Assessing cardiometabolic risk by imaging human adipose tissue

Dr. Alexios Antonopoulos MD PhD

Imaging Specialist CCT/CMR, EACVI Ambassador in CT/CMR

EACVI Scientific Documents Committee

Academic Consultant Cardiologist, 1st Cardiology Dpt, NKUA

Senior Researcher, Biomedical Research Foundation of the Academy of Athens
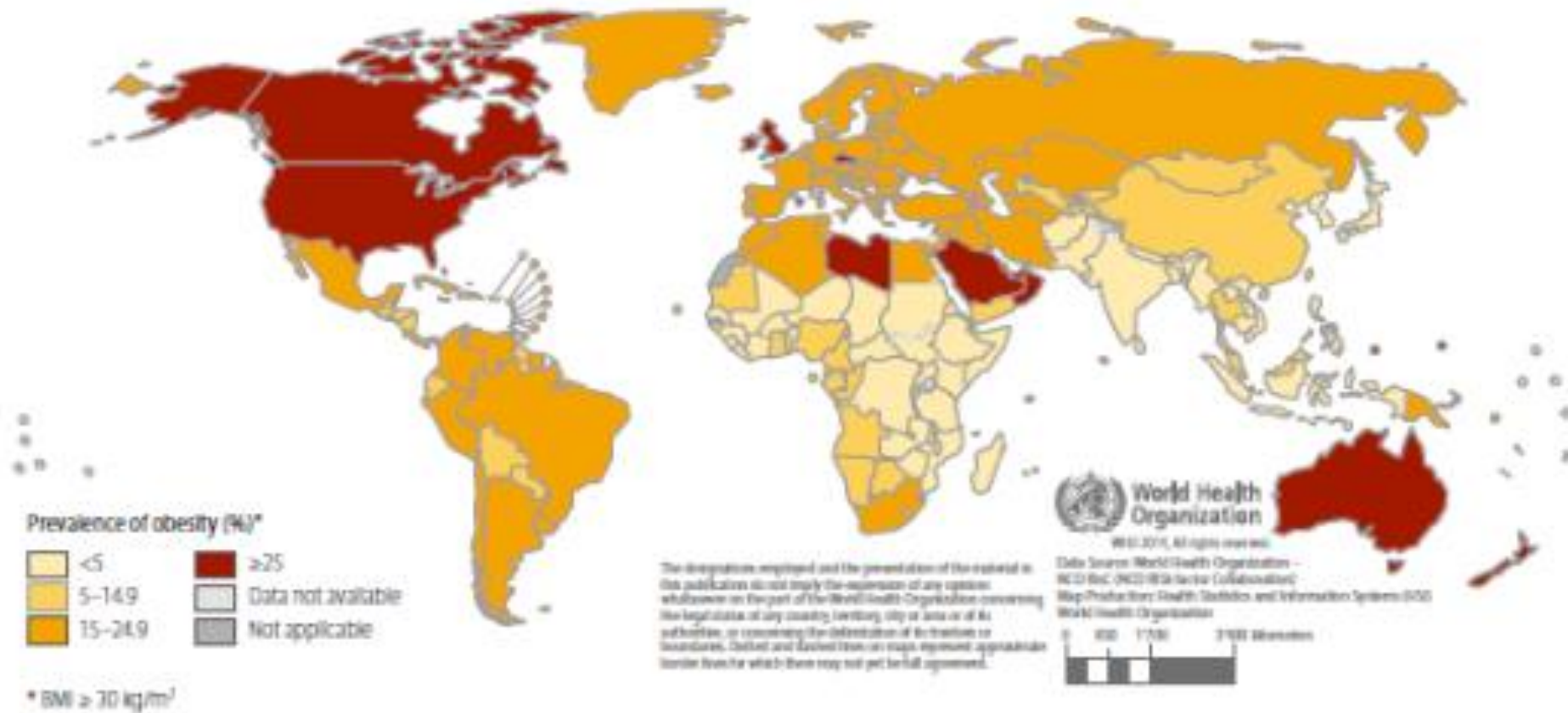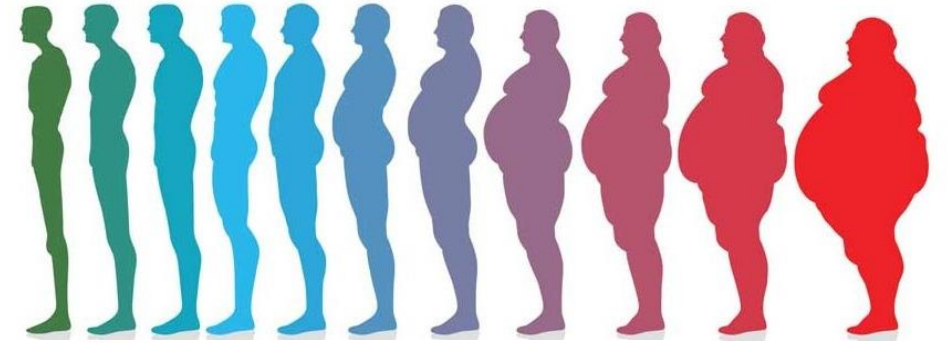
Disclosure of Interests

Patent holder of GB2015/052359, GR2018/0100490 and GR2018/0100510 with royalties paid to Caristo Diagnostics

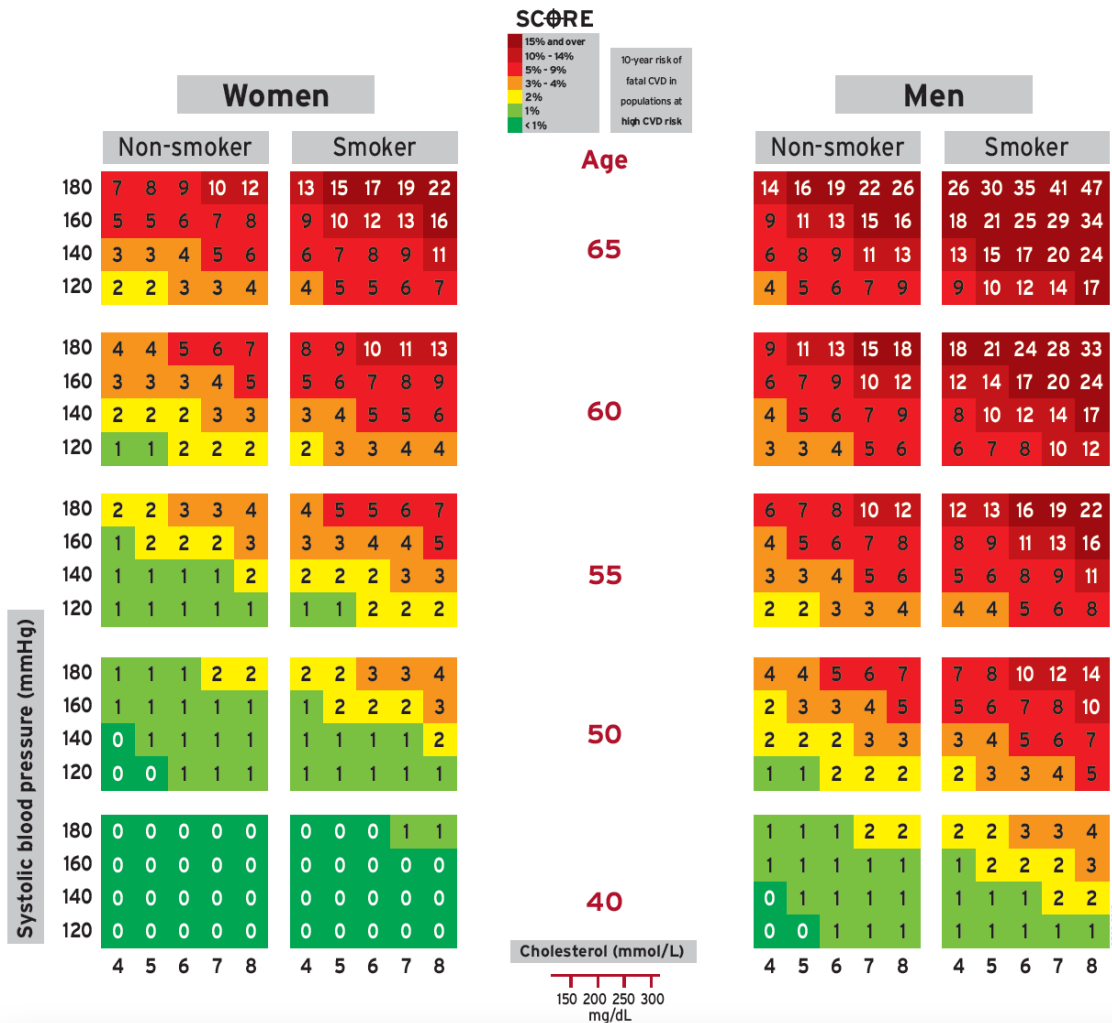Consultancy fees and shareholder (family member) of Caristo Diagnostics, a medical imaging company.

The Obesity Pandemic

Worldwide obesity has nearly tripled since 1975.

>1.9 billion adults overweight
>650 million were obese

Risk stratification for primary prevention of CVD


Risk may be higher than indicated in the chart in:

- Sedentary or obese subjects, especially those with central obesity

We need better ways to identify obese individuals at high risk for CV events

How to assess cardiometabolic risk in obesity?

- Anthropometric indices of obesity
- Cardiorespiratory fitness
- Metabolic status - plasma biomarkers
- Fat biopsies
- Adipose tissue imaging

