Biomarkers of Frailty: A Collaboration between CLSA, CFN, Metabolon & MIRA

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What is the Canadian Longitudinal Study on Aging (CLSA)?

A research platform – Infrastructure to enable state-of-the-art, interdisciplinary population-based *research* and *evidenced-based* decision-making that will lead to better health and quality of life for Canadians.

CLSA Research Platform

50,000 women and men aged 45 - 85 at baseline

Target: 20,000
Actual: 21,241
Randomly selected within provinces

Target: 30,000
Actual: 30,097
Randomly selected
within 25-50 km of 11 sites

Questionnaire
• By telephone (CATI)

Questionnaire
• In person, in home (CAPI)

2010 - 2015   2015   2018

Clinical/physical tests
Blood, urine
•   @ Data Collection Site

Participants aged 45 to 85 at baseline (51,338)

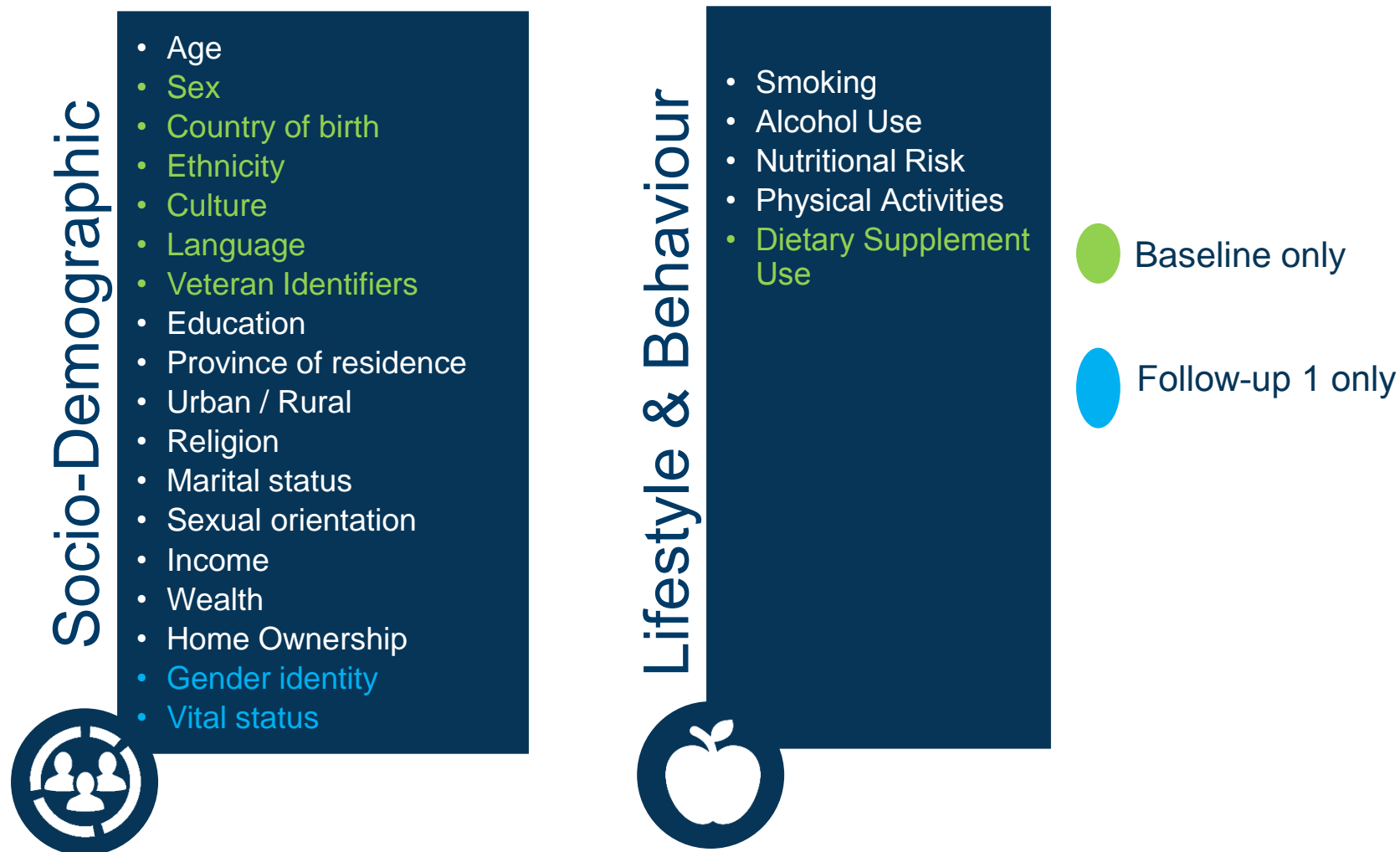20 Years

Baseline   FU-1   FU-2   FU-3   FU-4   FU-5   FU-6

Active follow-up every 3 years

Baseline & Follow-up 1 Questionnaires
Tracking & Comprehensive

Socio-Demographic

- Age
- Sex
- Country of birth
- Ethnicity
- Culture
- Language
- Veteran Identifiers
- Education
- Province of residence
- Urban / Rural
- Religion
- Marital status
- Sexual orientation
- Income
- Wealth
- Home Ownership
- Gender identity
- Vital status

