# *HepQuant Update:*

## *Progressing toward a Sensitive and Reliable Endpoint for Clinical Trials*

# *Safe Harbor and "Forward-looking Statements"*

"These materials include "forward-looking statements" within the meaning of the safe harbor provisions of the United States Private Securities Litigation Reform Act of 1995. These statements are based upon the current beliefs and expectations of HepQuant's management and are subject to significant risks and uncertainties. If underlying assumptions prove inaccurate or risks or uncertainties materialize, actual results may differ materially from those set forth in the forward-looking statements. HepQuant's products are not FDA approved, and are for investigational use only under an Investigational Device Exemption. These products are not offered for sale to physicians or patients. None of the descriptions of potential uses of the products should be interpreted as a statement that the products are safe or effective for the possible uses discussed."

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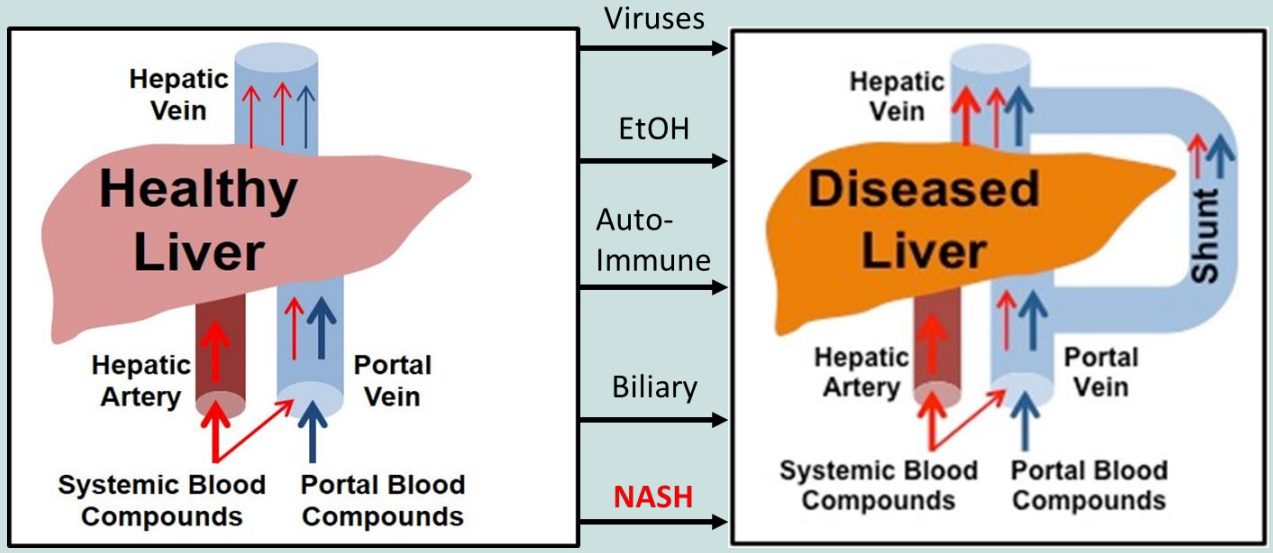
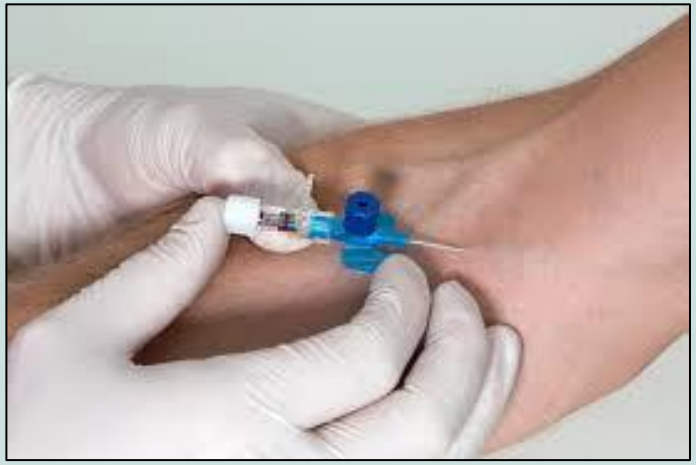
# *The HepQuant Platform of Tests*

# HepQuant SHUNT Test



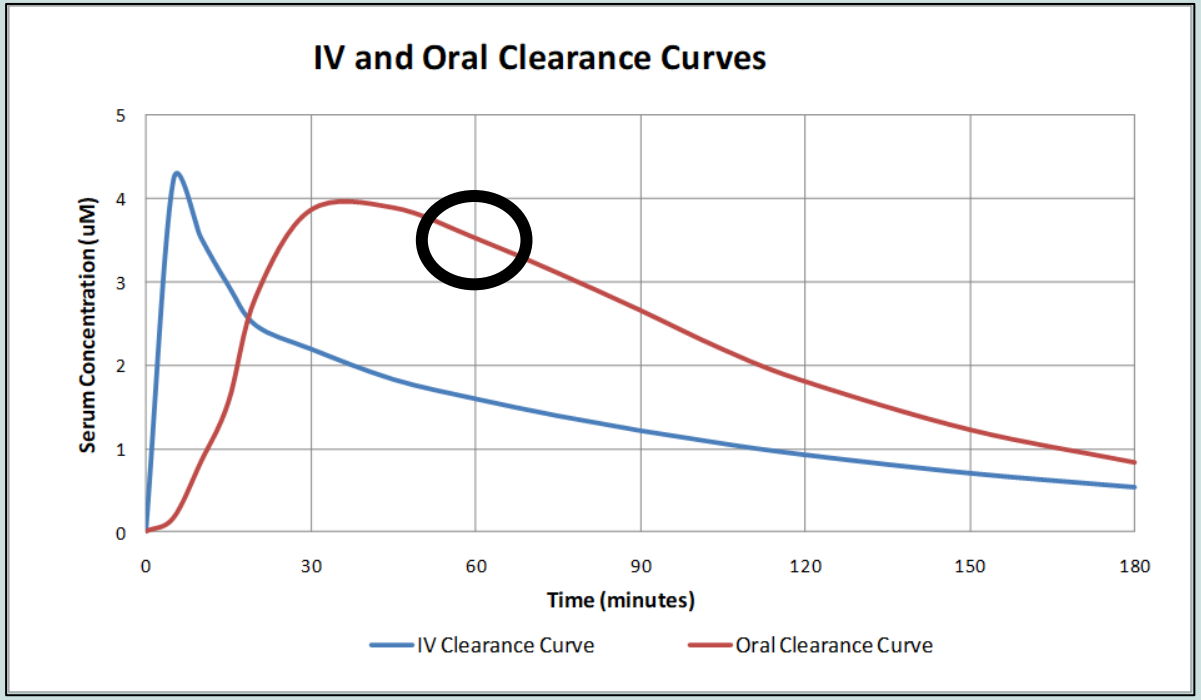



- Liver cell function – cholate uptake via specific hepatic transporters
- Dual blood inflows – portal and systemic (HFRs)
- Portal-systemic shunting (SHUNT%)

- Disease Severity Index (DSI) - global liver function

- Cholate is an endogenous primary bile acid in man.
- D4- and 13C-cholates are labeled with stable non-radioactive isotopes. NO RADIATION OR XENOBIOTIC.
- D4-cholate is delivered to the liver via portal vein and 13C-cholate via systemic circulation.
- D4-cholate, 40 mg, is taken orally and 13C-cholate, 20 mg, is administered intravenously.
- Blood is sampled at t = 5, 20, 45, 60 and 90 min via an indwelling intravenous catheter.