Anthropometric indices of obesity and CVD risk
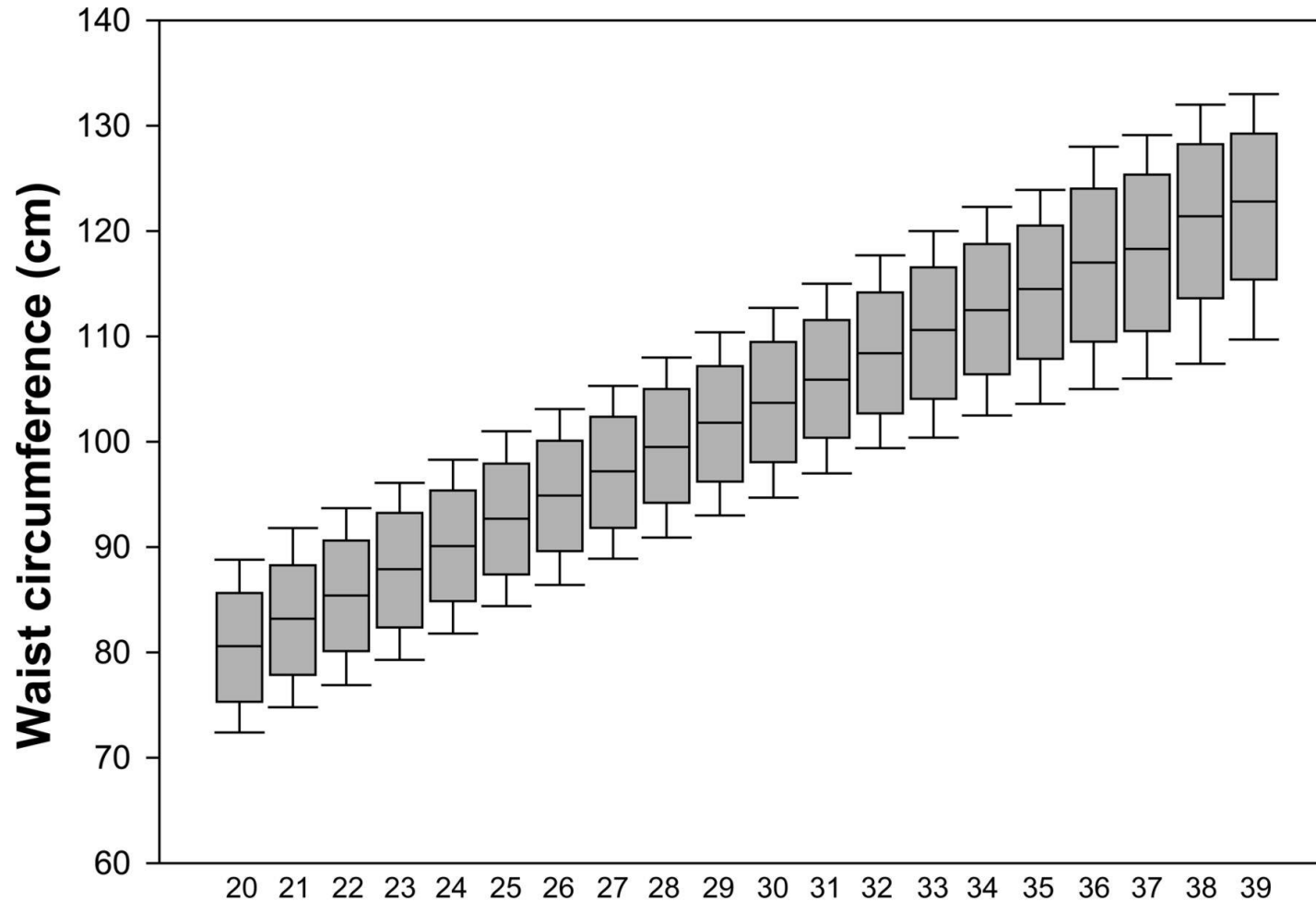
Do fat people live longer ???

"Obesity Paradox"

Antonopoulos AS et al. Cardiov Res 2017

Is obesity "protective" in chronic diseases?

Meta-analysis: Aune D et al; BMJ 2016
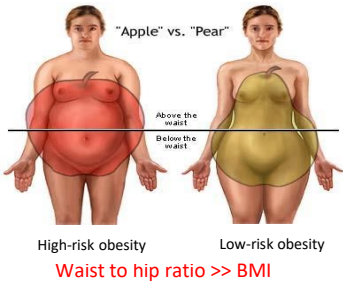230 cohort studies
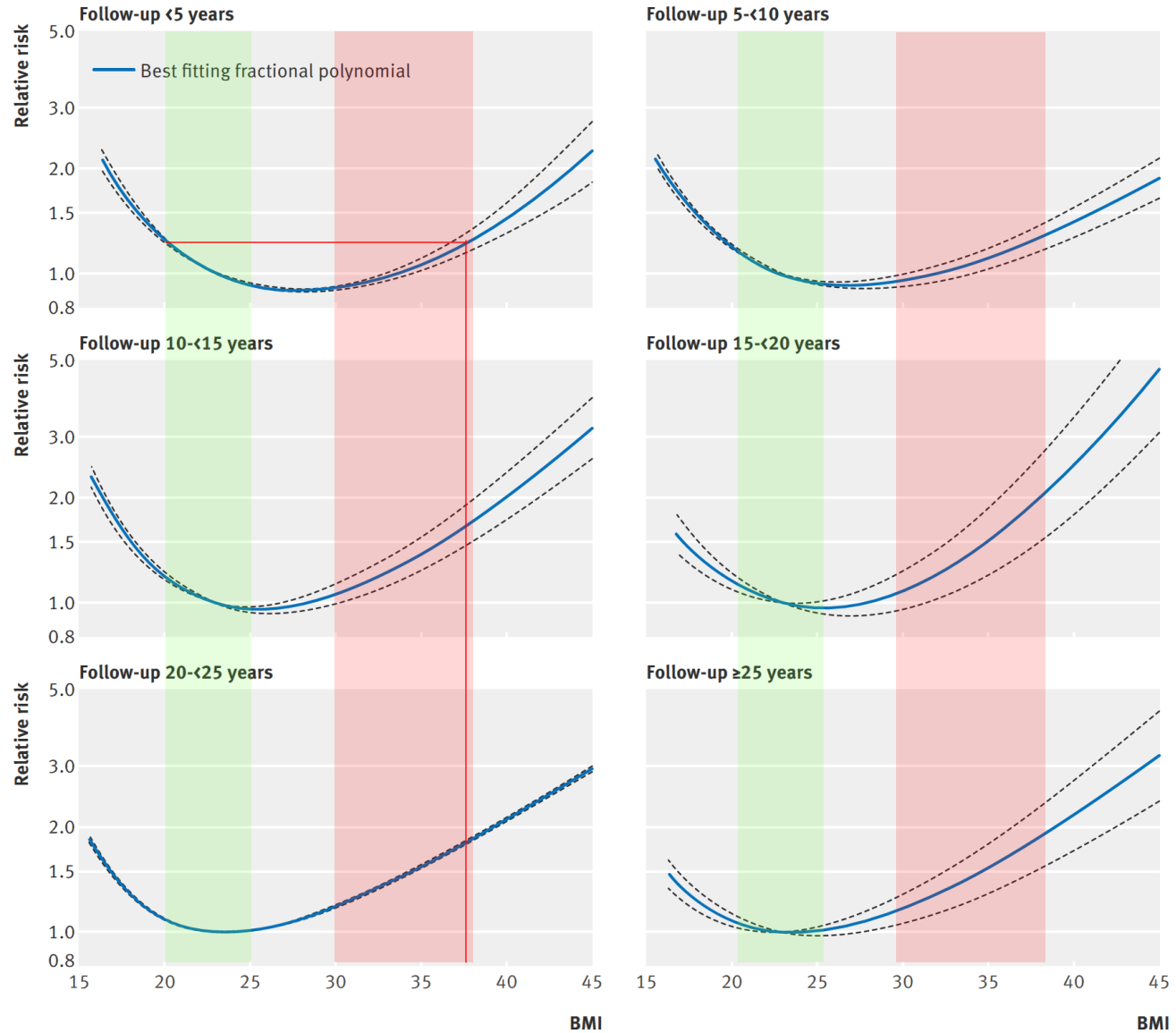30.3m participants
3.74m deaths

The epidemiologist's view:
-Confounders
-Chronic diseases
-Between-studies heterogeneity
-......

The endocrinologist's view:
-Location, location, location...

"Apple" vs. "Pear"

Above the waist
Below the waist

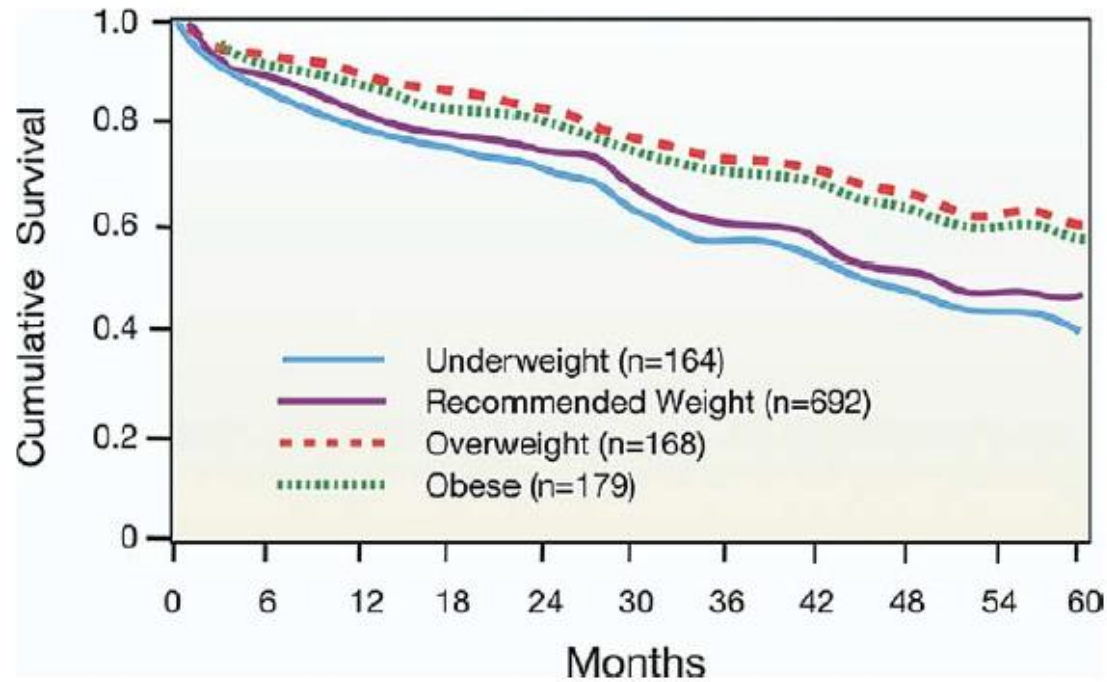High-risk obesity   Low-risk obesity

Waist to hip ratio >> BMI

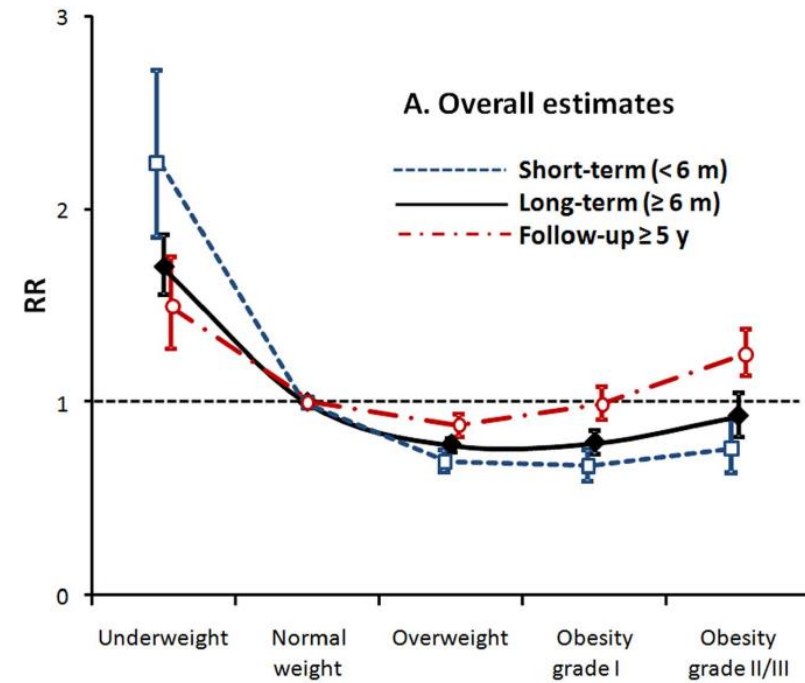**The clinical cardiologist's view:**

The "obesity paradox": A conundrum that needs solving

Patients with HF

J Am Coll Cardiol. 2009;53(21):1925-1932

Patients with CAD

Meta-analysis: Wang ZJ, et al. Heart 2015;0:1–8
89 studies
1,300,794 **patients with CAD**

Is the obesity paradox simply bad statistics?