Lifestyle & Behaviour

- Smoking
- Alcohol Use
- Nutritional Risk
- Physical Activities
- Dietary Supplement Use

● Baseline only

● Follow-up 1 only

Current Data Availability: www.clsa-elcv.ca/data-availability

Baseline & Follow-up 1 Questionnaires
Tracking & Comprehensive

🟢 Baseline only

🔵 Follow-up 1 only

Physical Health

- General Health
- Women's Health
- Vision
- Hearing
- Oral Health
- ADL/IADL
- Pain and Discomfort
- Health Care Utilization
- Injuries
- Falls
- Falls due to Consumer Products
- Preventative Health Behaviours
- Hearing Handicap Inventory for the Elderly
- Unmet Health Care Needs

- Self-Reported Chronic Conditions & Disease Symptoms:
  - Diabetes
  - Stroke/Cerebrovascular
  - Traumatic Brain Injury
  - Hypo & Hyperthyroidism
  - Hypertension
  - Ischemic Heart Disease
  - Osteoarthritis of the Hand
  - Osteoarthritis of the Hip
  - Osteoarthritis of the Knee
  - Musculoskeletal
  - Osteoporosis
  - Neurological
  - Neuropsychiatric
  - Asthma
  - COPD
  - Gastrointestinal
  - Cancer
  - Epilepsy

Current Data Availability: www.clsa-elcv.ca/data-availability

Baseline & Follow-up 1 Questionnaires
Tracking & Comprehensive

Psychological Health

- Depression
- General mental health (self-reported)
- Satisfaction with Life
- Post-traumatic Stress Disorder
- Cognitive Battery:
  - Rey I/Rey II
  - Animal Fluency Test
  - Mental Alternation Test
  - Meta Memory
  - Subjective Cognitive Decline
- Loneliness Scale
- Childhood Maltreatment Elder Abuse

Labour Force

- Retirement Status
- Pre-Retirement Labour Force Participation
- Pre-Retirement Labour Force Participation – open text
- Labour Force
- Labour Force – open text question
- Retirement Planning*
- Work Limitations Questionnaire

*Abbreviated in Comprehensive

Social Health

- Social Networks
- Social Support – Availability
- Social Participation
- Care Receiving 1/ Formal Care
- Care Receiving 2 / Informal Care
- Care Giving
- Social Inequality
- Online Social Networking
- Transportation, Mobility, Migration
- Built Environments
- Social Cohesion

Baseline only
Follow-up 1 only

Baseline & Follow-up 1 Questionnaires
Tracking & Comprehensive

Comprehensive Only

- Short Diet Questionnaire
- Disease Algorithms & Disease Symptoms
- Medications
- Sleep & Snoring
- Life Space Index
- Psychological Distress
- Personality Traits

Tracking Only

- Self-reported Height
- Self-reported Weight
- Functional Status
- Dietary Supplement Use
- Medication Use

Current Data Availability: www.clsa-elcv.ca/data-availability

Physical Assessments
Comprehensive Only

Biospecimens

- Blood
  - Serum
  - Plasma
  - Buffy Coat
  - Whole blood
  - Cells
  - DNA
  - PBMC
- Urine
  - Random

Psychological Measures

- Full Cognitive Battery*:
  - Miami Prospective Memory Test (event- & time-based)
  - Stroop – Victoria Version
  - Controlled Oral Word Association Test
  - Choice Reaction Time
  - Meta Memory
  - Subjective Cognitive decline

*Includes Rey, Mental Alternation & Animal Fluency Tests. Meta Memory, Subjective Cognitive Decline

Physical Measures

- Measured Height/Weight
- Body Mass Index
- Hip & Waist Circumference
- Blood pressure & Pulse Rate
- Body Composition
- Timed Get Up and Go
- Standing balance
- 4-metre walk
- Chair rise
- Grip strength
- Visual Acuity
- Fundus Scans (Diabetic Retinopathy & micro vessel disease)
- Tonometry
- Hearing
- Spirometry
- Carotid Intima Media Thickness
- Carotid Pulse Wave Velocity
- ECG, Aortic Calcification Bone Density by DXA Body Composition by DXA

Current Data Availability: www.clsa-elcv.ca/data-availability

Upcoming Follow-up 1 Releases

- Cognition
- Alphanumeric physical assessment data:
  - Spirometry
  - Hearing
  - Visual acuity, Tonometry, Fundus Scans
  - Electrocardiogram
  - Carotid-intima Media Thickness
- Hematology

Available Fall 2019

Upcoming Releases

- Chemistry FU-1
- Epigenetics (Baseline)
- Metabolomics (Baseline)
- Medications Baseline & FU-1

Anticipated 2019-2020

Follow-up 2 New Data

- Positive Mental Health
- Generalized Anxiety Disorder
- Digit Triplet Test for Hearing
- Sitting Height measurement
- Weight Perception
- Resiliency
- Intimate Partner Violence
- Post-traumatic Stress Disorder (re-introduced)