# *HepQuant STAT Test*



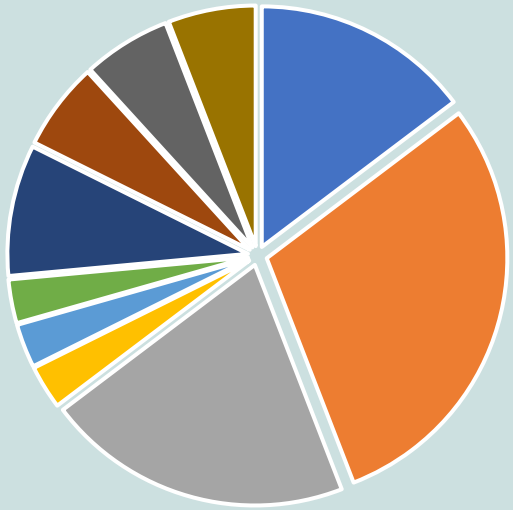

- **STAT is based on [D4-cholate] at t=60 minutes (red circle) of the oral clearance curve that generates Portal HFR.**
- **STAT estimates Portal HFR and DSI.**
- **Like DSI, STAT defined likelihood of cirrhosis, varices, large varices, and risk for future clinical outcomes.**

- **NO intravenous catheter**
- **Oral only (D4-cholate, 40 mg)**
- **Timed blood draw at t = 60 minutes**
- **Quantifies STAT and estimates Portal HFR and DSI**

# *HepQuant's "VALUE ADD" to Drug Development and Clinical Trials*

# Partnering Opportunities: HepQuant R&D Spans all CLD and Liver Research

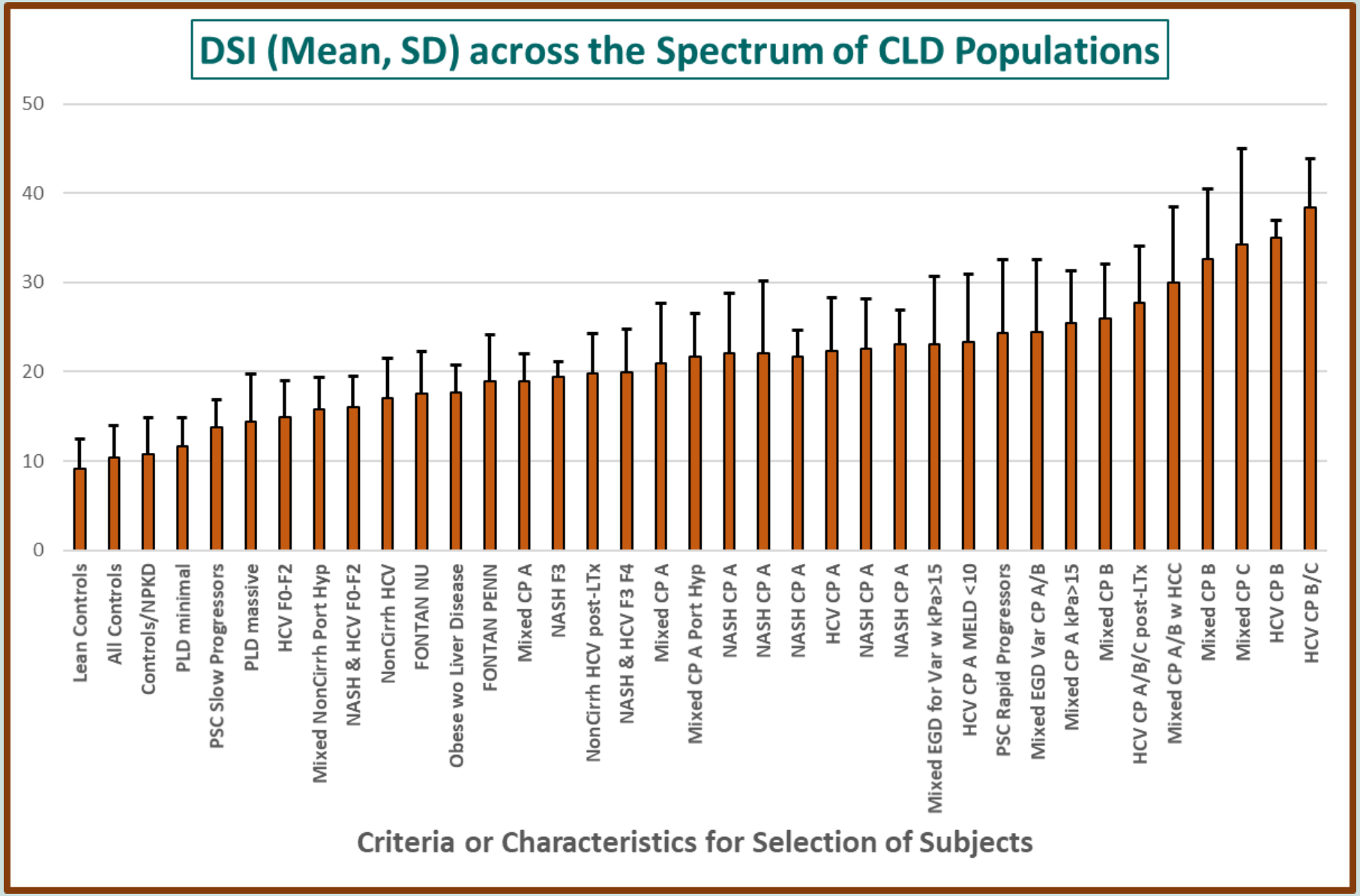
**22 Trials Completed, 5+ Ongoing**



- **CLD (includes NASH)**
- **NASH**
- HCV
- **Hepatic Impairment**
- PBC
- PSC
- **HCC**
- **Portal Hypertension**
- TIPS
- Congenital Heart  FONTAN

**2,000+ Tests / 1,000+ Subjects Completed**

**New cutting-edge treatment developers are talking with HepQuant to measure liver effects (telomere lengthening, RNA therapies, regenerative therapies, etc.**



# HepQuant Test Results are Reliable within a Single Individual

| | All Subjects | MDD | NASH + HCV | MDD |
|---|---|---|---|---|
| | ICC (95% CI) | | ICC (95% CI) | |
| DSI | **0.94 (0.90, 0.96)** | **1.47** | **0.92 (0.87, 0.96)** | **1.54** |
| Systemic HFR | 0.82 (0.73, 0.89) | 1.21 | 0.69 (0.52, 0.82) | 1.57 |
| Portal HFR | 0.84 (0.76, 0.90) | 5.18 | 0.85 (0.75, 0.92) | 3.17 |
| SHUNT | 0.74 (0.63, 0.84) | 10.2 | 0.78 (0.65, 0.88) | 9.3 |
| STAT | 0.90 (0.84, 0.94) | 0.18 | 0.88 (0.79, 0.93) | 0.23 |
| Hepatic Reserve | 0.95 (0.92, 0.97) | 2.6 | 0.93 (0.88, 0.96) | 3.2 |