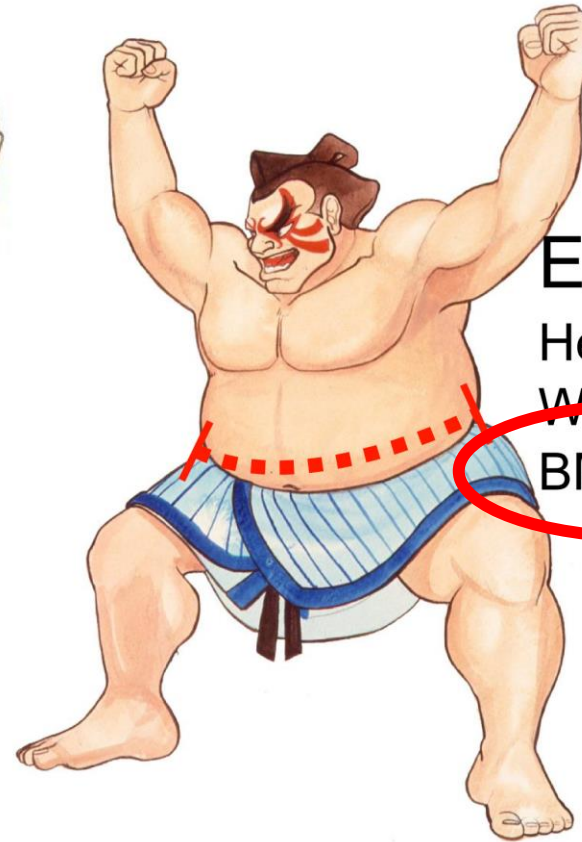

Zangief
Height: 7' 0"
Weight: 400 lbs
BMI: 39.5

E. Honda
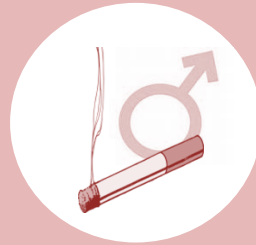Height: 6' 1"
Weight: 302 lbs
BMI: 40

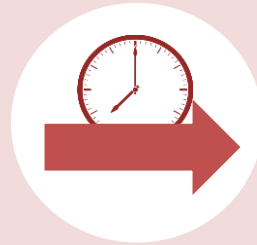# How can the notion of obesity paradox be explained?

THE INHERENT STATISTICAL LIMITATIONS OF CLINICAL STUDIES
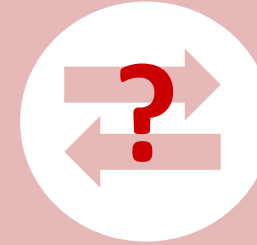
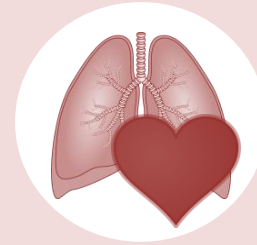| Selection & Survival bias | Confounders (unadjusted for) | Inadequate follow-up period | Treatment bias | The reference BMI group | Reverse causality |

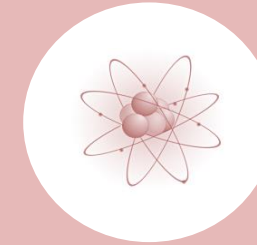## THE USE OF BMI AS AN INDEX OF OBESITY AND ITS LIMITATIONS
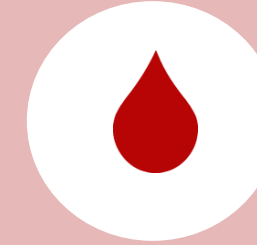
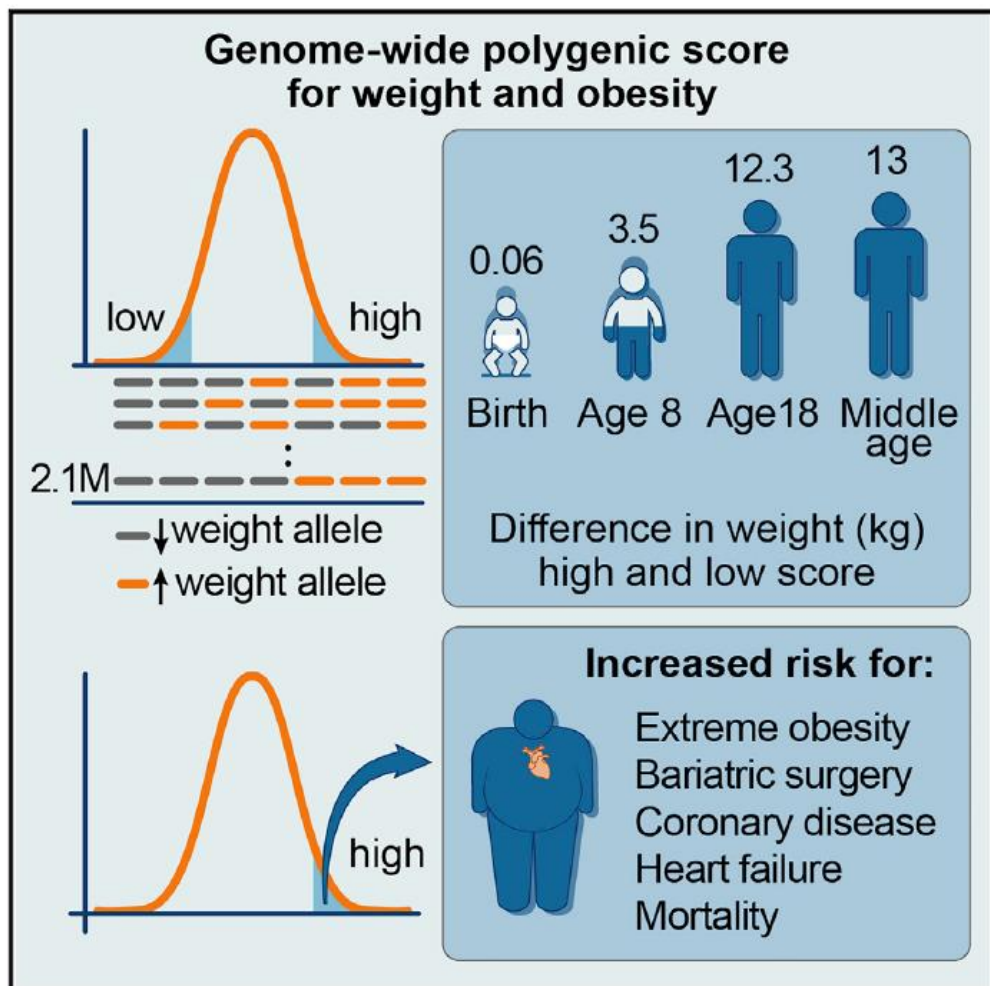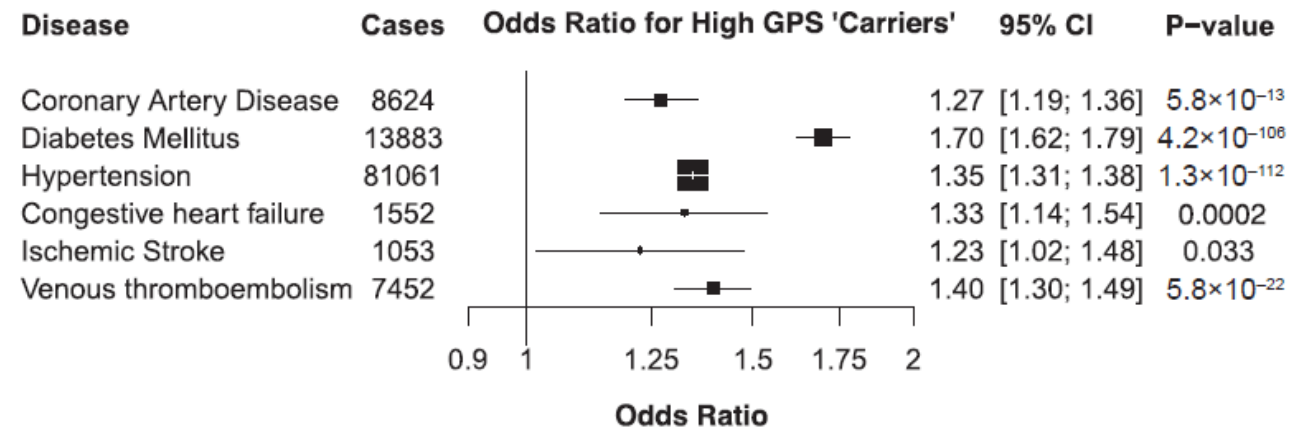| Fat distribution | Body composition | Cardiorespiratory fitness | Metabolic reserve | Coronary anatomy | Bleeding complications |

Antonopoulos AS et al. Obes Rev 2016

Establishing a causal relationship between obesity and CVD risk

300,000 individuals genome wide screening 2.1 m SNPs assessed

Genome-wide polygenic score for weight and obesity

low high

0.06 3.5 12.3 13

Birth Age 8 Age18 Middle age

Difference in weight (kg) high and low score

2.1M

— ↓weight allele
— ↑weight allele

Increased risk for:

Extreme obesity
Bariatric surgery
Coronary disease
Heart failure
Mortality

high

High GPS carrier had increased risk for
- morbid obesity
- Bariatric surgery
- CVD development

Disease	Cases	Odds Ratio for High GPS 'Carriers'		95% CI	P-value
Coronary Artery Disease	8624		1.27	[1.19; 1.36]	5.8×10^{-13}
Diabetes Mellitus	13883		1.70	[1.62; 1.79]	4.2×10^{-106}
Hypertension	81061		1.35	[1.31; 1.38]	1.3×10^{-112}
Congestive heart failure	1552		1.33	[1.14; 1.54]	0.0002
Ischemic Stroke	1053		1.23	[1.02; 1.48]	0.033
Venous thromboembolism	7452		1.40	[1.30; 1.49]	5.8×10^{-22}