Data collection launched Summer 2018

Ends 2021

CLSA Biomarkers for Frailty Research

THE JOURNAL OF FRAILTY&AGING

PROCEEDINGS OF THE CANADIAN FRAILTY NETWORK WORKSHOP: IDENTIFYING BIOMARKERS OF FRAILTY TO SUPPORT FRAILTY RISK ASSESSMENT, DIAGNOSIS AND PROGNOSIS. TORONTO, JANUARY 15, 2018
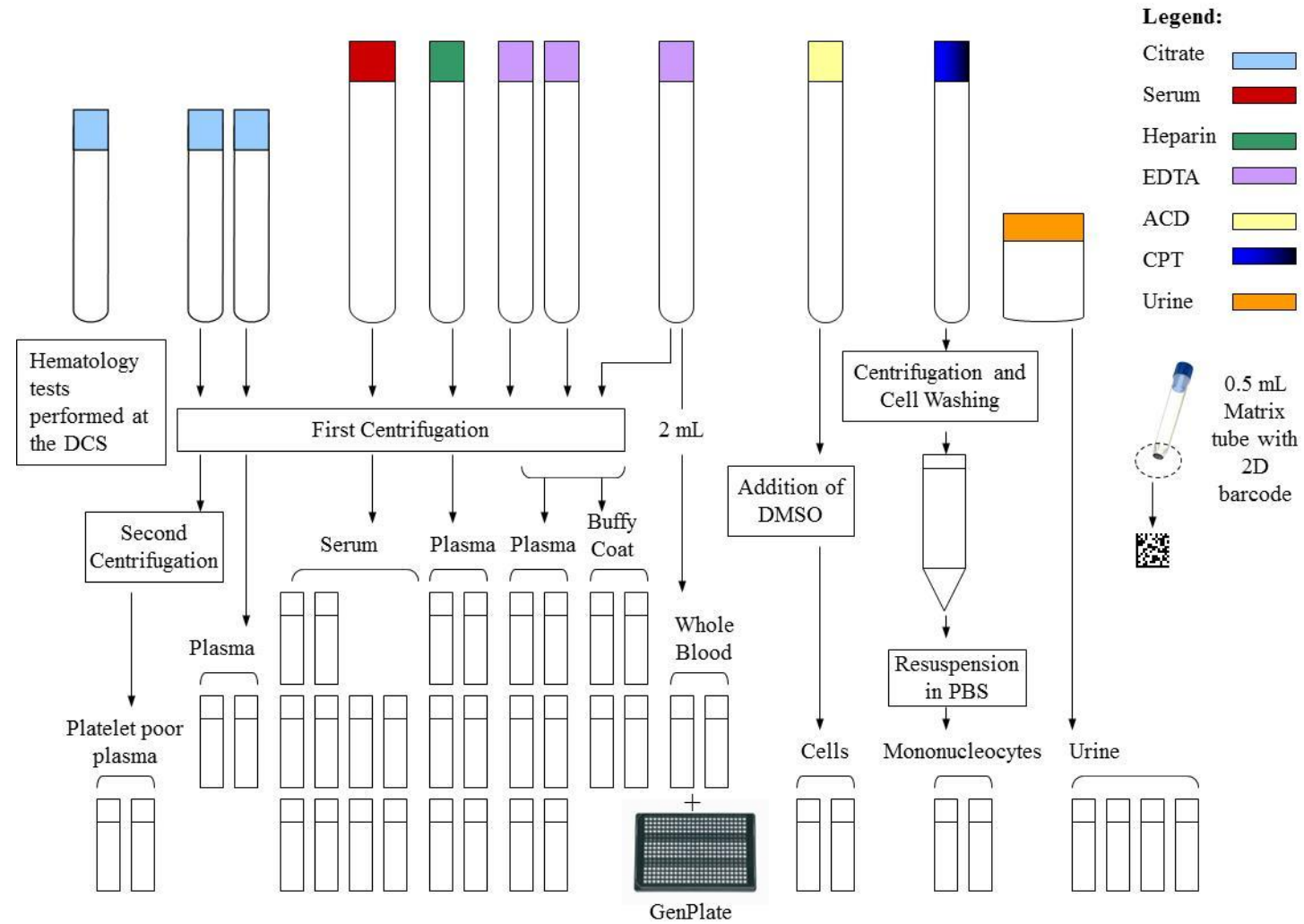
J. MUSCEDERE, P.M. KIM, J. AFILALO, C. BALION, V.E. BARACOS, D. BOWDISH, M. CESARI,J.D. ERUSALIMSKY, T. FÜLÖP, G. HECKMAN, S.E. HOWLETT, R.G. KHADAROO, J.L. KIRKLAND, L. RODRIGUEZ MAÑAS, E. MARZETTI, G. PARÉ, P. RAINA,
K. ROCKWOOD, A. SINCLAIR, C. SKAPPAK, C. VERSCHOOR, S. WALTER
FOR THE CANADIAN FRAILTY NETWORK

