# BASELINE PARAMETERS FOR NAFLD/NASH CLINICAL TRIALS

Recommendations from a Liver Forum Working Group

# Purpose and Action Steps

- To increase the uniformity of data collected across NAFLD/NASH clinical trials at screening and the baseline or pre-treatment initiation visit
- To make these data comparable and assist the regulatory agencies' efforts to determine efficacy and safety
- Experts from the stakeholder groups invited to engage in dialogue and deliberation to identify a standardized set of baseline parameters to recommend for inclusion in all clinical trials for NASH therapies

# Methodological Approach

- Working Group assessed the state of the science with regard to recently completed and current clinical trials for NAFL and NASH
  - Reviewed recent NAFL/NASH-related randomized clinical trials for general patterns of study entry criteria and baseline data
  - Made recommendations for broad categories of data to include in eligibility screenings and baseline assessments
- For each category, specific variables assessed for relevancy to
  - Mode of action (i.e., liver fibrosis versus liver inflammation)
  - Phase of trial
  - Essential versus ideal

# Broad Categories for Recommended Measurements

- Demographics
- Anthropometrics
- Diet & Activity
  - Physical activity
  - Diet/Nutrition
  - Alcohol consumption
  - Smoking/substance use
- Comorbidities
- Concomitant medication
- Laboratory tests
- Liver histology
- Potential non-invasive diagnostic tools
  - Imaging
  - Biomarkers
- Quality of life
  - Liver disease related QOL

# Demographics

- Gender
  - NAFLD more prevalent in males than females
  - differences in insulin resistance, visceral adiposity, life style, sex hormones, other factors
- Age
  - effects on liver -- > decreased volume, blood flow, and mitochondrial dysfunction
  - risk factor for
    - fibrosis progression
    - comorbid conditions such as cardiovascular disease
- Race and ethnicity

# Demographics

| | Proof of Concept, Phase 2a/ 2b | Phase 3 |
|---|---|---|
| **Age** | E | E |
| **Gender** | E | E |
| **Race** | I | E |
| **Ethnicity** | I | E |

Key: E = Essential, I = Ideal, C= Consider

# Diet and Activity
# Alcohol, Smoking and Substance Use

- SOC for activity and diet  is recommended to be implemented for NAFLD patients prior to enrollment in clinical trials
  - NCEP Step1 for non-diabetics and ADA diet for diabetics
- Consider the time period between the screening visit and baseline visit as a “run-in period” to stabilize diet and activity level
- Baseline level of physical activity can be readily obtained using the International Physical Activity Questionnaire (IPAQ)
- Audit C, a validated tool for alcohol consumption, could be augmented with biochemistry markers, such as PEth for confirmation

# Activity/Diet - Baseline Parameters Recommendations

| | Proof of Concept, Phase 2a/ 2b | Phase 3 |
|---|---|---|
| Diet Assessment* | E | E |
| Activity Assessment* | E | E |
| Alcohol consumption (AUDIT C) | E | E |
| Alcohol exposure (PEth) | C | C |
| Smoking habits | I | E |

Key: E = Essential; I = Ideal; C = Consider
PEth: PHOSPHATIDYLETHANOL
* Avoid major changes during the trial

# Anthropometrics

| | Proof of Concept, Phase 2a/ 2b | Phase 3 |
|---|---|---|
| Weight | E | E |
| BMI | E | E |
| Waist Circumference* | E | E |
| Blood Pressure | E | E |

Key: E = Essential

*Consider other circumferences

# Comorbidities and other liver diseases

- Essential to capture across all phases of trials and drug mechanisms
- Some of them should be considered as exclusion criteria

  - type 2 diabetes mellitus,
  - hypertension,
    - In some studies, HTN has been associated with fibrosis progression
  - hyperlipidemia,
  - cardiovascular disease,
  - obstructive sleep apnea,
  - Autoimmune disease,
  - Obesity and history of weight loss/bariatric surgery

# Comorbidities and other liver diseases

- Essential to capture across all phases of trials and drug mechanisms
- Some of them should be considered as exclusion criteria
  - Presence of other chronic liver disease (viral and autoimmune hepatitis, history of DILI, alcoholic liver disease, Wilson's disease, genetic hemochromatosis, history of biliary tract disease, genetic hemochromatosis, known or suspected HCC, Gilbert 's disease
  - Depression

- Specific comorbidities important to consider with regard to drug clearance, tolerability, and safety.
- Renal function effects on drug clearance and metabolism.

# Concomitant Medications

- Essential to capture regardless of phase of trial or medication mechanism
- Some medications may have anti-NASH effects directly or indirectly by targeting elements of the metabolic syndrome.

- Antidiabetics
  - Glitazones
  - SGLT2 inhibitors
  - GLP1 agonists.DPP4 inhibitors
  - Metformin/ Sulfonylureas
  - Insulin
- Vitamin E
- PUFA/W3FA
- Ursodeoxycholic acid
- Statins and fibrates
- Antihypertensive drugs

# Laboratory Tests: Metabolic Profile, Liver Panel, Hematology and other Blood Chemistries

Essential regardless of phase of trial or drug mechanism

- to ensure suitability for inclusion
- To track dynamic changes due to activity of the study drug or metabolites
- To detect effect modifiers in analysis of endpoints
- potential influence on drug clearance