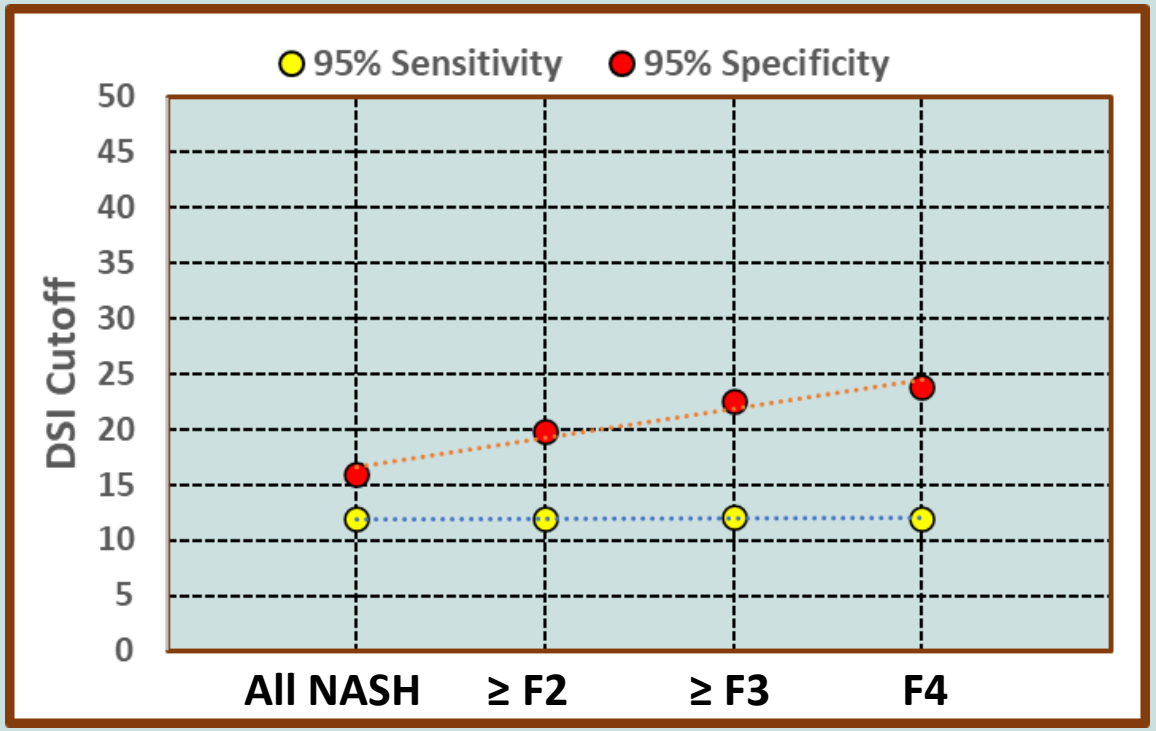
Abbreviations: Systemic and Portal HFR (mL $kg^{-1}$ $min^{-1}$), the hepatic filtration rate derived from the clearance of IV 13C-cholate or PO 4D-cholate, respectively, corrected for body weight; SHUNT (%), Systemic HFR/Portal HFR; DSI, disease severity index which is a score derived from modeling Systemic HFR, Portal HFR, and SHUNT for future clinical outcomes; STAT, [4D-cholate] at 60 minute time point normalized for body weight.

Burton JR, et al. The within-individual reproducibility of the disease severity index from the HepQuant SHUNT test of liver function and physiology. Transl Res. 2021 Jan 2:S1931-5244(20)30321-2. doi: 10.1016/j.trsl.2020.12.010. Online ahead of print. PMID: 33400995

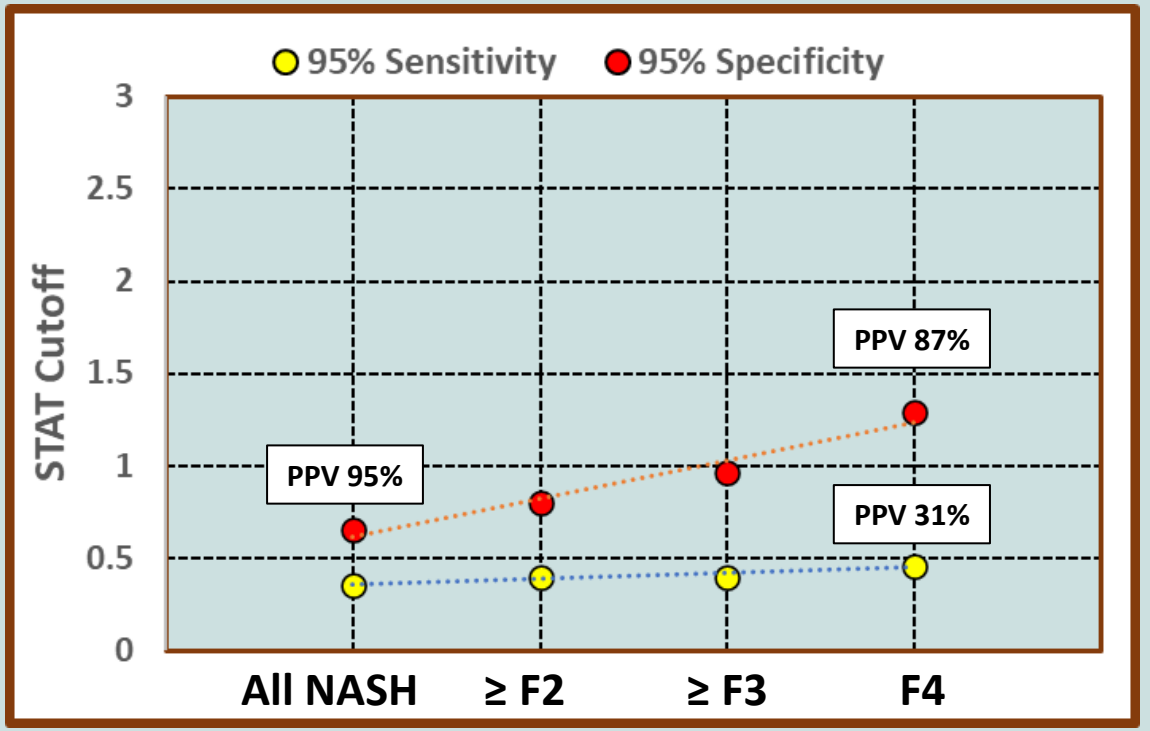
Matheson GJ. 2019. We need to talk about reliability: making better use of test-retest studies for study design and interpretation. PeerJ 7:e6918 http://doi.org/10.7717/peerj.6918 For calculation of **minimum detectable difference (MDD)** – the difference between two measurements in an individual that is sufficiently large that it is unlikely to have been due to chance alone.

