechanism based signature for fibrogenesis (645 genes)

van Koppen et al. Cell Mol Gastroenterol Hepatol 2018;5:83–98

Verification independent testing cohort

An independent cohort of 128 NASH patients with biopsy-proven fibrosis

A



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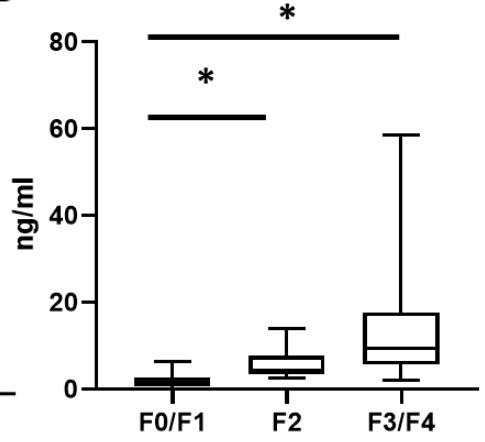

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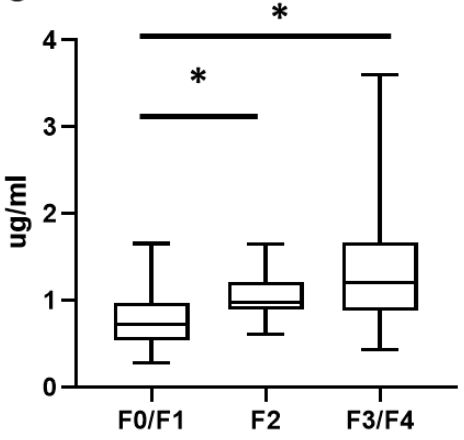

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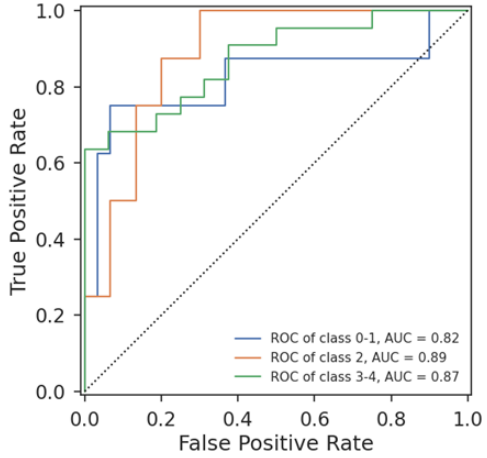

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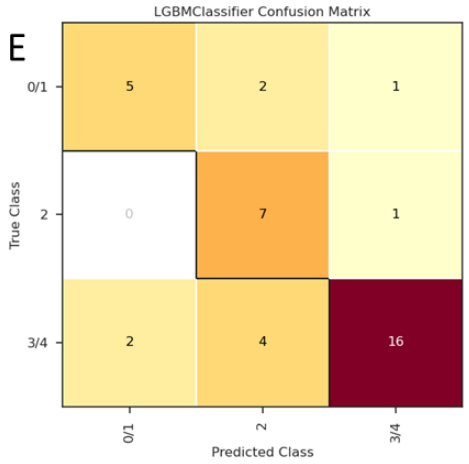