0.9 1 1.25 1.5 1.75 2

Odds Ratio

Khera et al. Cell 2019; 177: 587–596

OBESITY is Harmful

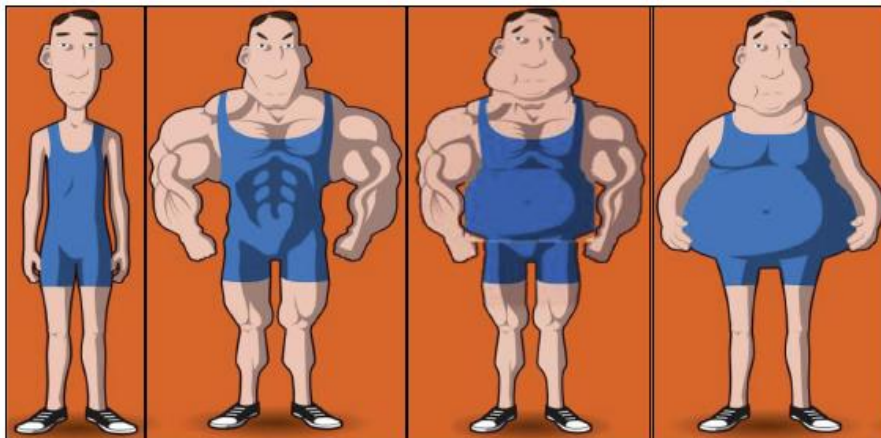But obesity is a heterogeneous mixture of distinct metabolic phenotypes
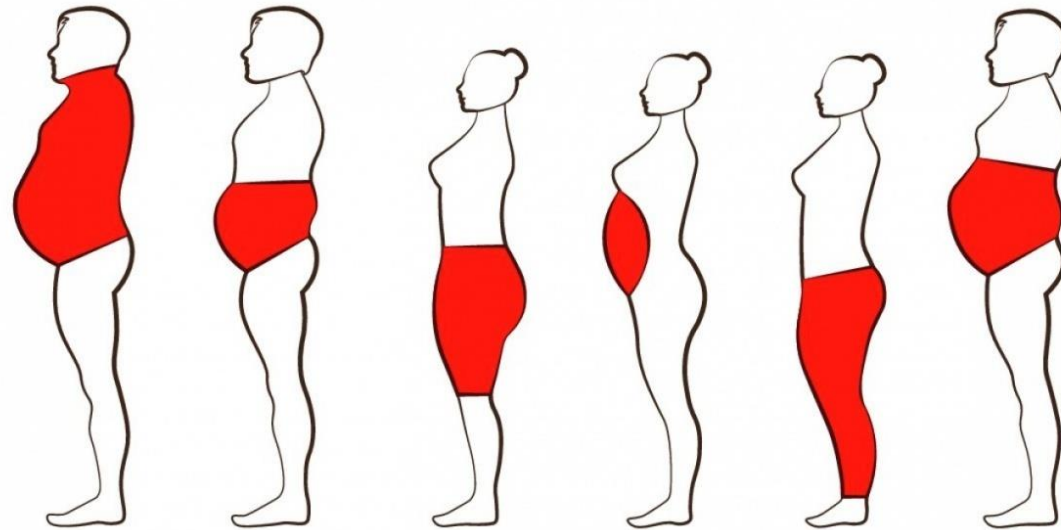
& anthropometric indices of obesity <u>do not accurately </u>describe
the obesity-related burden to cardiovascular health

# Obesity, cardiorespiratory fitness and CV risk

the 'Fat but fit' hypothesis

Body Composition and Obesity Phenotypes	Normal weight	Athlete	Nonsarcopenic Obese	Sarcopenic Obese
BMI (kg/m^2)	18.5-25	≥30	≥ 30	≥ 30
Fat Mass	Normal	Decreased	Increased	Increased
Lean Mass	Normal	Increased	Increased	Decreased
Cardio - Respiratory Fitness	Normal	Increased	*Mild Impairment?*	*Severe Impairment?*

N=2316 men with no known vascular disease
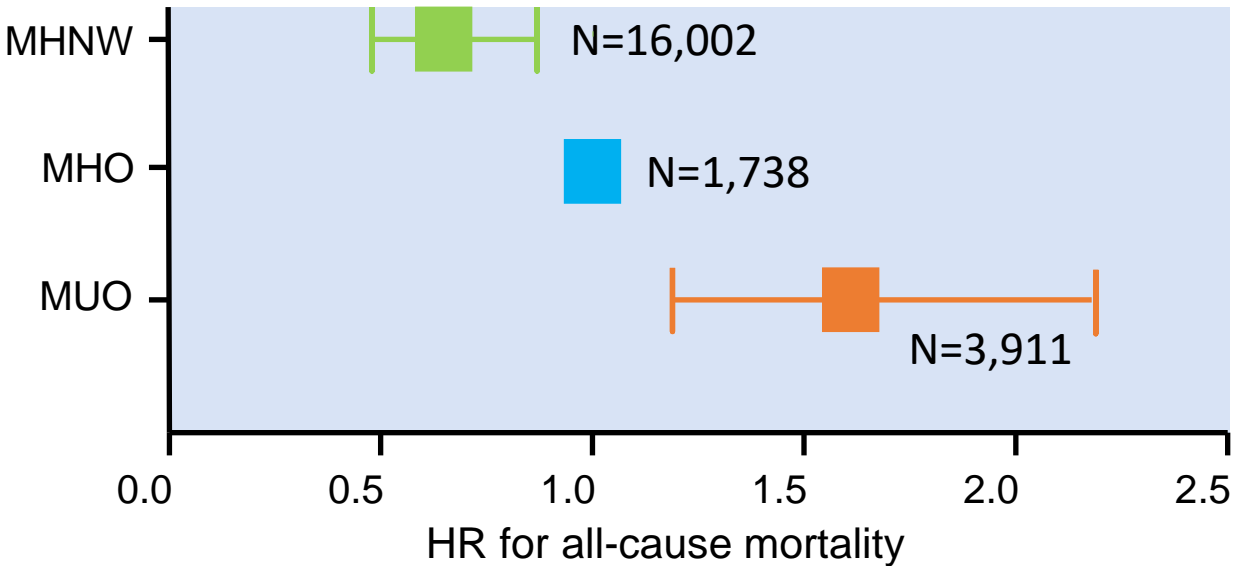Fitness by treadmill test (METS)
Obesity by BMI
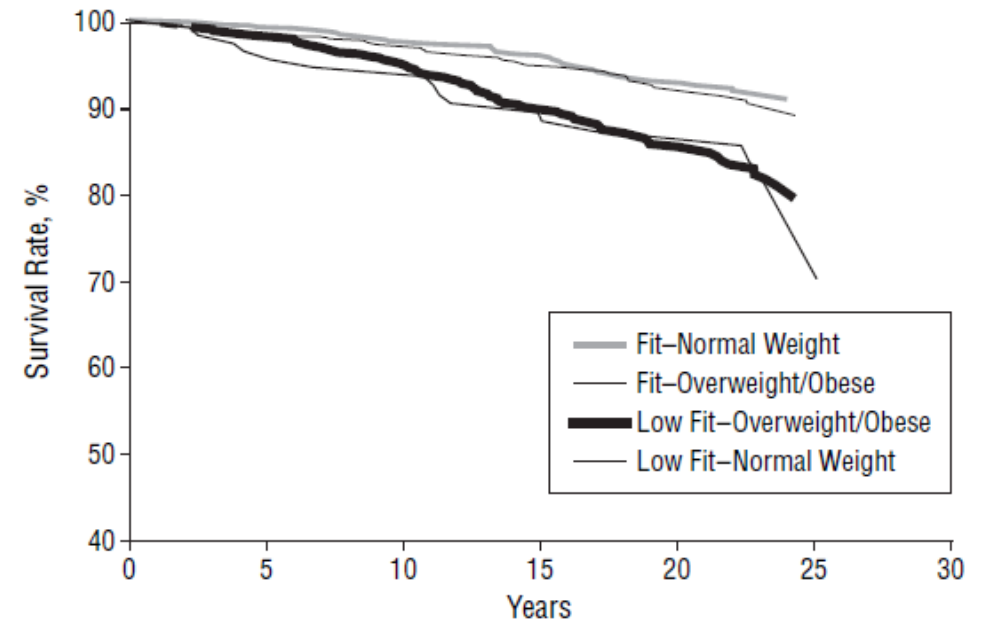
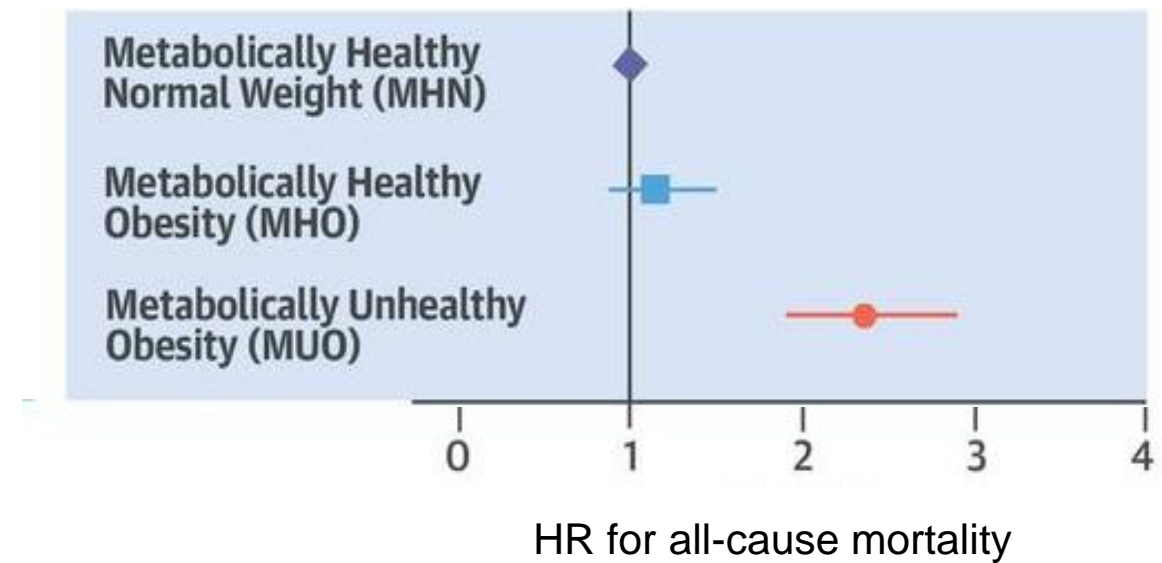Poor fitness and not obesity is a risk factor for adverse outcomes!

Church et al. Arch Intern Med. 2005;165:2114-2120

Metabolically healthy obesity and CVD risk

MHO: obesity individuals, BMI > 30kg/m^2 but without metabolic abnormalities
(only 0 or 1 of MetS criteria)