# HepQuant in NASH and Relationship to Fibrosis Stage

## DSI




## STAT




* N=93: F0/F1 16, F2 12, F3 34, F4 31. ** N=50, 30 Lean, 16 Overweight, and 4 Obese healthy controls.

| | | Diagnostic Performance | | | |
|---|---|---|---|---|---|
| | | AUROC | Youden Cutoff | Sensitivity | Specificity |
| **NASH All** | FIB4 | NA | NA | NA | NA |
| | ALT | 0.678 | 40 | 0.632 | 0.648 |
| | NIS4 | 0.83 | 0.4 | 0.777 | 0.762 |
| | OWL | NA | NA | 0.773 | 0.668 |
| | ELF | NA | NA | NA | NA |
| | PROC3 | NA | NA | NA | NA |
| | FM-VCTE | NA | NA | NA | NA |
| | | | | | |
| | HQ-STAT | 0.875 | 0.55 | 0.75 | 0.86 |
| | HQ-DSI | 0.945 | 16.05 | 0.84 | 0.96 |

| | | Diagnostic Performance | | | |
|---|---|---|---|---|---|
| | | AUROC | Youden Cutoff | Sensitivity | Specificity |
| **NASH ≥ F2** | FIB4 | 0.796 | 1.4 | 0.654 | 0.808 |
| | ALT | NA | NA | NA | NA |
| | NIS4 | 0.874 | 0.4 | 0.823 | 0.799 |
| | OWL | NA | NA | NA | NA |
| | ELF | 0.828 | 9.5 | 0.718 | 0.815 |
| | PROC3 | 0.809 | 17.6 | 0.698 | 0.81 |
| | FM-VCTE | 0.841 | 0.5 | 0.667 | 0.864 |
| | | | | | |
| | HQ-STAT | 0.856 | 0.55 | 0.81 | 0.77 |
| | HQ-DSI | 0.903 | 16.05 | 0.88 | 0.82 |

| | | Diagnostic Performance | | | |
|---|---|---|---|---|---|
| | | AUROC | Youden Cutoff | Sensitivity | Specificity |
| **NASH F3 or F4** | FIB4 | 0.793 | 1.4 | 0.751 | 0.686 |
| | ALT | NA | NA | NA | NA |
| | NIS4 | 0.788 | 0.6 | 0.729 | 0.748 |
| | OWL | NA | NA | NA | NA |
| | ELF | 0.835 | 9.6 | 0.808 | 0.702 |
| | PROC3 | 0.764 | 18.8 | 0.714 | 0.714 |
| | FM-VCTE | 0.858 | 0.6 | 0.762 | 0.813 |
| | | | | | |
| | HQ-STAT | 0.844 | 0.66 | 0.69 | 0.87 |
| | HQ-DSI | 0.884 | 16.05 | 0.92 | 0.74 |

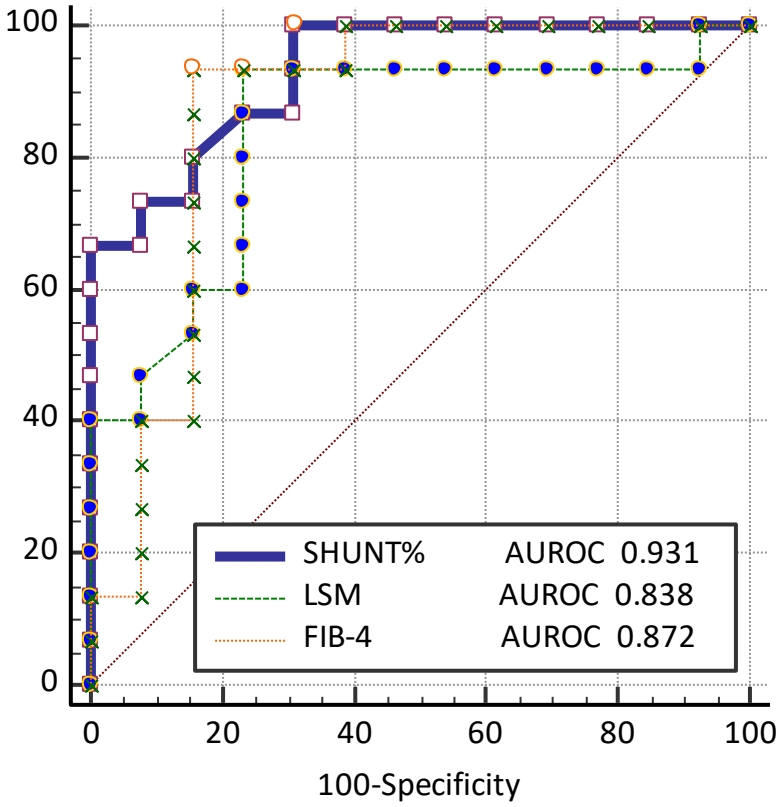
| | | Diagnostic Performance | | | |
|---|---|---|---|---|---|
| | | AUROC | Youden Cutoff | Sensitivity | Specificity |
| **NASH F4** | FIB4 | 0.81 | 1.5 | 0.85 | 0.634 |
| | ALT | NA | NA | NA | NA |
| | NIS4 | 0.725 | 0.6 | 0.781 | 0.614 |
| | OWL | NA | NA | NA | NA |
| | ELF | 0.855 | 10.1 | 0.821 | 0.733 |
| | PROC3 | 0.728 | 21.1 | 0.662 | 0.685 |
| | FM-VCTE | 0.897 | 0.6 | 0.942 | 0.704 |
| | | | | | |
| | HQ-STAT | 0.857 | 0.66 | 0.87 | 0.75 |
| | HQ-DSI | 0.877 | 19.51 | 0.81 | 0.84 |

For FIB4, ALT, NIS4, OWL, ELF, PROC3, FM-VCTE: N=220 w NAFL and 853 w NASH (from NIMBLE Stage-1, AASLD 2021).
For HQ-STAT and HQ-DSI: N=93: F0/F1 16, F2 12, F3 34, F4 31; plus N=30 Lean, N=16 Overweight, and N=4 Obese healthy controls.