F

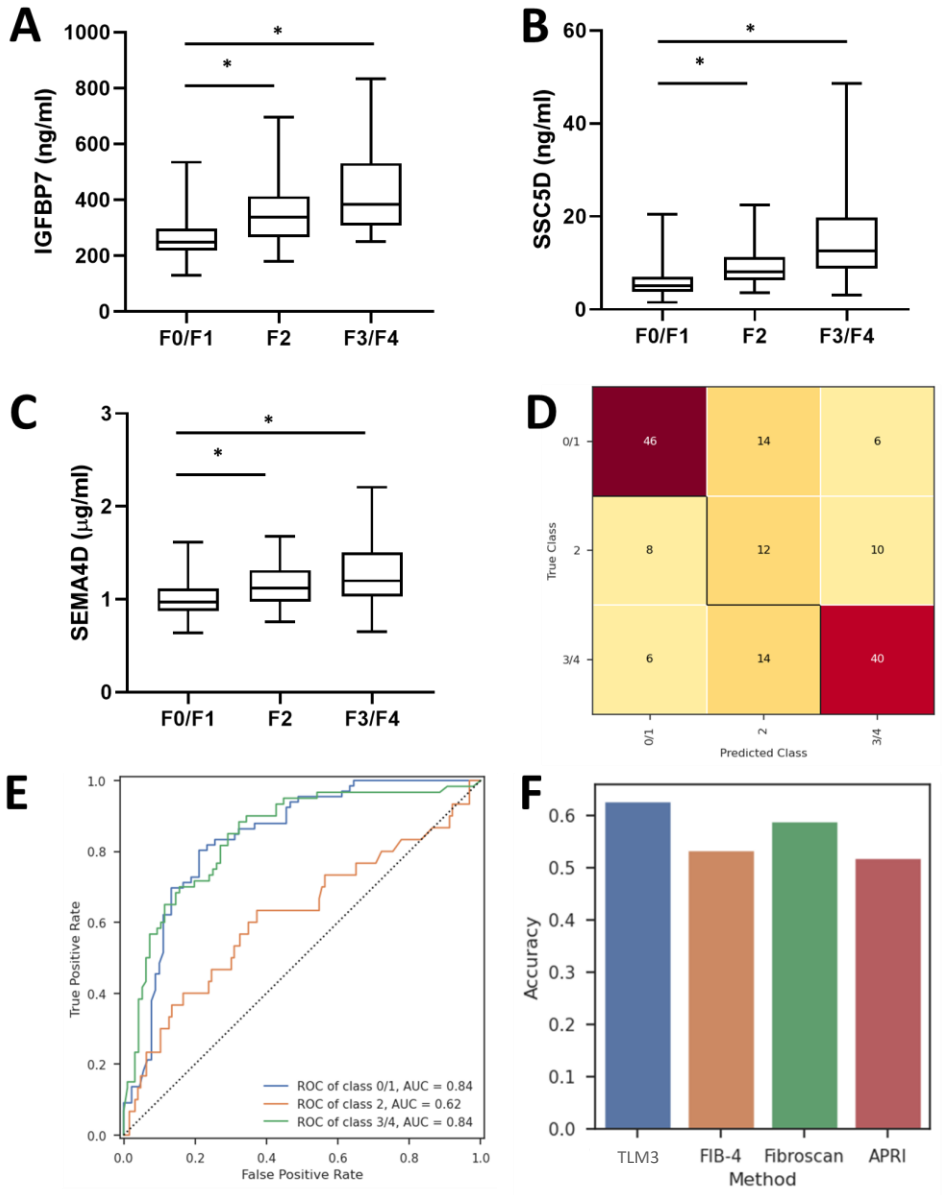
	Sensitivity	Specificity
F0-F1	0.625	0.903
F2	0.875	0.960
F3-F4	0.727	0.700

Validation of the model independer cohort

UNIVERSITY OF COPENHAGEN

- Validation of the LGBM model was performed using serum protein levels of the biomarkers analyzed in samples from patients in an independent Danish cohort.

- We successfully replicated the high-performance predictions in the testing cohort.
- We showed that the overall accuracy of our model outperformed the FIB-4, APRI and FibroScan predictions.



Conclusion

- We established a workflow starting from the disease mechanism to identify functional biomarkers using a translational model.
- This approach enabled the identification of candidate biomarkers for diagnosing hepatic fibrosis in MASLD.
- These biomarkers were clinically validated in two independent cohorts of biopsy-proven patients.
- In partnership with university medical centers in Leiden, Amsterdam, and Copenhagen.