Principles to Guide Future CFN biomarker initiatives

- 1) Biomarkers should reflect a pathophysiological pathway or mechanism that is fundamental to frailty onset, development/progression and severity. Conceptually there maybe two categories of biomarkers:
  - i) Biomarkers that are linked with frailty but are not causal to the pathophysiology of frailty. These would not be actionable.
  - ii) Biomarkers that are a component of the pathophysiology of frailty and have a causal role. These would be actionable such that the modulation of the biomarker could directly affect the onset or severity of frailty and/or progression of frailty.
- 2) The utility of biomarkers can be classified into two different types:
  - i) Biomarkers to increase the utility of (or support) existing clinical frailty measures (e.g., FI).
  - ii) Biomarkers to be used independently of clinical frailty measures.
- 3) Biomarkers should be able to be embedded in clinical assessments and tools, but more research on how to best achieve this is needed. Concomitant use of both a clinical frailty assessment instrument and biomarkers is likely to be the optimal method to bring about personalized frailty assessment and individualized care plans.

- 4) Biomarkers chosen for a clinical tool should be evaluated on their ability to accomplish the ultimate clinical purpose. For instance, biomarkers used for diagnosis may be different from those used for risk assessment, which may differ from those used for prognosis
- 5) Different care settings are likely to require different biomarkers due to variation in prevalence of both frailty and biomarkers in different populations.
- 6) Any candidate biomarker should be validated in different populations, care settings and environments.
- 7) An ideal frailty biomarker would be able to measure the effectiveness of an intervention.
- 8) Practical considerations related to ease of measurement (i.e., special instruments and/or expertise required) and ease of securing biological samples (e.g., tissue biopsy vs blood sample collection) should be considered when selecting frailty biomarkers.

CLSA biospecimens

Legend:
- Citrate
- Serum
- Heparin
- EDTA
- ACD
- CPT
- Urine

Hematology tests performed at the DCS

First Centrifugation

2 mL

Centrifugation and Cell Washing

Second Centrifugation

Serum

Plasma

Plasma

Buffy Coat

Addition of DMSO

Resuspension in PBS

Plasma

Platelet poor plasma

Whole Blood

GenPlate

Cells

Mononucleocytes

Urine

0.5 mL Matrix tube with 2D barcode

Core Biomarkers in the CLSA

	Category	N	Biomarkers		
Available from Baseline	**HEMATOLOGY** Data Collection Sites (DCS) Analysis repeated every 3 years)	25,427	• Erythrocytes • Granulocytes • Hematocrit • Hemoglobin	• Lymphocytes • Platelets • MCV • MCH	• MCHC • MPV • RBC • RDW
	CHEMISTRY Calgary Laboratory Services (CLS) (Analysis repeated every 3 years)	27,012	Albumin • Alanine aminotransferase (ALT) • C-reactive protein (CRP) • Creatinine • Cholesterol • Ferritin • Free T4 • Hemoglobin A1c (n = 26,961) • HDL • LDL Non-HDL	Thyroid-stimulating hormone (TSH) • Triglycerides • 25-Hydroxyvitamin D • eGFR • TNF-Alpha (n=10,000) (FU-1) • IL6 (n=10,000) (FU-1) • NT-ProBNP (FU-1) • hsTroponin • Electrolytes (n=10,000)	
	GENETICS Genetic and Epigenetic Centre (GEC)	26,871*	• Genotypes (Affymetrix Axiom array, 794k SNPs) • Imputation (Haplotype Reference Consortium release 1.1, 39.2M SNPs)		
Available in 2019	**EPIGENETICS** Epigenetic Centre (EC) (Repeated every 3 years)	1,488	• DNA methylation • DNA extracted from PBMCs • 850K Infinium MethylationEPIC BeadChip (Illumina)		
	Metabolomics Metabolon	10,000	• METABOLON Platform • ~1,300 metabolites		