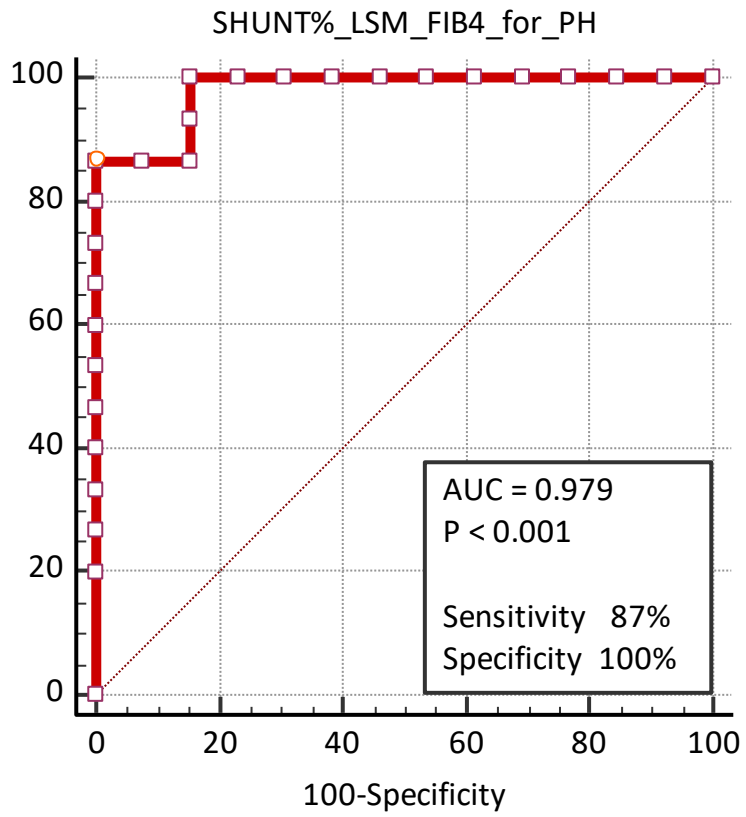
# *HepQuant Predicts Clinical Outcomes*

# *Portal Hypertension*

## Individual Non-Invasive Tests

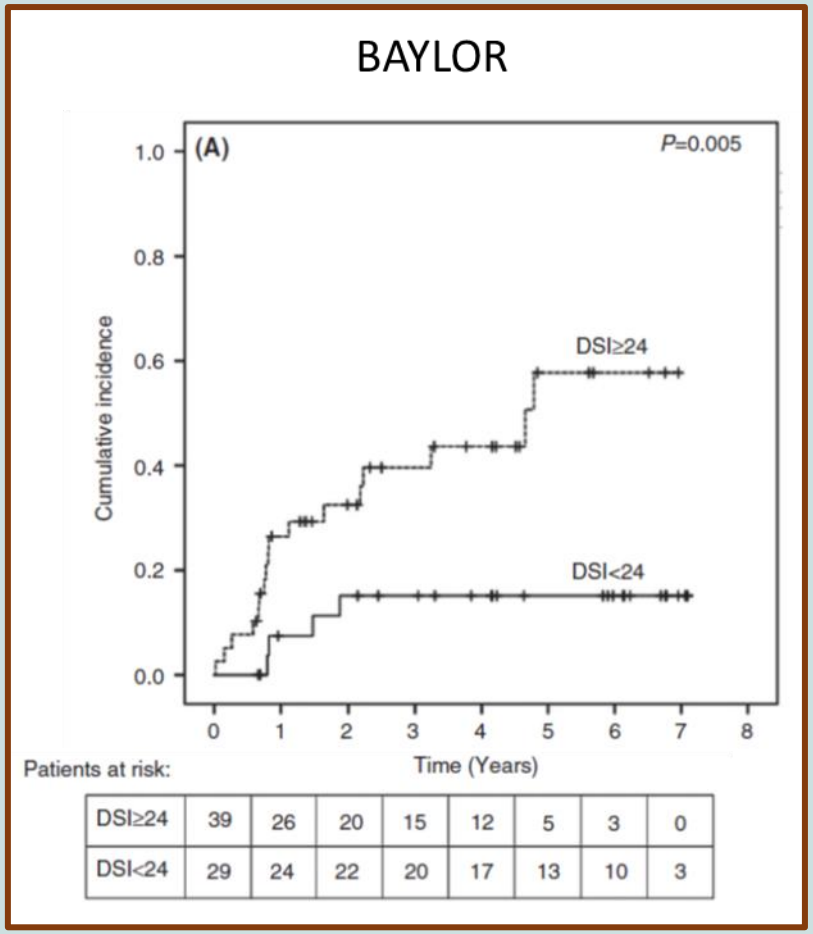



## Combination of Non-Invasive Tests

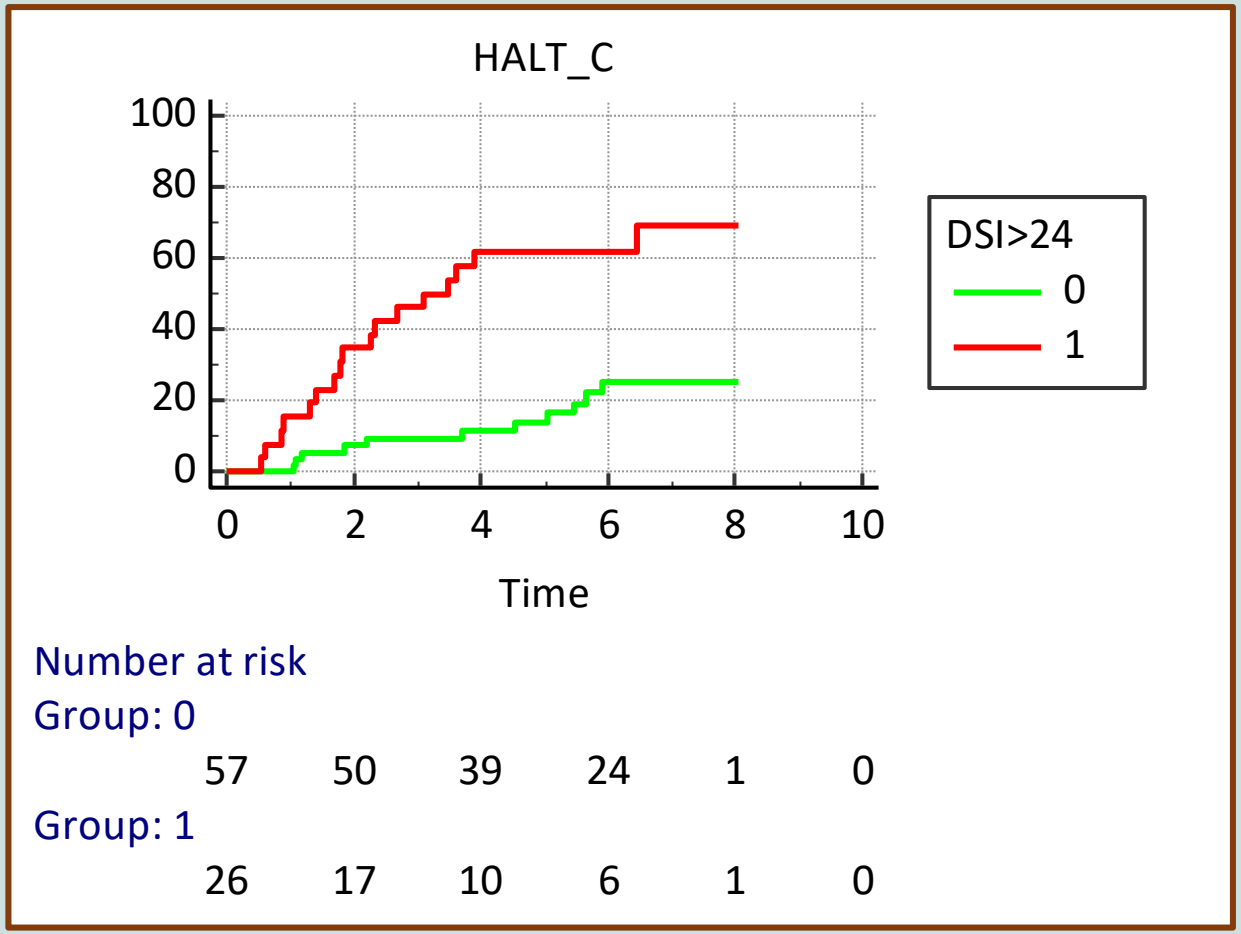



*HepQuant SHUNT Detects Portal Hypertension in Early Stages of Clinically Compensated Chronic Liver Disease.* Amanda Wieland, et al. Clin Gastroenterol Hepatol 2021 Apr 22;S1542-3565(21)00464-X. doi: 10.1016/j.cgh.2021.04.030