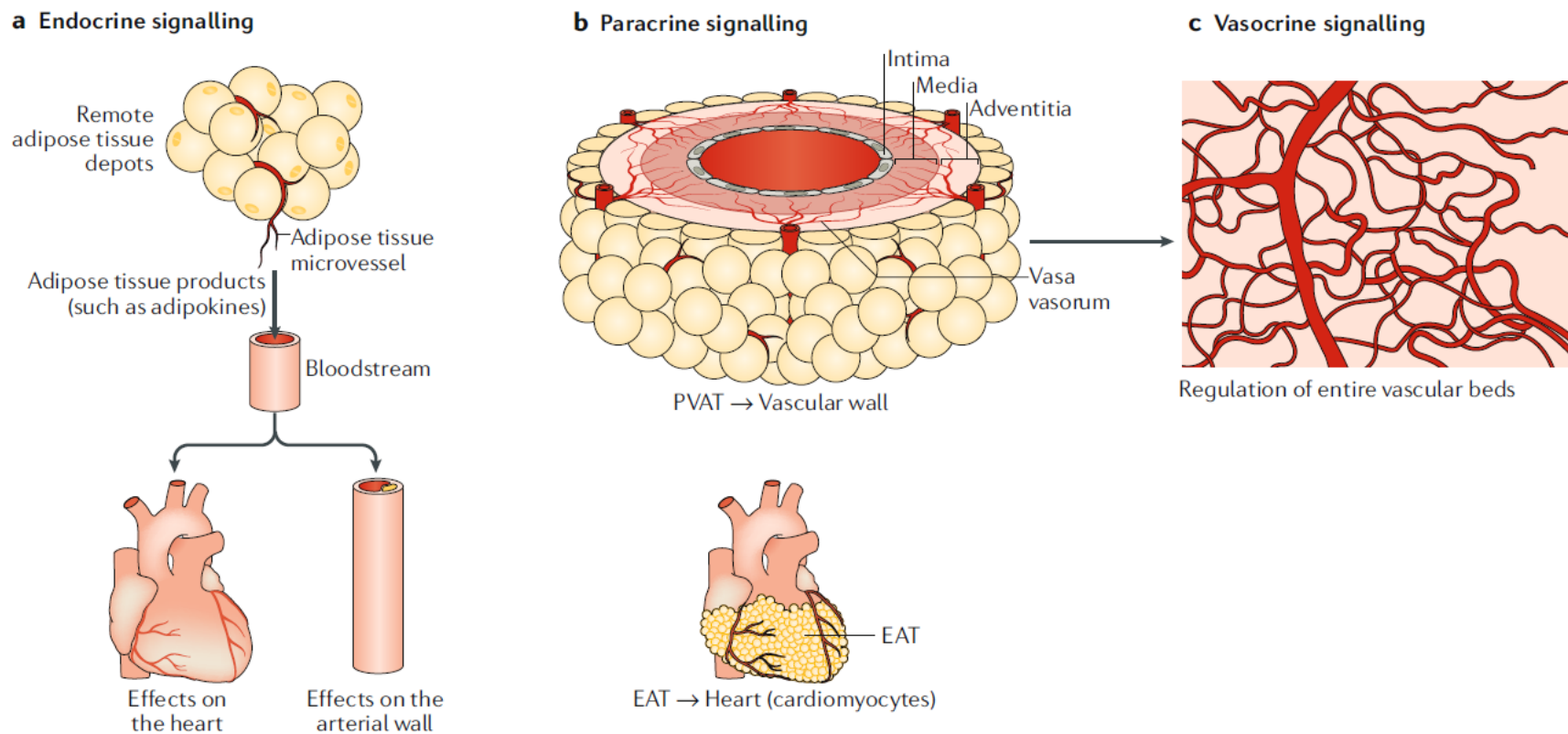

Ortega et al EHJ 2013 N=43,265 adults

Mongraw-Chaffin et al JACC 2018 N=6,809 adults

How to assess cardiometabolic risk in obesity?

- Anthropometric indices
- Cardiorespiratory fitness
- Metabolic status - plasma biomarkers
- Fat biopsies
- Adipose tissue imaging

How to define "benign" vs. "malignant" adiposity ?

a Endocrine signalling

Remote adipose tissue depots

Adipose tissue microvessel

Adipose tissue products (such as adipokines)

Bloodstream

Effects on the heart

Effects on the arterial wall

b Paracrine signalling

Intima
Media
Adventitia

Vasa vasorum

PVAT → Vascular wall

EAT

EAT → Heart (cardiomyocytes)

c Vasocrine signalling

Regulation of entire vascular beds

Study of adipose tissue biology

- Fat biopsies *(invasive, not suitable for screening)*
- Quantify adipose tissue products (proteome/secretome)
- Adipose tissue imaging

Adipose tissue imaging

a Ultrasonography

EAT →

Thickness

Subcutaneous adipose tissue

Femoral adipose tissue
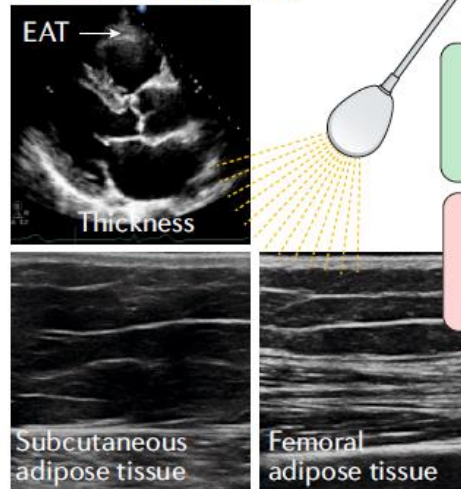
- Low cost
- Wide availability
- Ease of use
- No radiation

- Low reproducibility
- Operator-dependent
- Relies on 2D measurements

b CT imaging

SAT and VAT

EAT and ThAT

PVAT
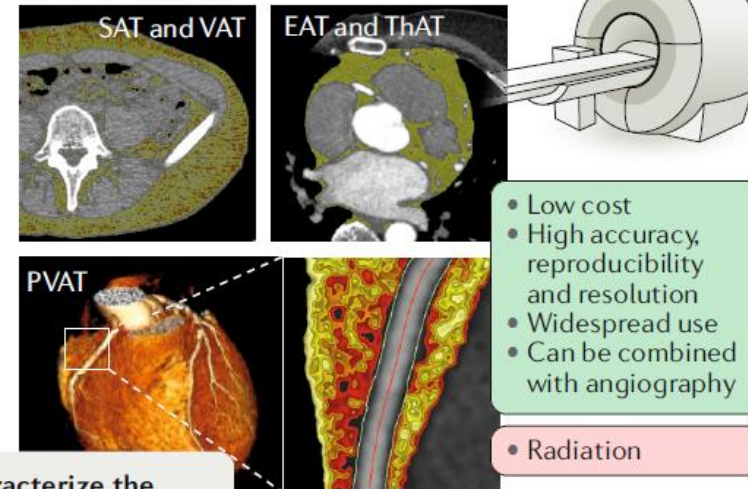
- Low cost
- High accuracy, reproducibility and resolution
- Widespread use
- Can be combined with angiography

- Radiation

How to characterize the adipose tissue in clinical practice

Height

WHtR

WHR

BMI

Weight

c MRI
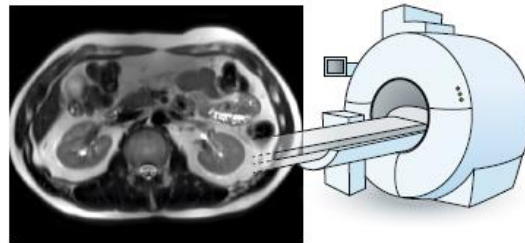
- Accurate volumetric quantification
- No radiation
- Can be combined with spectroscopy, for example, to detect adipose tissue browning

- High cost
- Limited availability
- Long acquisition times
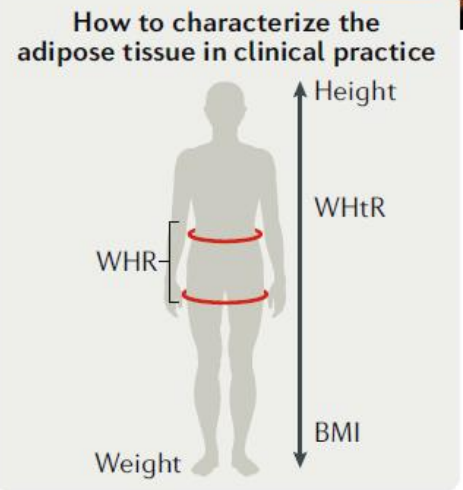
d PET–CT and/or PET–MRI
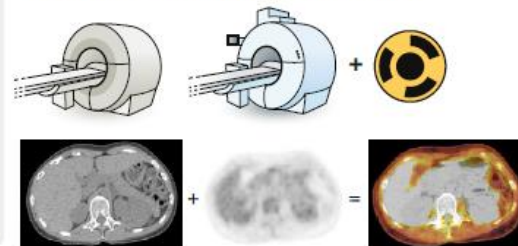
- Provides functional information (such as adipose tissue inflammation and browning)
- Continuous development of novel radiotracers

- High cost
- Limited availability
- Poor spatial resolution
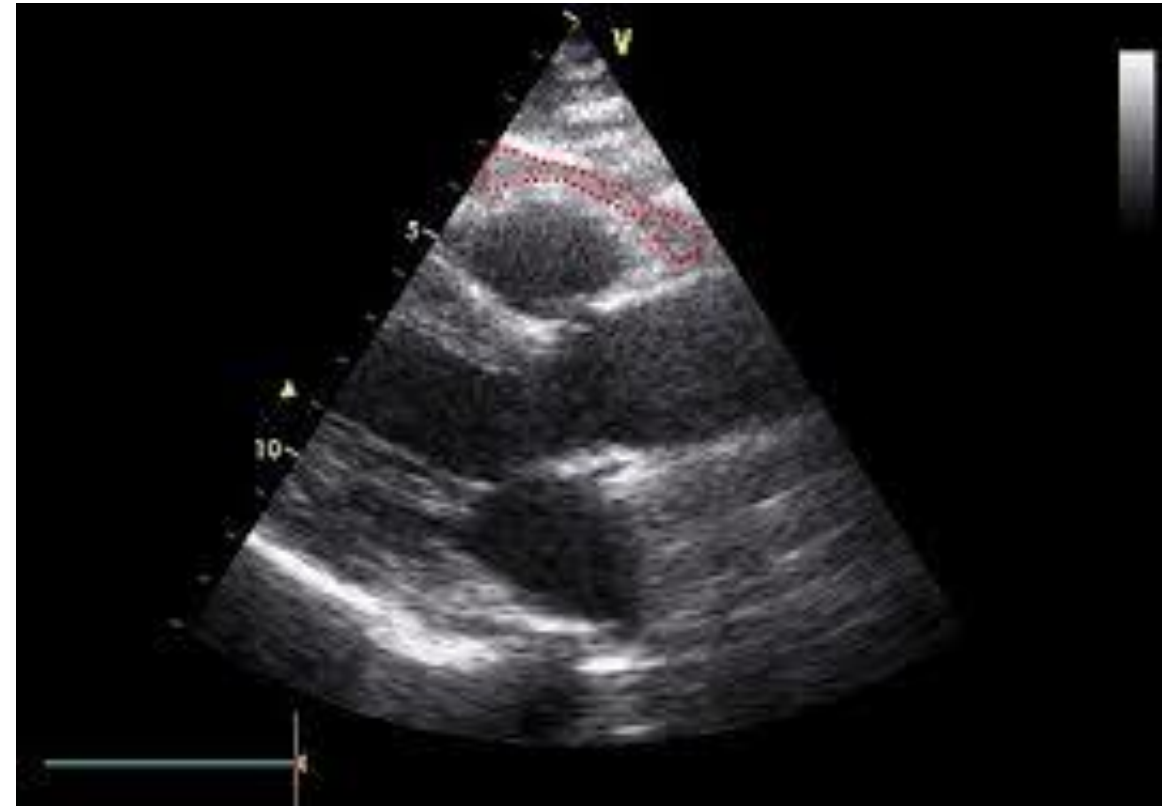- High radiation dose