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Article

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Development of a novel non-invasive biomarker panel for hepatic fibrosis in MASLD

Received: 23 August 2023

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Check for updates

Lars Verschuren[1,13], Anne Linde Mak[2,13], Arianne van Koppen[1], Serdar Özsezen[1], Sonia Difrancesco[1], Martien P. M. Caspers[1], Jessica Snabel[1], David van der Meer[3], Anne-Marieke van Dijk[2], Elias Badal Rashu[4], Puria Nabilou[4], Mikkel Parsberg Werge[4], Koen van Son[2], Robert Kleemann[1], Amanda J. Kiliaan[5], Eric J. Hazebroek[6], André Boonstra[7], Willem P. Brouwer[7], Michail Doukas[8], Saurabh Gupta[9], Cornelis Kluft[10], Max Nieuwdorp[2], Joanne Verheij[11], Lise Lotte Gluud[4], Adriaan G. Holleboom[2,14], Maarten E. Tushuizen[12,14] & Roeland Hanemaaijer[1,14]

Verschuren L., et al. Nat Commun **15**, 4564 (2024)

Applications:

1. Improved Diagnosis: Provides better diagnostic accuracy compared to current NITs.

2. Reduced Screening Failure: The use of TLM3 can significantly reduce screening failures observed in Fibroscan, particularly useful in clinical trials.

Reduced Screening Failure

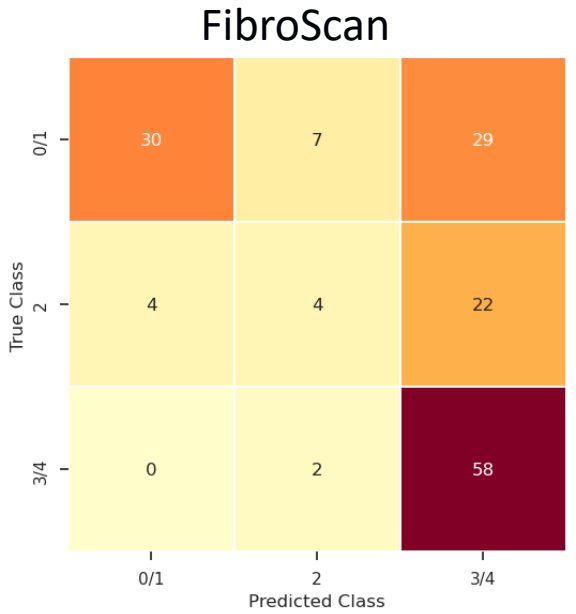
The most commonly used non-invasive test (NIT) for screening fibrosis in MASLD is transient elastography (TE), often known by FibroScan.

There are various factors that can cause variability of FibroScan analysis (Liver Stiffness Measurements; LSM) such as, hepatic inflammation[1], obesity[2], T2DM[3].

This leads often in overdiagnosis of patients predicted to be F3/F4 while biopsy shows F0/F1 or F2.

This results in clinical trials to screen a larger number of patients (biopsy) before the number of patients to be included is reached.

The use of TLM3 can significantly reduce screening failures observed in Fibroscan, particularly useful in clinical trials.



1) *World Journal of Gastroenterology*, 27, 641 – 653;
2) *Journal of hepatology*, 56 3, 564-70;
3) *European Journal of Gastroenterology & Hepatology* (2019).