# Decompensation, Death, Transplant




Spectrum of Etiologies of Cirrhosis (20% had DSI>35; 13% had DSI<15)




HCV Etiology of Cirrhosis (2% had DSI>35; 8% had DSI<15)

*Predicting clinical decompensation in patients with cirrhosis using the Hepquant-SHUNT test. Fallahzadeh MA, et al. Aliment Pharmacol Ther. 2021;53:928–938.*

# THE HEPQUANT SHUNT TEST PREDICTS THE LIKELIHOOD OF FINDING ESOPHAGEAL VARICES AND PARTICULARLY LARGE VARICES AT ENDOSCOPY

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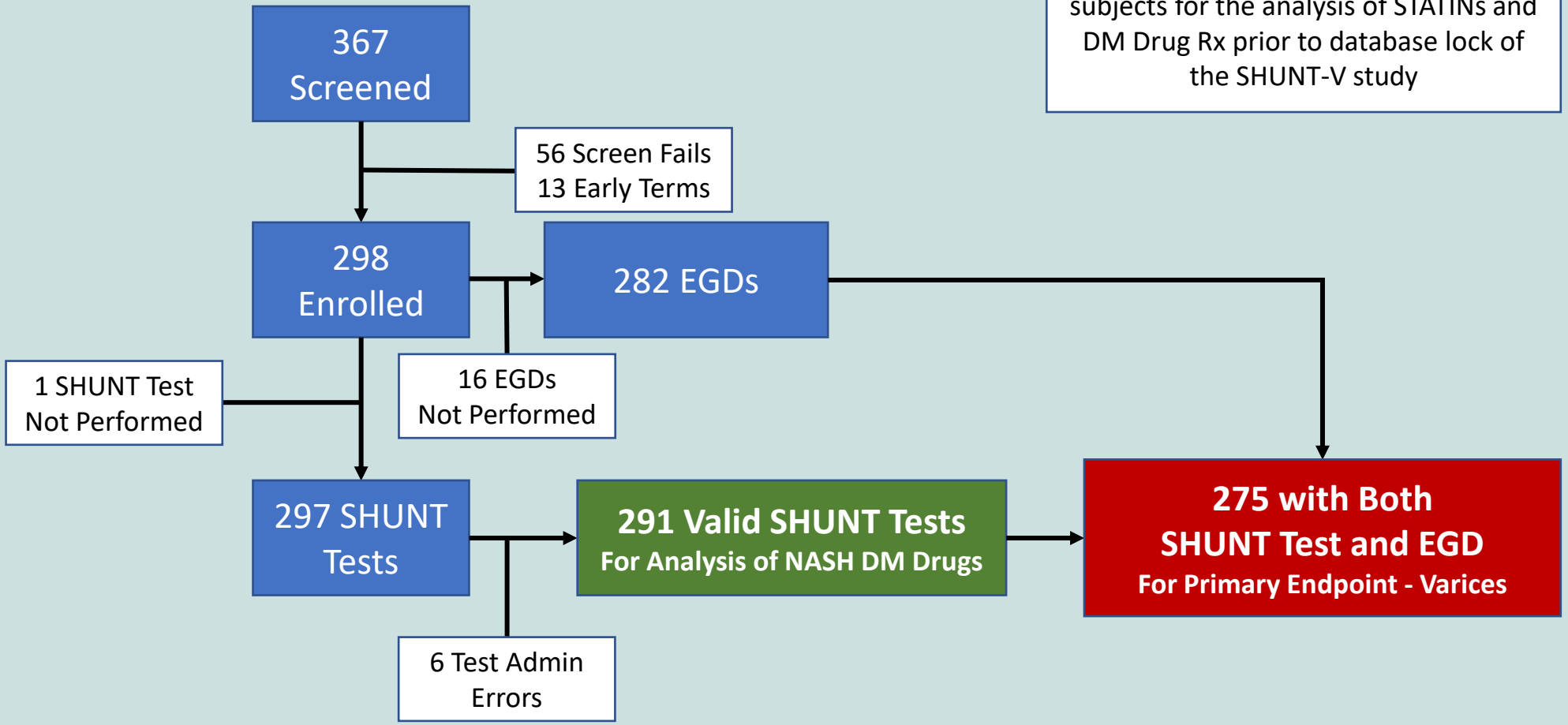
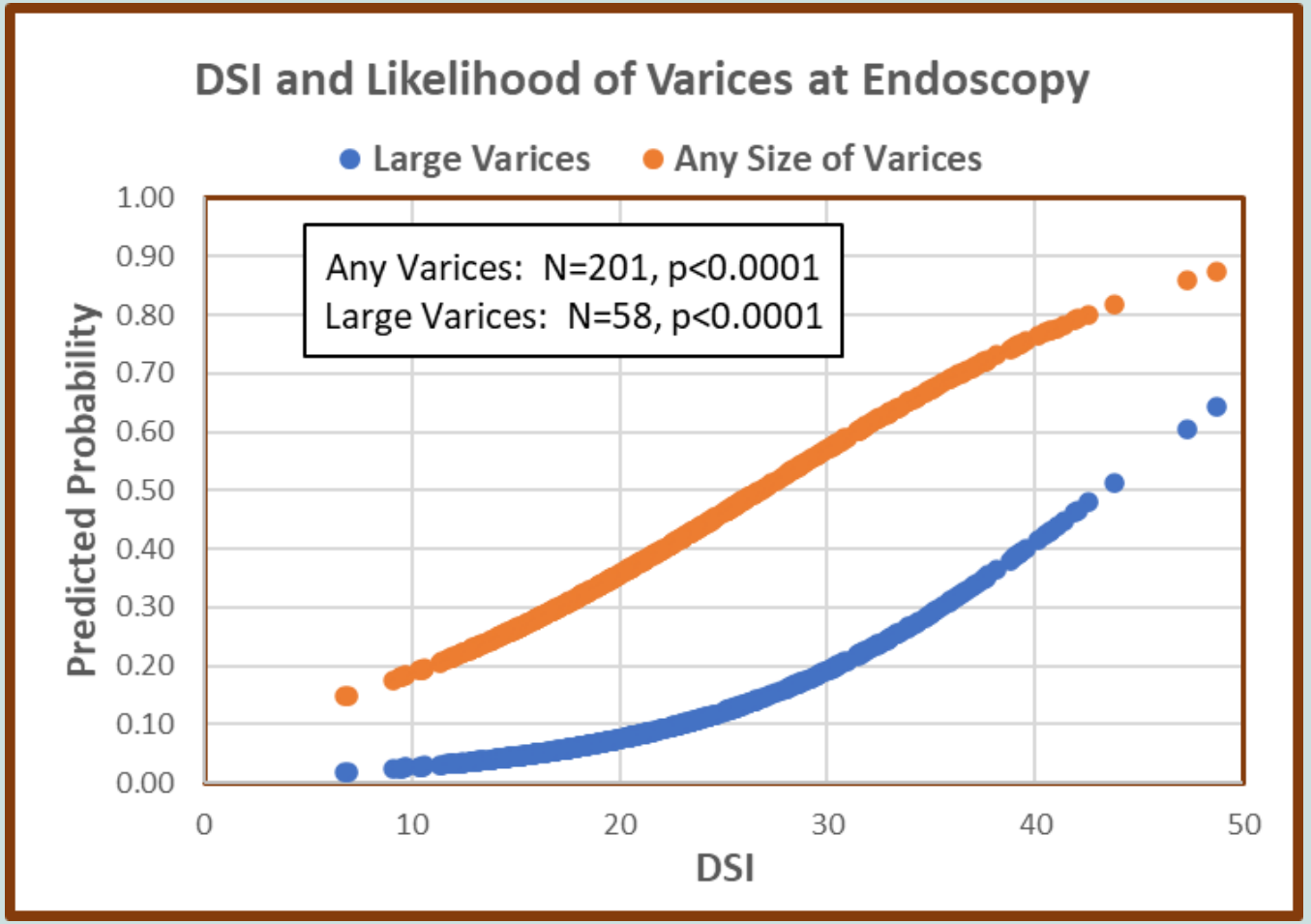
# SHUNT-V Study Flow Chart