Adipose tissue imaging

by ultrasound

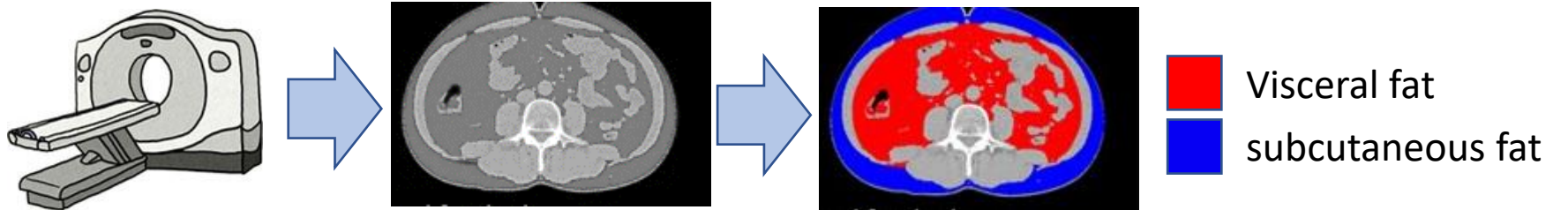Subcutaneous and visceral abdominal fat
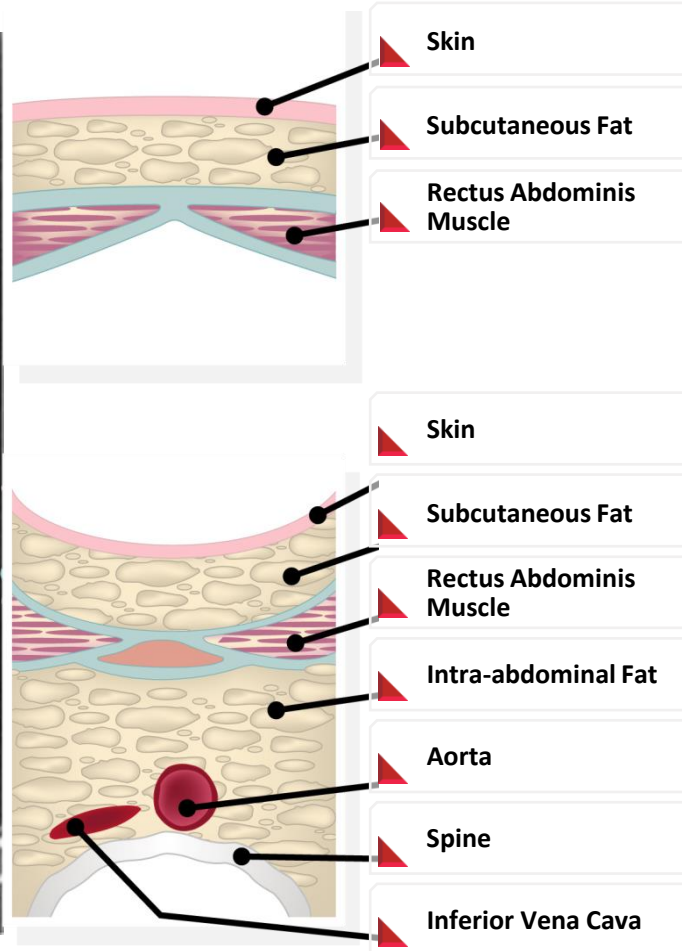
Epicardial adipose tissue

Thickness of human fat pads as a marker of increased metabolic risk
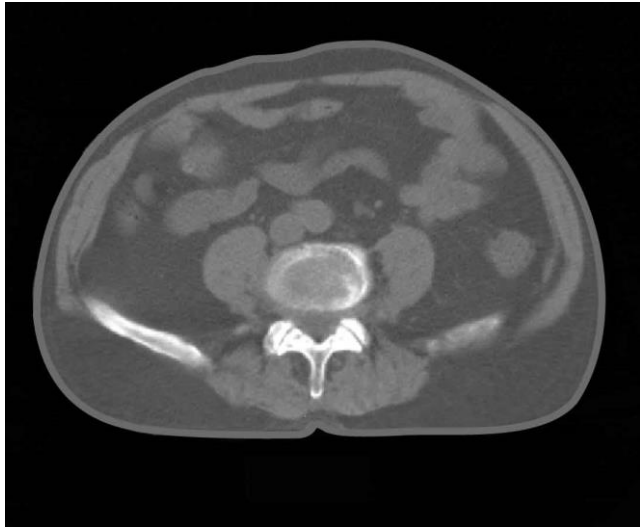
Visceral vs. abdominal fat
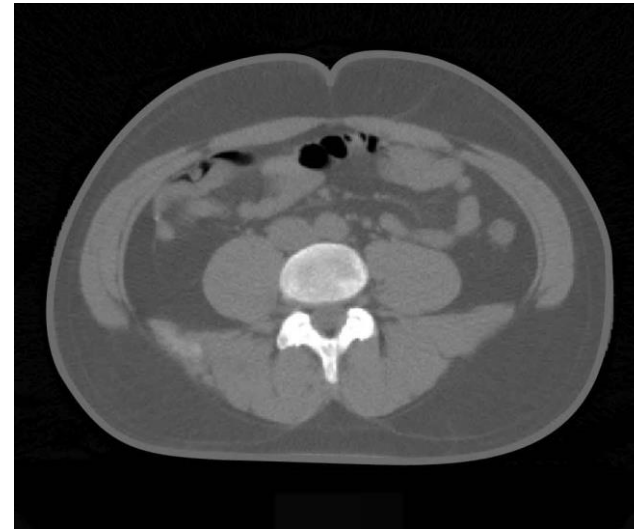
CT imaging of abdominal fat

Fat: -190 to -30 HU

- Visceral fat
- subcutaneous fat

Same WC but different fat distribution

Case 1

- **Waist Circumference: 91 cm**
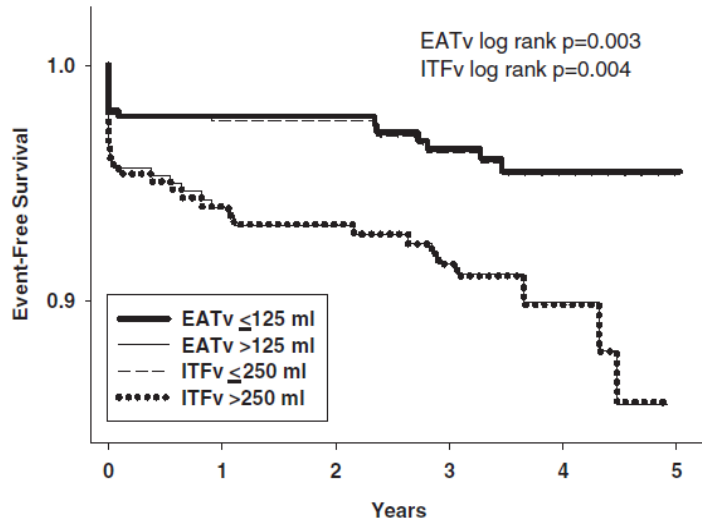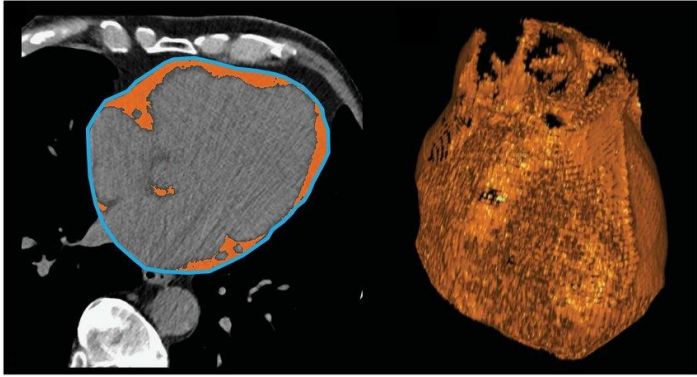- **Intra-abdominal Fat: 190 cm^2**
- **Subcutaneous Fat: 162 cm^2**

Case 2

- **Waist Circumference: 93 cm**
- **Intra-abdominal Fat: 98 cm^2**
- **Subcutaneous Fat: 274 cm^2**
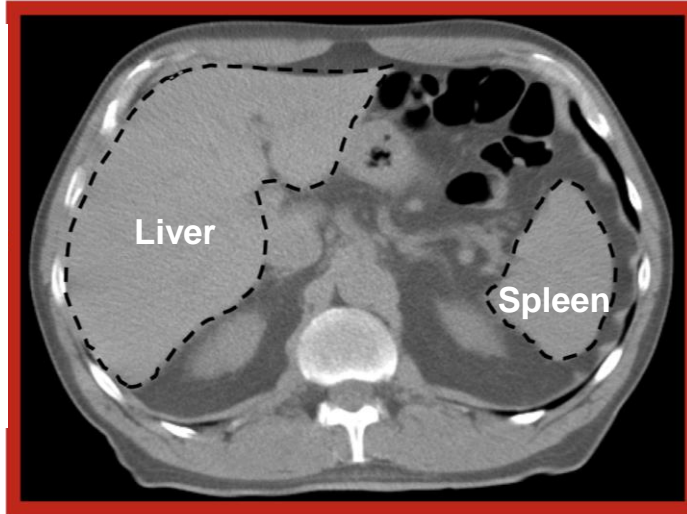
Visceral vs. abdominal fat and cardiovascular disease risk
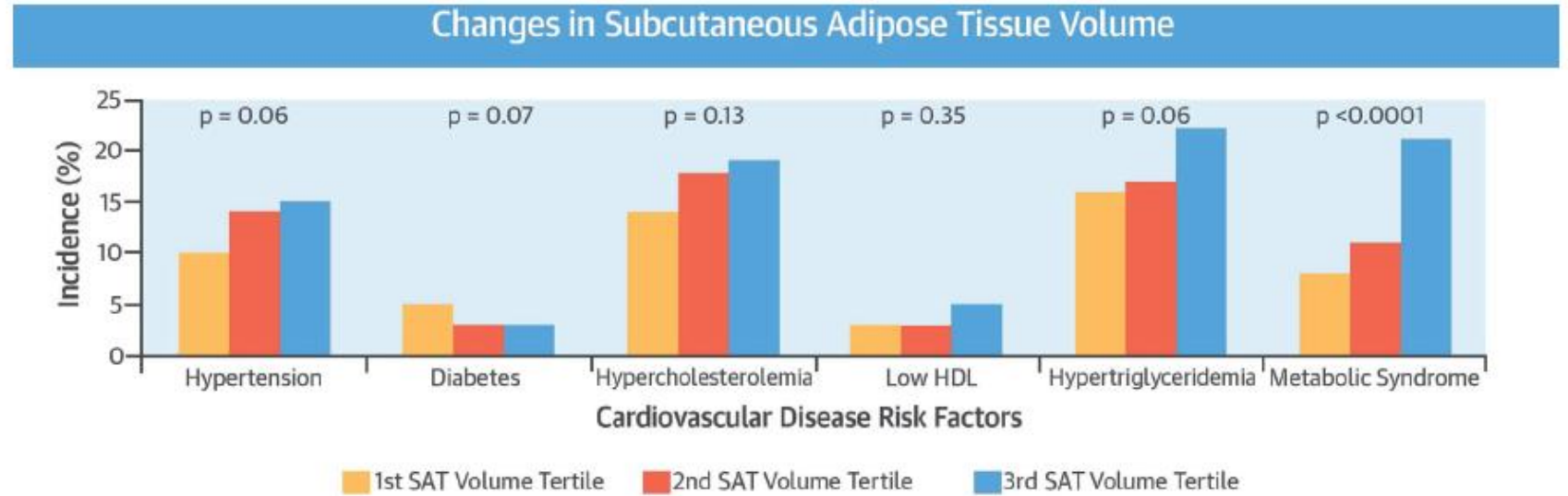
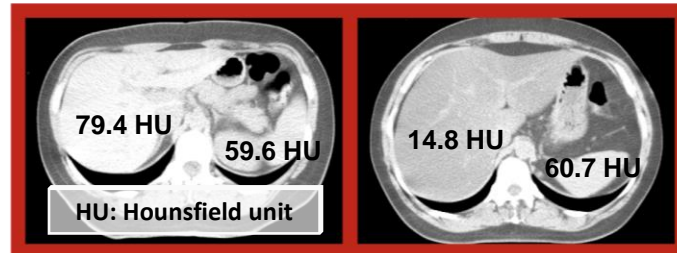# Ectopic adiposity and cardiovascular disease risk
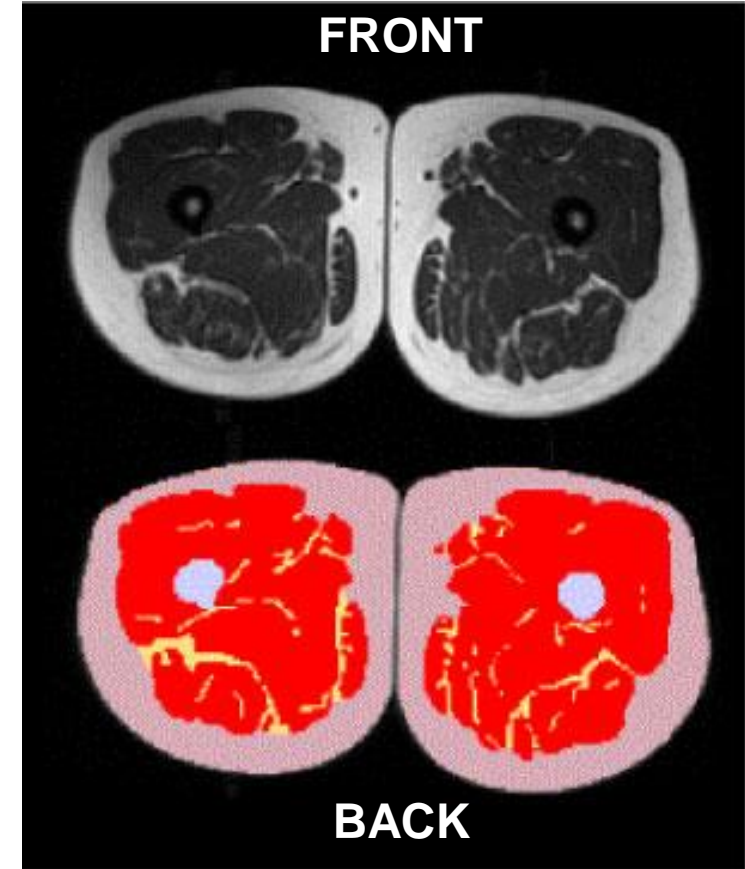
Epicardial adiposity