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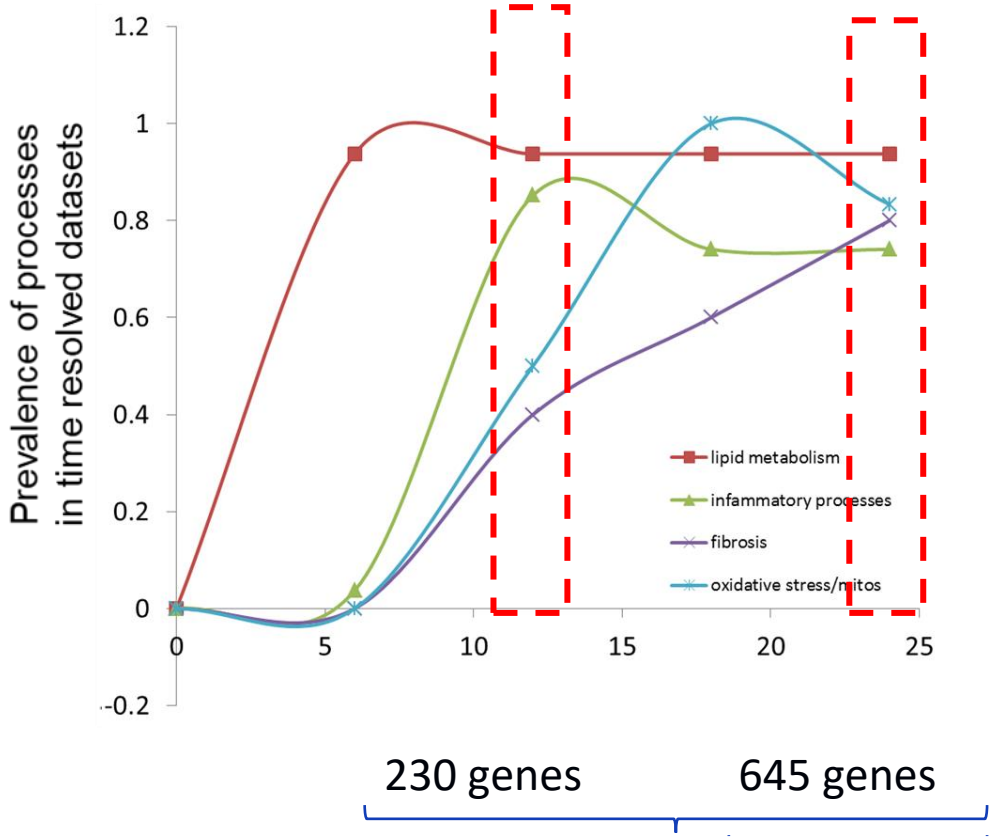
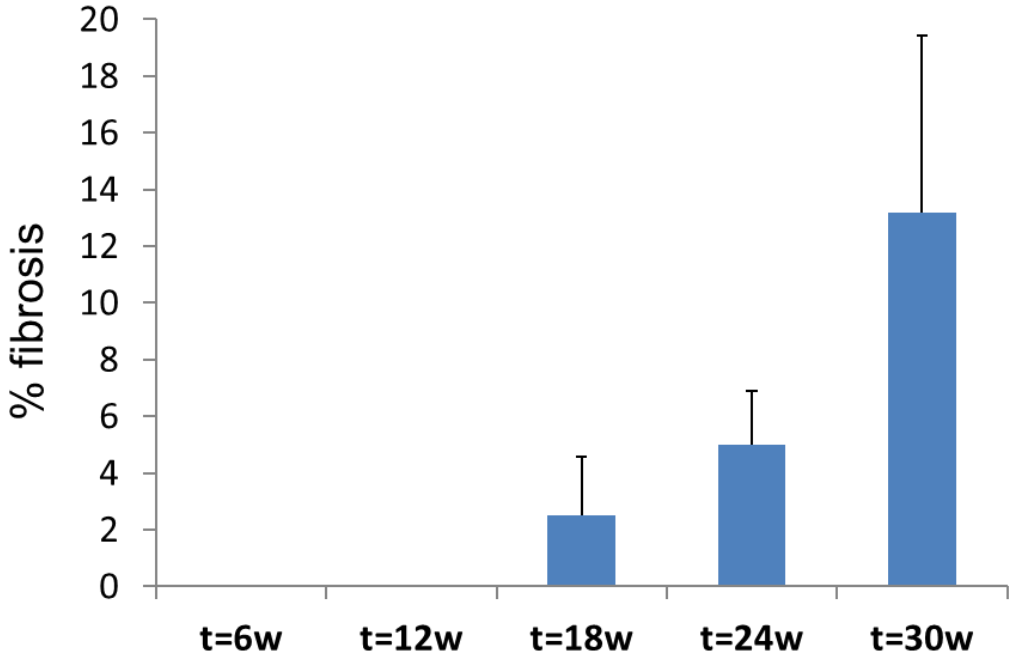
Projects in Development