Metabolon's HD4 Global Metabolomics Profiling
Screens >5000 Named and >7000 Unnamed Biochemicals And Reports Back All Detected
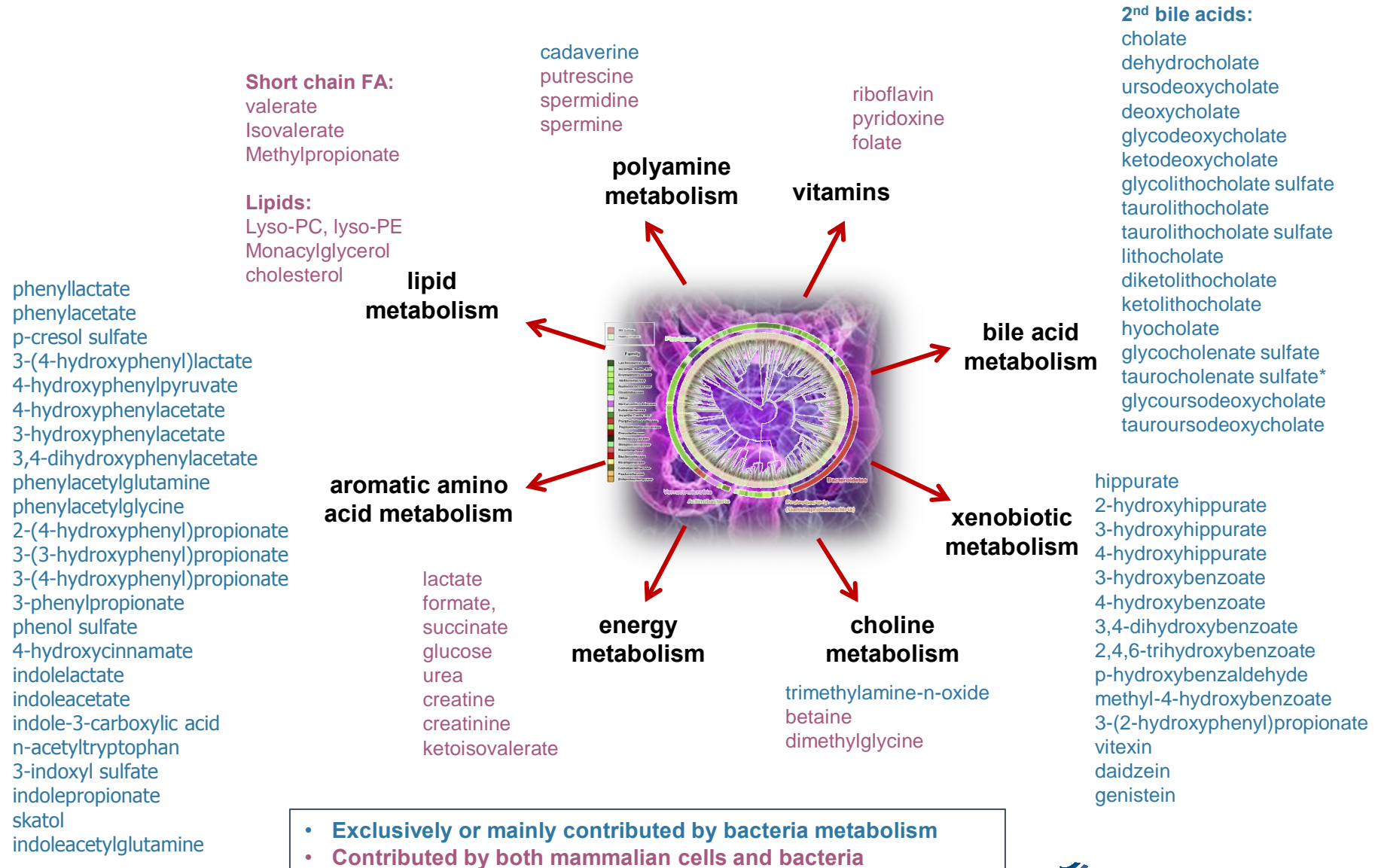
Metabolite Coverage

The DiscoveryHD4 platform provides the industry's broadest class coverage from a single sample.

Around 1,000 metabolites across diverse classes can be measured* from 100 µL of plasma/serum, 50-100 mg of tissue, or a 50-100 µL cell pellet with approximately 5% CVs.

* The metabolites detected from the above classes can vary based on the type of sample and the abundance levels in those samples.

Amino Acid Metabolism	Cofactor & Vitamin Metabolism	Nucleotide Metabolism	Microbiome Metabolism
Amino Acid catabolism	Ascorbate metabolism	Degradation of nucleotides	2° Bile acids
Bioactive intermediates & trace amines	CoA metabolism	Deoxyribonucleotides	Aromatic amino acids
Glutathione metabolism	FAD metabolism	DNA damage	Energy
Inflammatory mediators	Folate metabolism	FAD metabolism	Choline/carnitine
Microbiome metabolism	NAD/NADP metabolism	Modified nucleotides	Xenobiotics
Polyamines/ornithine metabolism	PLP metabolism	Nucleotide Coenzymes	Fatty acids/short chain medium chain
Urea Cycle	SAM metabolism	Purine and pyrimidine *de novo* synthesis	Vitamins
	Many other cofactors and vitamins (tocopherol, B12, Biotin)	Purine and pyrimidine salvage synthesis	Polyamines
		Ribose metabolism	

Carbohydrate Metabolism	Energy Metabolism	Lipid Metabolism	Novel Metabolites
Gluconeogenesis	Acyl-carnitines	Bile acids	Novel drug metabolites
Glucose metabolism	Beta-oxidation	Bioactive lipids	Novel xenobiotics
Glycogen metabolism	Creatine metabolism	Cholesterol	Novel microbiota metabolites
Glycosylation pathways	FAD metabolism	Fatty acids	Novel by-products of non-canonical host metabolism
Metabolism of other carbon sources	Glycolysis	Sphingosine	
Metabolism of sugars (fructose, galactose)	Mitochondrial function	Inflammatory mediators	
Polyol metabolism	Pentose phosphate pathway	Lysolipids	
Pyruvate metabolism		Sterols	
		Oxidized lipids (COX, LOX)	