For AASLD abstract/presentation:

Data were available for 270 of the 291 subjects for the analysis of STATINs and DM Drug Rx prior to database lock of the SHUNT-V study



# Probability of Finding Varices at EGD for a Given DSI




**There were highly significant relationships of finding any varices or large varices at EGD with DSI.**

# Conclusions and Key Takeaways

- The HepQuant DSI and SHUNT% predict the likelihood of finding esophageal varices, particularly large varices, at endoscopy across a wide spectrum of patient characteristics, disease etiologies, and severity of CLD.
- The HepQuant SHUNT Test may be useful in the decision to avoid or proceed with endoscopic screening or surveillance.

# *HepQuant Measures Treatment Effects*

# TREATMENT WITH HMG-CoA REDUCTASE INHIBITORS (STATINS) IS ASSOCIATED WITH PRESERVATION OF HEPATIC FUNCTION IN ADVANCED CHRONIC LIVER DISEASE (CLD): RESULTS FROM THE SHUNT-V STUDY

Dr. Robert S. Rahimi, Annette C. and Harold C. Simmons Transplant Institute, Baylor University Medical Center, Baylor Scott and White, Dallas, TX, Dr. Kathryn Jean Lucas, Diabetes and Endocrinology Consultants, North Carolina, USA, Dr. Ethan M Weinberg, Gastroenterology, Hospital of University of Pennsylvania, Dr. Zeid Kayali, Inland Empire Liver Foundation, Dr. Vishal K. Bhagat, Gastroenterology Consultants of Southwest Virginia, Dr. Andrew Paul Keaveny, MD, Transplant Center-Liver, Mayo Clinic Florida, Dr. Parvez S. Mantry, The Liver Institute, Methodist Dallas Medical Center, Dr. Alastair D. Smith, Syneos Health, John M Kittelson, Biostatistics and Informatics, Colorado School of Public Health, Steve M. Helmke, Hepquant, LLC, Dr. Gregory T Everson, Hepquant LLC and SHUNT-V Investigators Group

# In Multivariable Analysis the Use of STATINs or METFORMIN were Independently Associated with Lower SHUNT% and Lower DSI

| | Impact on SHUNT% | | Impact on DSI | |
|---|---|---|---|---|
| | Decline in SHUNT% | p | Decline in DSI | p |
| **Statin** | **-6.3%** | **0.0132** | **-3.3269** | **0.0025** |
| **Metformin** | **-5.9%** | **0.0475** | **-2.4337** | **0.0574** |
| **Diabetes Diagnosis** | -1.4% | 0.64 | -0.7239 | 0.5736 |
| **NASH Diagnosis** | -1.3% | 0.61 | -0.2246 | 0.8343 |

**The combined effect of the use of STATINs plus METFORMIN was**
**20% less portal-systemic shunting (lower SHUNT%) and 20% better function (lower DSI).**

# Key Takeaways

- The results support a positive effect of STATIN and Metformin therapy on severity of chronic liver disease – and, the need for an interventional study of STATINS and Metformin in CLD

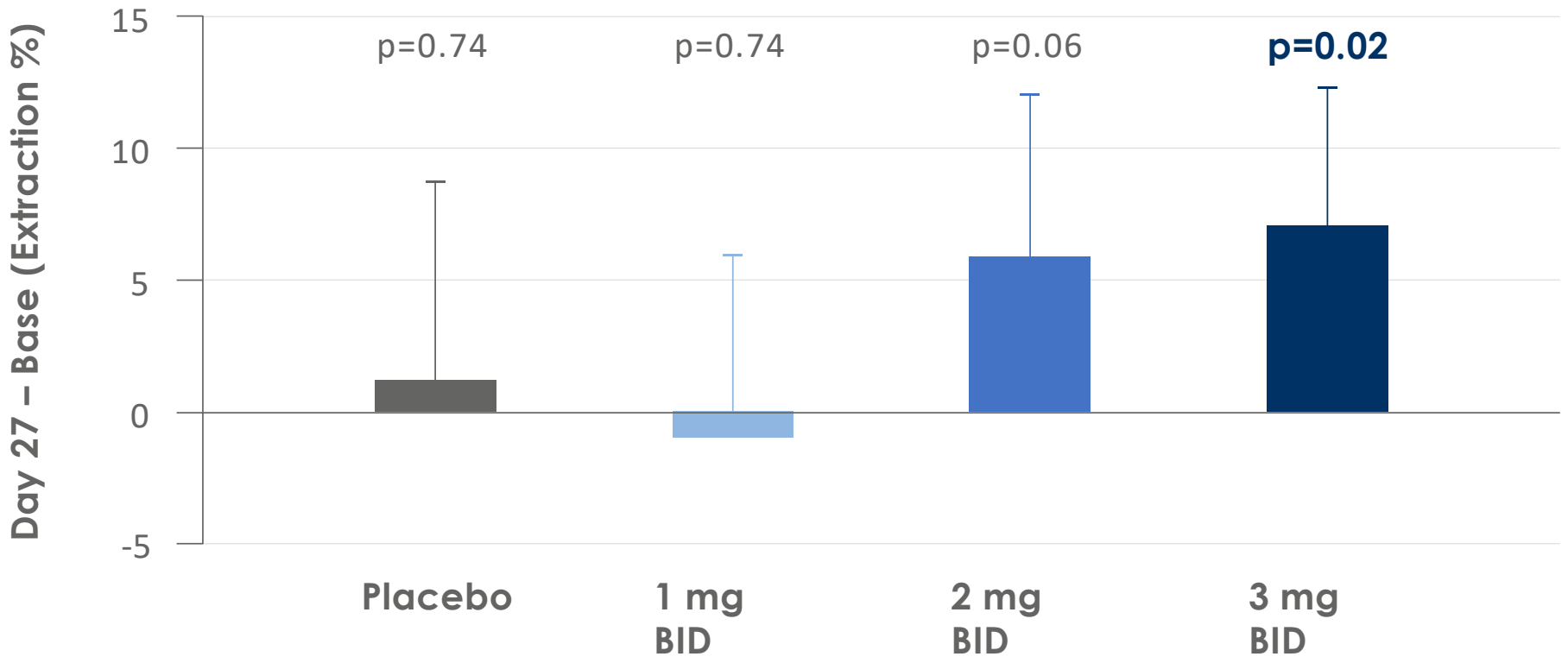
- The detection of the STATIN and Metformin effects by the dual cholate SHUNT test support further investigation of the use of this test in understanding effects of drug therapy of liver disease