3. Prognostic Tools: Developing tools to predict disease progression.

4. Monitoring Early Efficacy: Creating methods to monitor early treatment efficacy, with a focus on disease mechanisms.

Dynamics of disease pre-clinical study





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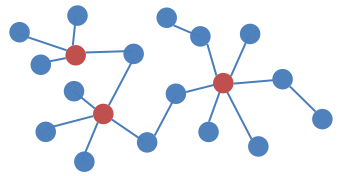

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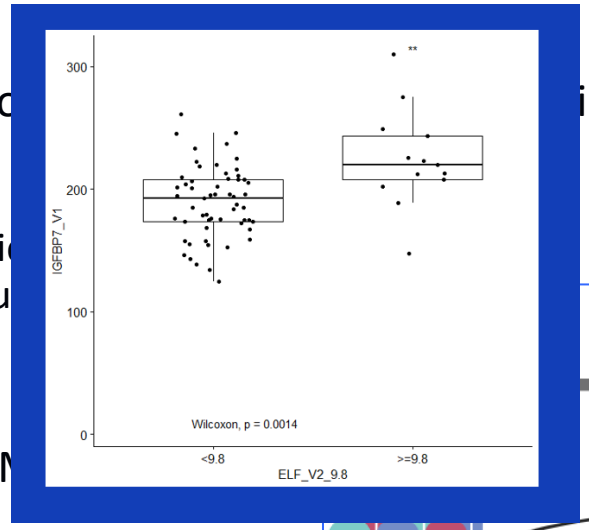
PoC study: Early diagnosis liver fibrosis

- Identification of candidate pro ibrosis

  - From dynamic preclinical study
    in combination with human clini
  - Can be measured in human seru
  - Assays available

- Initiated collaboration with AM

  - Longitudinal study, selection of 80 people
  - 6 years follow up
  - 40 with fibrosis, 40 without fibrosis
  - Biomarkers have been measured in serum
  - Next step: analysis / ML model building



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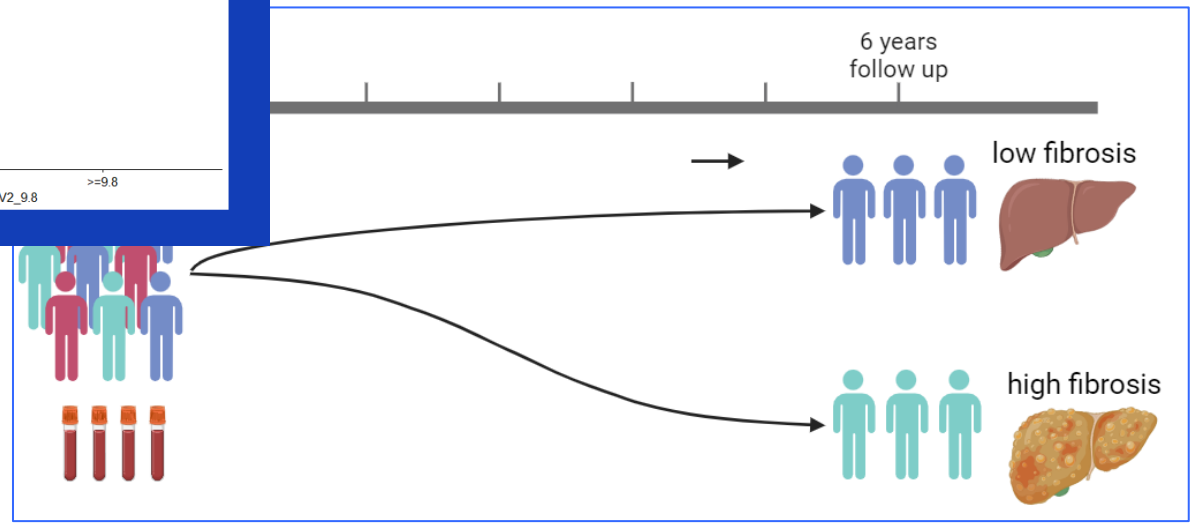

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