Fatty liver

$$\frac{\text{Mean Liver Attenuation Value}}{\text{Mean Spleen Attenuation Value}} \longrightarrow \text{CTL/CTS (\text{"Fatty Liver" Index})}$$
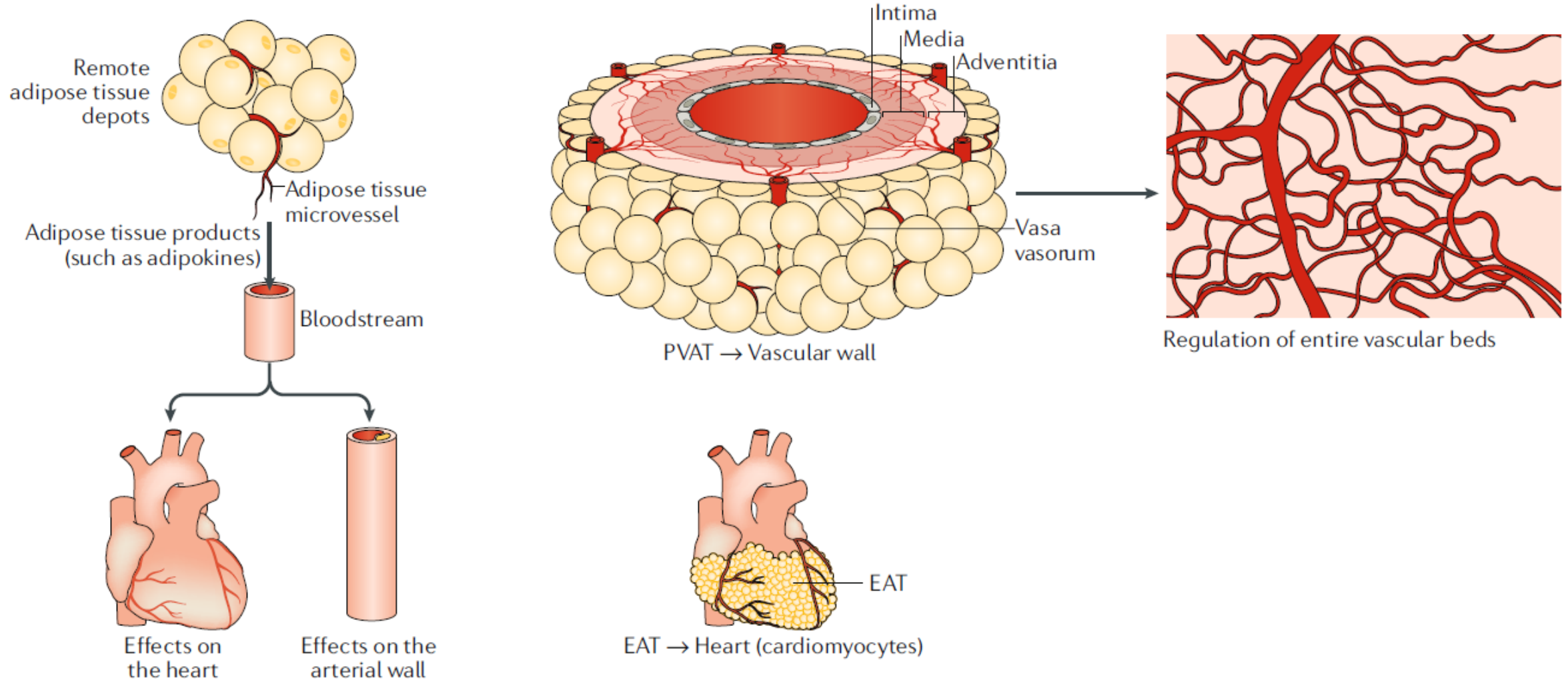
Normal Liver CTL/CTS = 1.33	"Fatty Liver" CTL/CTS = 0.24

Intramuscular fat

FRONT

BACK

Forouzandeh Circ Cardiovasc Imaging. 2013;6:58-66.

Han et al. Cardiovasc Diabetol (2017) 16:54

Human adipose tissue depots – perivascular fat

Remote adipose tissue depots

Adipose tissue products (such as adipokines)

Adipose tissue microvessel

Bloodstream

Effects on the heart

Effects on the arterial wall

Intima
Media
Adventitia

Vasa vasorum

PVAT → Vascular wall

Regulation of entire vascular beds

EAT

EAT → Heart (cardiomyocytes)