METABOLON®

About 200 molecules are derived from bacterial metabolism

2nd bile acids:
cholate
dehydrocholate
ursodeoxycholate
deoxycholate
glycodeoxycholate
ketodeoxycholate
glycolithocholate sulfate
taurolithocholate
taurolithocholate sulfate
lithocholate
diketolithocholate
ketolithocholate
hyocholate
glycocholenate sulfate
taurocholenate sulfate*
glycoursodeoxycholate
tauroursodeoxycholate

Short chain FA:
valerate
Isovalerate
Methylpropionate

Lipids:
Lyso-PC, lyso-PE
Monacylglycerol
cholesterol

cadaverine
putrescine
spermidine
spermine

riboflavin
pyridoxine
folate

polyamine metabolism

vitamins

lipid metabolism

bile acid metabolism

phenyllactate
phenylacetate
p-cresol sulfate
3-(4-hydroxyphenyl)lactate
4-hydroxyphenylpyruvate
4-hydroxyphenylacetate
3-hydroxyphenylacetate
3,4-dihydroxyphenylacetate
phenylacetylglutamine
phenylacetylglycine
2-(4-hydroxyphenyl)propionate
3-(3-hydroxyphenyl)propionate
3-(4-hydroxyphenyl)propionate
3-phenylpropionate
phenol sulfate
4-hydroxycinnamate
indolelactate
indoleacetate
indole-3-carboxylic acid
n-acetyltryptophan
3-indoxyl sulfate
indolepropionate
skatol
indoleacetylglutamine

aromatic amino acid metabolism

lactate
formate,
succinate
glucose
urea
creatine
creatinine
ketoisovalerate

energy metabolism

choline metabolism

trimethylamine-n-oxide
betaine
dimethylglycine

xenobiotic metabolism

hippurate
2-hydroxyhippurate
3-hydroxyhippurate
4-hydroxyhippurate
3-hydroxybenzoate
4-hydroxybenzoate
3,4-dihydroxybenzoate
2,4,6-trihydroxybenzoate
p-hydroxybenzaldehyde
methyl-4-hydroxybenzoate
3-(2-hydroxyphenyl)propionate
vitexin
daidzein
genistein

- **Exclusively or mainly contributed by bacteria metabolism**
- **Contributed by both mammalian cells and bacteria**

METABOLON®

The Metabolome

The metabolome is tightly connected with other "omes."The metabolome interacts and reflects the activity of the genome, transcriptome, and proteome.
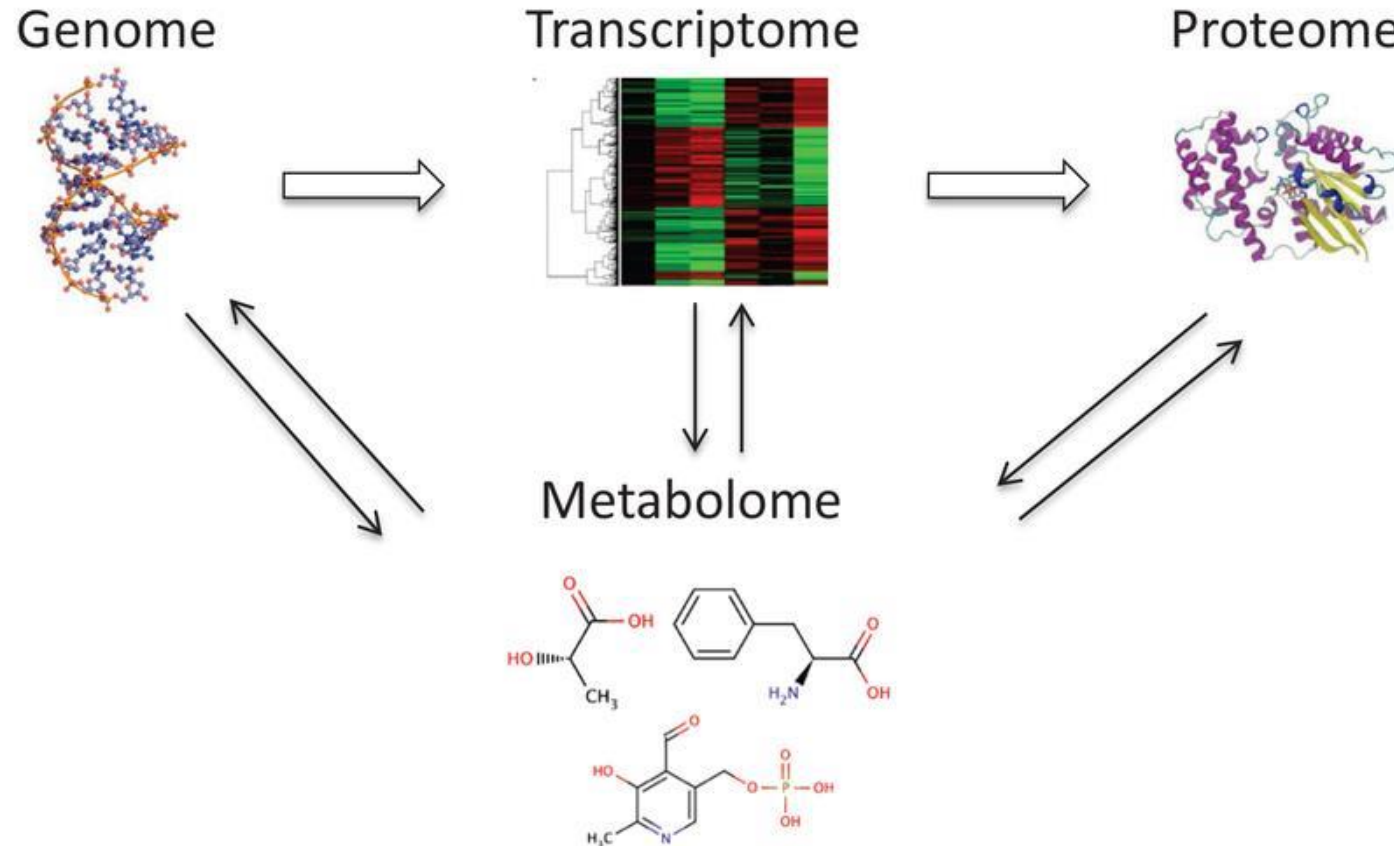