# Analysis from a Phase 1b Drug Trial

# Boehringer Ingelheim BI 685509

# 28-day Dose-Dependent Improvement in Hepatic Extraction (1-SHUNT) with BI 685509, N=24

**Figure 2. Change in the first pass extraction % of d4-CA (1 – SHUNT %) from the portal inflow**




BID, twice daily; CA, cholate.

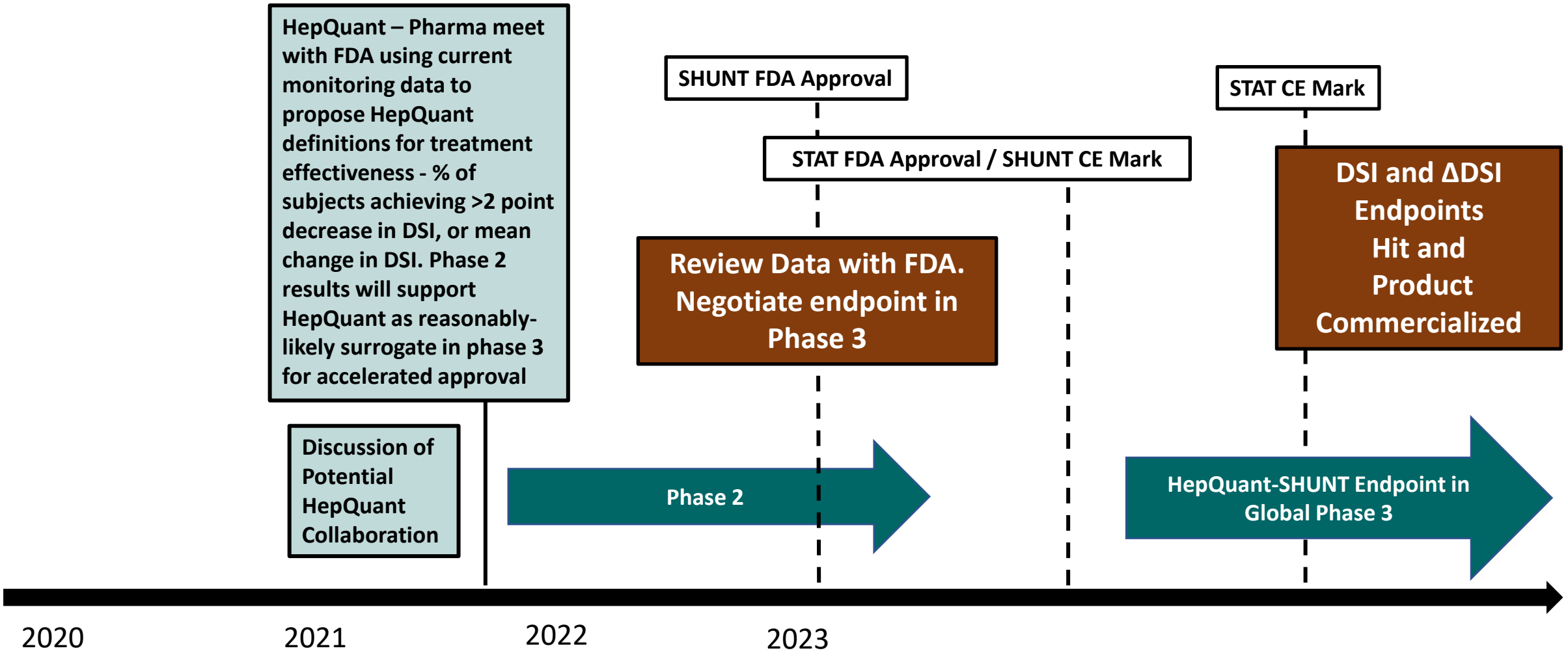
# Key Takeaways

- The results from this subgroup analysis indicate that BI 685509 improved hepatic perfusion in subjects with CP-A cirrhosis. In addition, first-pass extraction of d4-CA was increased with BI 685509 in a dose-dependent manner, with a significant increase observed with 3 mg BID (p=0.02)

- The HepQuant SHUNT test successfully defined differences in baseline disease severity between dose groups, detecting early treatment effects and defining dose response using small sample size

# Potential HepQuant Endpoint Collaboration

Milestones



# HepQuant for Hepatic Impairment Studies

# Predicting Drug Pharmacokinetics

| Compound | PK Measurement | Compartment (Pathways) |
|---|---|---|
| unconj-OCA | Clearance (h*ng $mL^{-1}$ $mg^{-1}$) | Cytosol (transport/BA conjugation) |
| Galactose | Clearance (mg $min^{-1}$ $kg^{-1}$) | Cytosol (transport/galactokinase) |
| Antipyrine | Clearance (mL $min^{-1}$) | Microsome (CYP 1A2, 2B6, 2C8, 2C9, 2C18, 3A4) |
| Caffeine | Elimination rate ($h^{-1}$) | Microsome (CYP 1A1, 1A2, 2A6, 2E1, 3A) |
| Lidocaine | Metabolite MEGX (ng $mL^{-1}$) | Microsome (CYP 3A4, 1A2) |
| $^{13}$C-Methionine | Metabolite $^{13}$C-$CO_2$ (score) | Mitochondria (oxidation) |

2021 AASLD/FDA DILI Conference Drug Induced Liver Injury: New Developments, Innovative Tools and Assessment Strategies in Patients with Underlying Liver Disease or Cancer
April 20 – 22, 2021 Virtual Conference

# Key Takeaways

- The improved resolution of drug PK by SHUNT test HepQuant DSI and SHUNT% over Child-Pugh scoring could improve selection of drug dose for patients with liver disease.

- Quantification of baseline hepatic function by HepQuant could reduce the risk of misclassification of the severity of liver disease in hepatic impairment studies.

# Regulatory and Commercialization Timelines

# HepQuant's Commercial Path – Value-added Milestones