- Liver Panel
- Glucose and Lipid profile
- Hematology
- Other chemistries

# Metabolic Panel: glucose and lipid profile

| | Proof of Concept, Phase 2a/ 2b | Phase 3 |
|---|---|---|
| Fasting Glucose | E | E |
| Fasting Insulin | E | E |
| Hemoglobin A1c | E | E |
| HOMA | NR | NR |
| Glucose Clamp | NR | NR |
| Total Cholesterol and fractions (LDLc and HDLc) | E | E |
| Triglycerides | E | E |

Key: E = Essential, NR = Not Recommended

# Laboratory Tests: Liver Panel, Hematology and other Blood Chemistries

| | Proof of Concept, Phase 2a/ 2b | Phase 3 |
|---|---|---|
| ALT | E | E |
| AST | E | E |
| ALP | E | E |
| Total Bilirubin | E | E |
| GGT | E | E |
| Albumin | E | E |
| Prothrombin Time/ INR | E | E |
| Hemoglobin | E | E |
| Hematocrit | E | E |
| Platelets | E | E |
| BUN | E | E |
| Creatinine | E | E |

# Liver Histology

- NASH Histological confirmation
- NASH CRN scoring system : NAS and fibrosis stage
  - good inter-rater reproducibility
- SAF score : a semi-quantitative score of steatosis, activity (lobular inflammation and hepatocellular ballooning ) and fibrosis
  - good inter-rater reproducibility
  - separates steatosis from necro-inflammation

| | Proof of Concept, Phase 2a/ 2b | Phase 3 |
|---|---|---|
| Liver Histology | I/E | E |

Key: E = Essential, I = Ideal

# Specialized Biomarkers

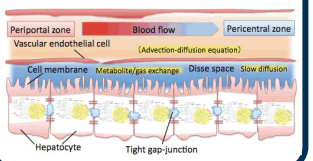
- Urgent need of validated non-invasive biomarker
- Final recommendations for standardized baseline assessment need further validation
- Opportunity to accelerate investigation and validate use
- To confirm mechanisms of action and/or PK/PD purposes

# Specialized Biomarkers

**Oxidative Stress and Apoptosis**

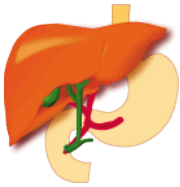
Malondialdehyde, TBARS, Oxidized LDL
Cytokeratin-18 (CK-18) fragments
Ferritin

**Hepatocyte Function**




Alanine aminotransferase (ALT)
Aspartate aminotransferase (AST)
Alkaline phosphatase (ALP)
Gamma-glutamyl-transpeptidase (GGT)
Acute phase reactants: α1-antitrypsin, ceruloplasmin, haptoglobin, GlycA
Albumin, Lipid panel, Platelet count
Measures of insulin resistance

**Fibrosis**



Fibronectin
Hyaluronic acid (HA)
Type IV collagen S
Procollagen type III N-terminal Peptide (PIIINP)
Tissue inhibitor of metalloproteinase 1 (TIMP-1)
Small VLDL particle number

**Inflammation**

Cytokines: TNF-α, IL-1β, IL-6, IL-8, IFNγ, TGFβ
Adipokines: Adiponectin, Leptin, Resistin
Acute phase reactants: hsCRP, GlycA
Chemoattractants: MCP-1
Ferritin, Soluble CD14
VLDL size and small LDL particle number

**Multiple panels**

**1.** Adapted from Fitzpatrick et al., Noninvasive biomarkers in NAFLD World, *J Gastroenterol* 2014;20(31):10851-10863.
**2.** Armutcu et al., Adv in Clin Chem 2013;61:67-125.

# Genetic markers

- Genetic polymorphisms associated with liver disease severity and/or cardiovascular risk in NAFLD

- PNPLA3, TM6SF2 and GCKR
  - variants have specific ethnic distributions
  - PNPLA3 accounts for up to 72% of inter-ethnic variation in hepatic triglyceride content in the Dallas Heart Study

**Essential** storing DNA for future genetic (including genome wide) analyses; obtain appropriate informed consent
**Essential** PNPLA3 in PhIII

# Imaging

## Liver Fat

► Ultrasound low sensitivity ( <20%-30% of liver fat)

► Magnetic resonance-based

- MRI-PDFF / MRS
- Multiparametric MRI

## Liver Stiffness

► Vibration-controlled transient elastography (VCTE)

► US elastography (pSWE or ARFI and 2D-SWD)

► Magnetic resonance elastography

MRI-PDFF: Magnetic resonance imagins-protocol density fat fraction
MRS: Magnetic resonance spectroscopy
pSWE: point shear wave elastography-ARFI: acoustic radiation force impulse imaging
2D-SWD 2D-shear wave elastography

# Imaging

| | POC/Ph2a/2b | Ph3/clinical outcomes |
|---|---|---|
| MRI/MRS | E in POC trials when changes in liver fat is the primary endpoint | NA |
| Multiparametric MRI | I (to enrich a population in a POC when biopsy confirmation is not available) | Need further validation |
| VCE/Fibroscan | C/ I (to enrich a population in a POC when biopsy confirmation is not available) | C/ I (early identification of patients progressing to cirrhosis) |

# Health Related Quality Of Life

- NAFLD-specific HRQOL measures lacking
- Currently available: short form 36 (SF-36) and the chronic liver disease questionnaire (CLDQ)
- Ideally would want tool that evaluates HRQOL distinct from features of metabolic syndrome
- In absence of validated NAFLD HRQOL too, current recommend SF-36 (used by NASH CRN in practice) and CLDQ (validated for chronic liver disease)

| | Proof of Concept, Phase 2a/2b | Phase 3 |
|---|---|---|
| **SF-36** | I /C in Ph2b | E |
| **CLDQ** | I/C in Ph2b | E |