The end result of all biological & environmental interactions is the definitive way to fully understand the phenotype

METABOLON®

NEW OMICS TECHNOLOGIES IN THE CLSA

- Proteomics (O-LINK Multi-Plex Platform)
  - Inflammation panel (1500 CLSA Participants)
    - Available in Fall of 2020
- Whole Genome Sequencing on 500 CLSA participants over two time periods (Pilot)
  - Genetic Instability (Fall of 2020)
- Whole Exome Sequencing discussions in progress
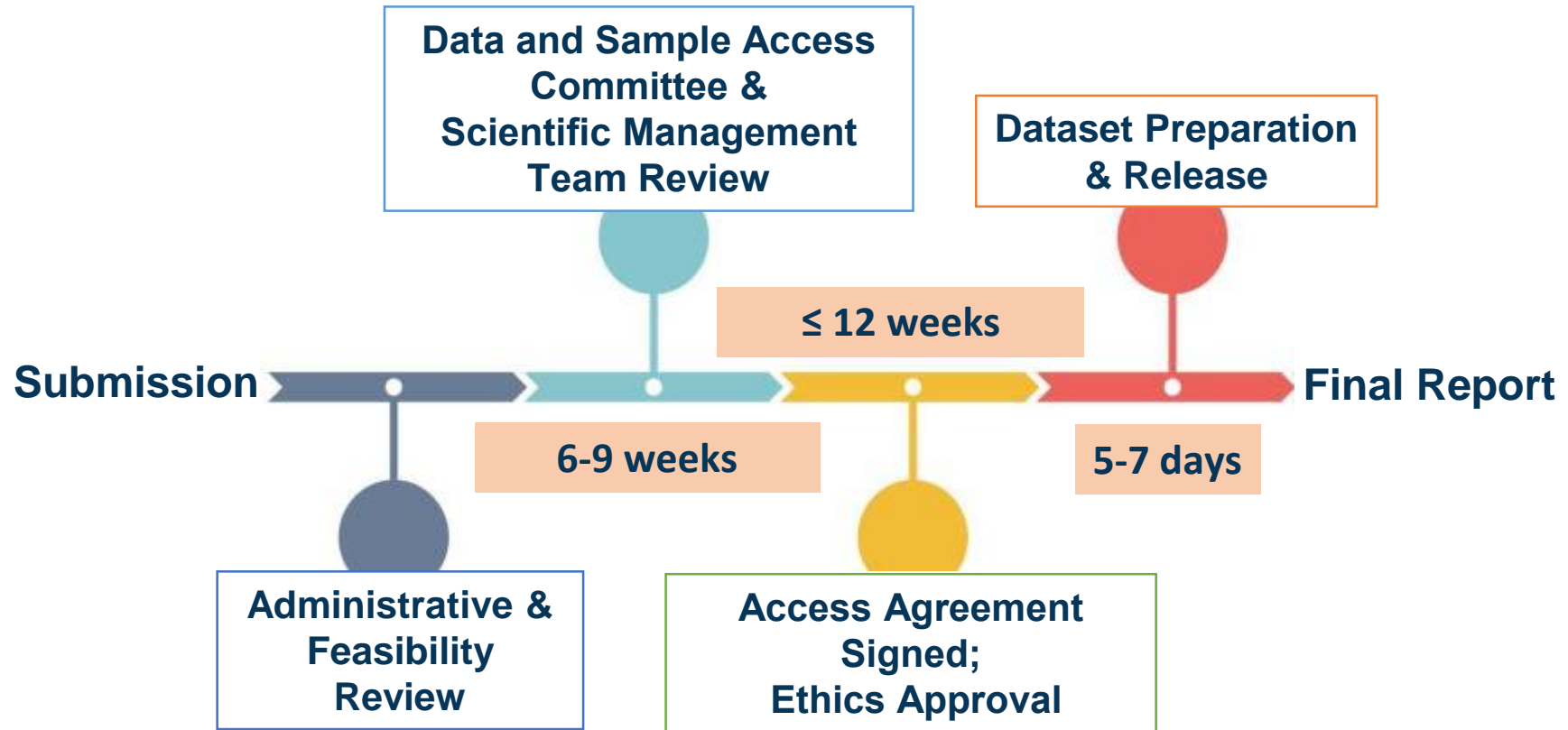
Opportunity for Research on Frailty
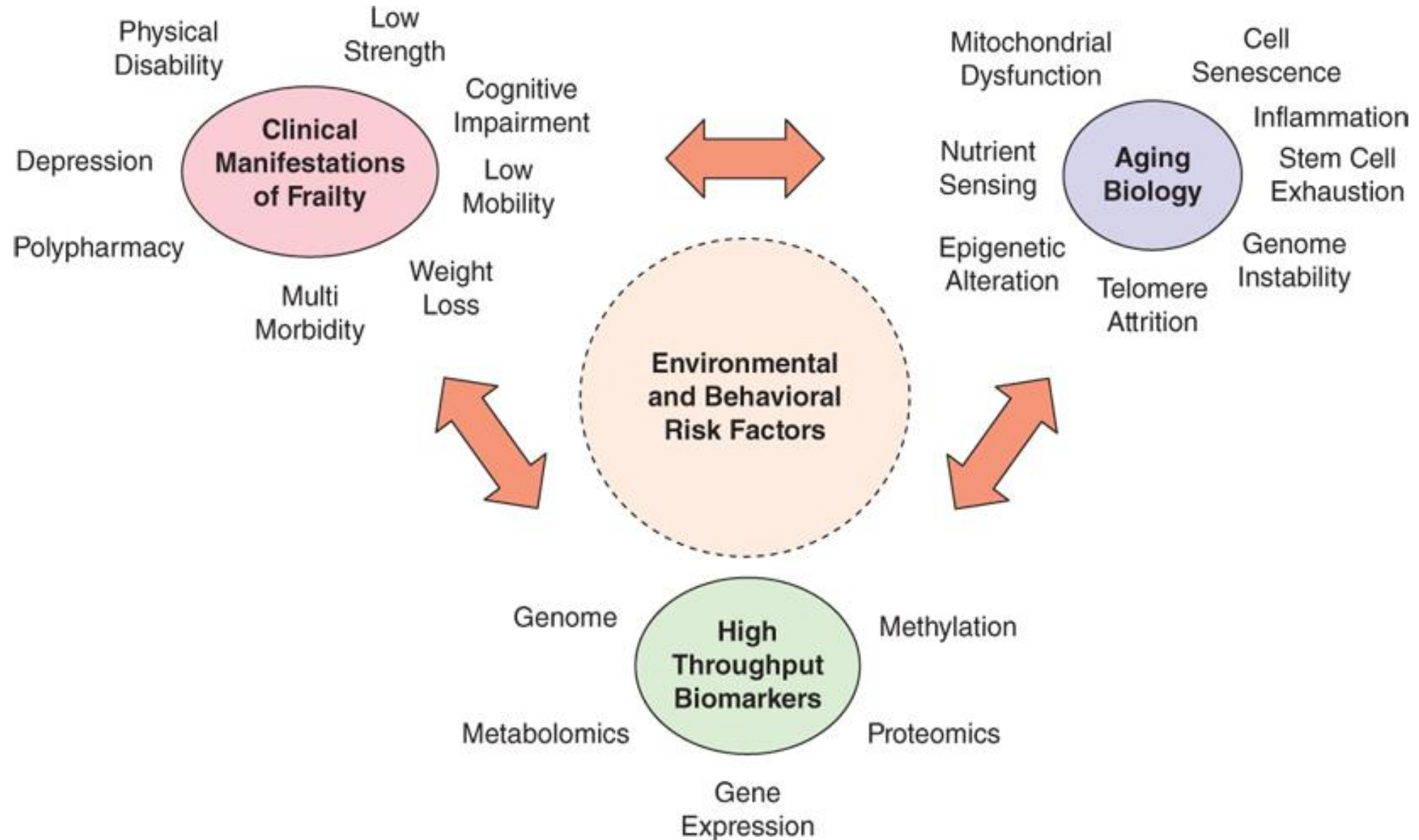
Applying for Data Access

- **Magnolia**, a new web-based data access application system
- User account requests: **access@clsa-elcv.ca**
- 2-3 working days to receive login information

Data Access Fees

- **Partial Cost Recovery Model**
  - **Alphanumeric data**
    - CAD $3,000 for researchers based in Canada
    - CAD $5,000 for researchers based at institutions outside of Canada
    - **Graduate students** using data solely for thesis research & **Postdoctoral fellows** using data solely for the postdoctoral project are eligible for a **fee waiver (once as postdoc)**
    - Trainees must be enrolled at a Canadian institution or be supported by Canadian funds if working outside Canada
  - **Images & Complex data**
    - Additional fees of CAD $1,000 apply for access to image files, raw data and datasets that require more complex customization

Contact:
Data inquiries: access@clsa-elcv.ca
General inquiries: info@clsa-elcv.ca

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www.clsa-elcv.ca