The role of adipose tissue dysfunction in T2DM

"Adipocentricity" in T2DM

Healthy WAT expansion
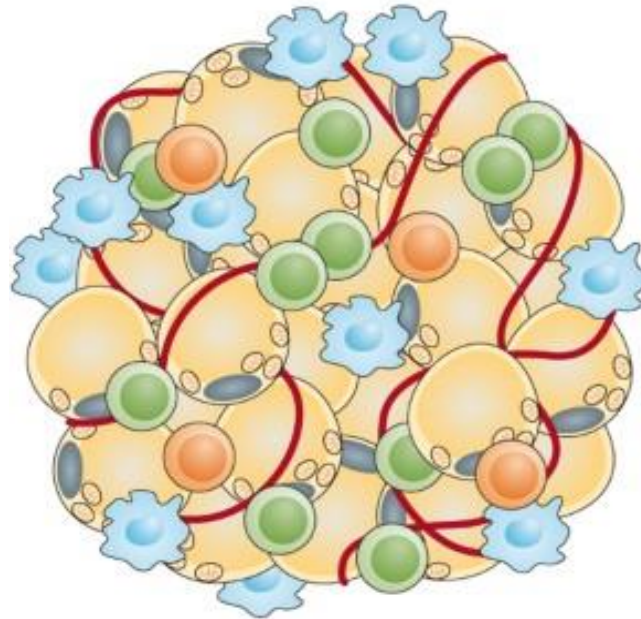
- Adipocyte hyperplasia
- Anti-inflammatory state
  ($\uparrow$ M2 ATMs and $\uparrow$ T$_{regs}$)
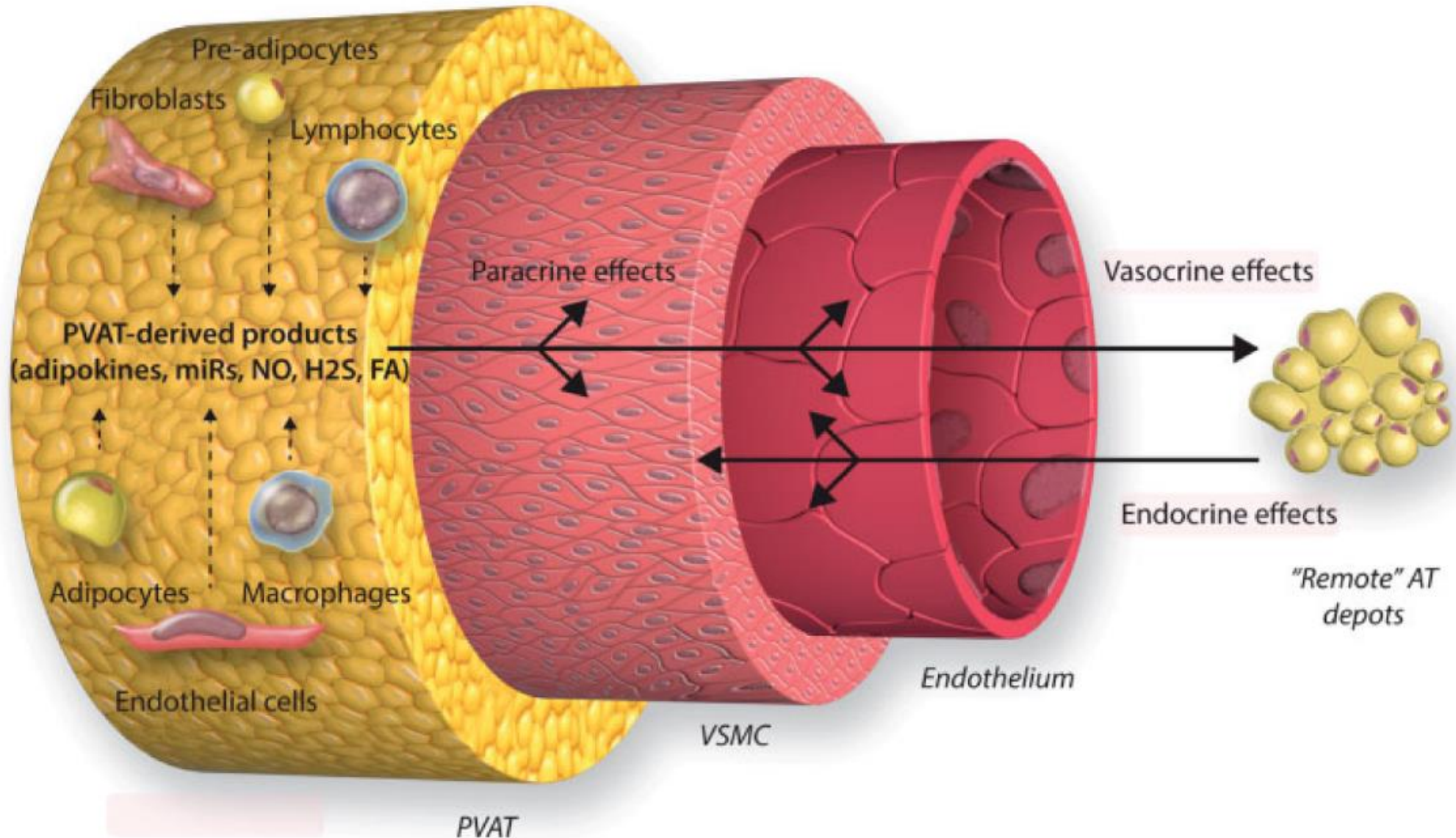- $\uparrow$ Formation of new vasculature
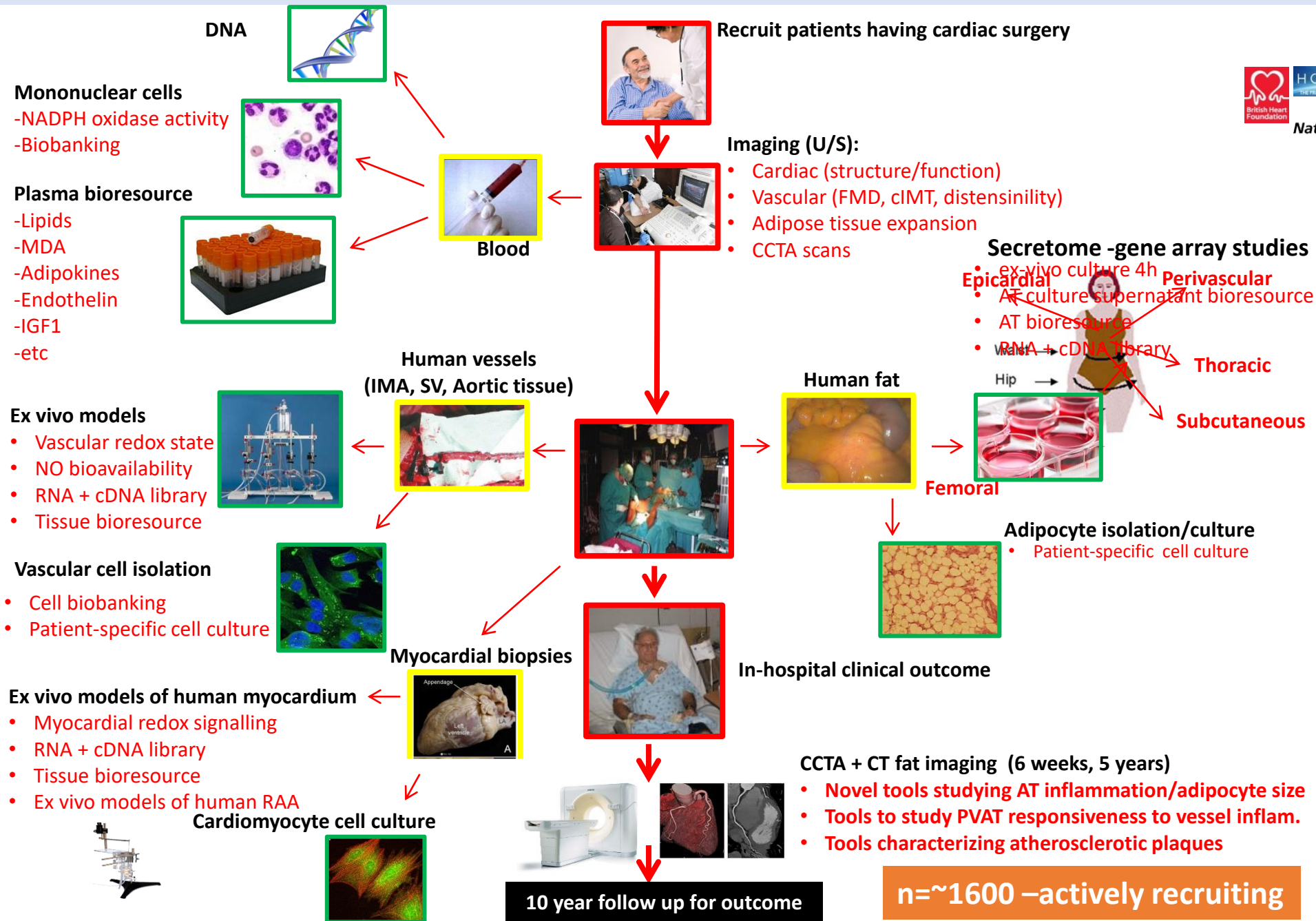
Metabolically healthy obesity and CVD risk
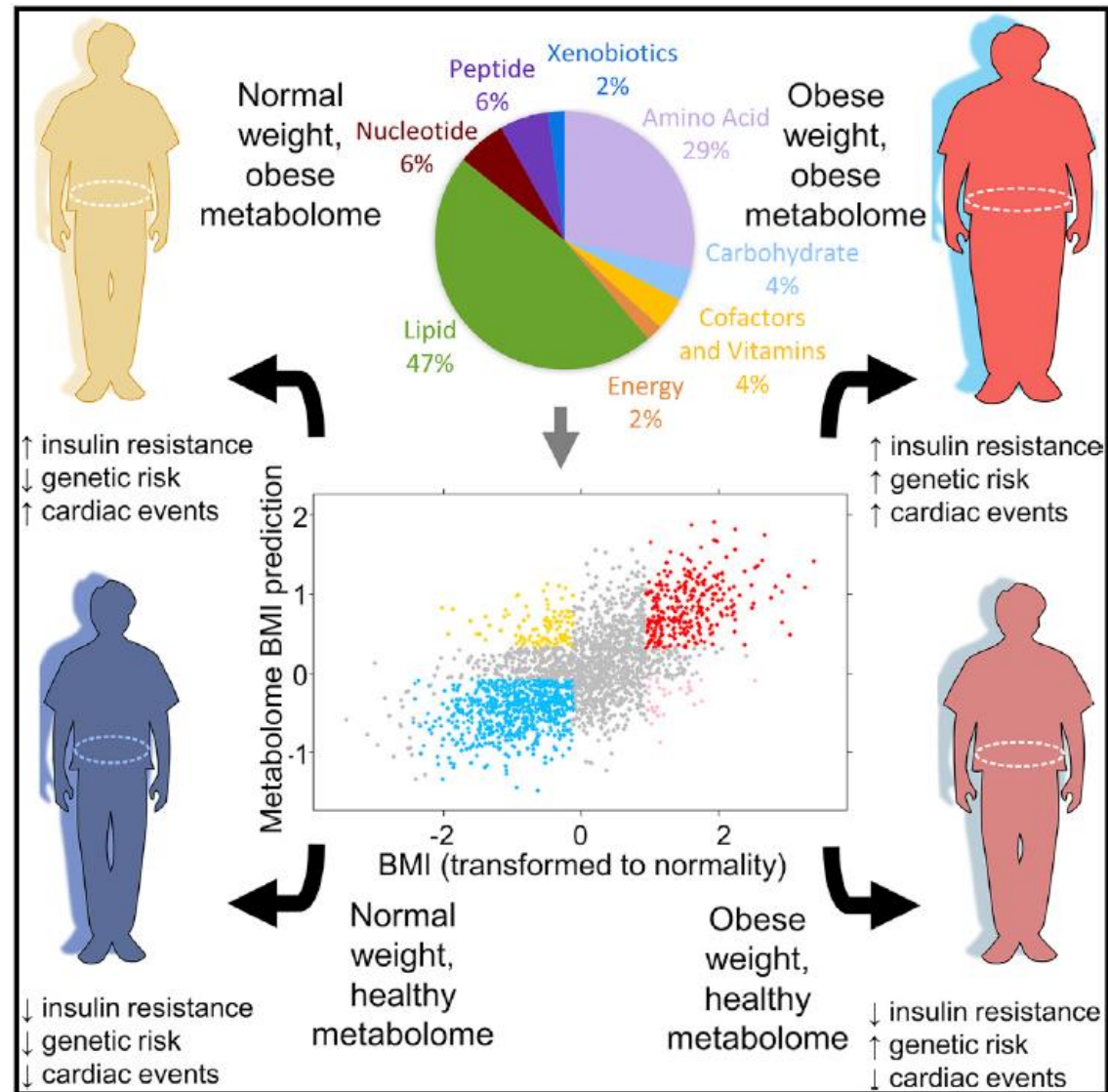
Metabolic profiling may be a more accurate way to assess cardiovascular risk than BMI.

Abnormal metabolome linked to ~5-fold increase in CV events compared to BMI-matched people but with an opposing metabolome.
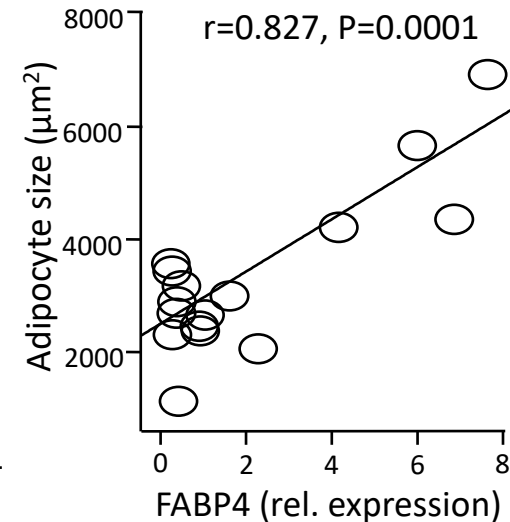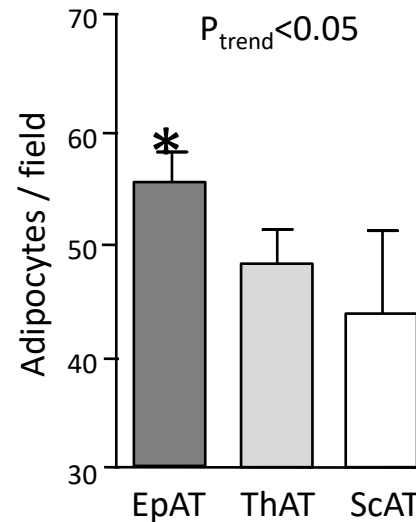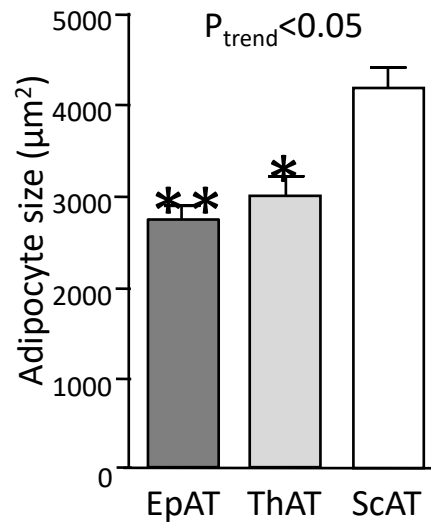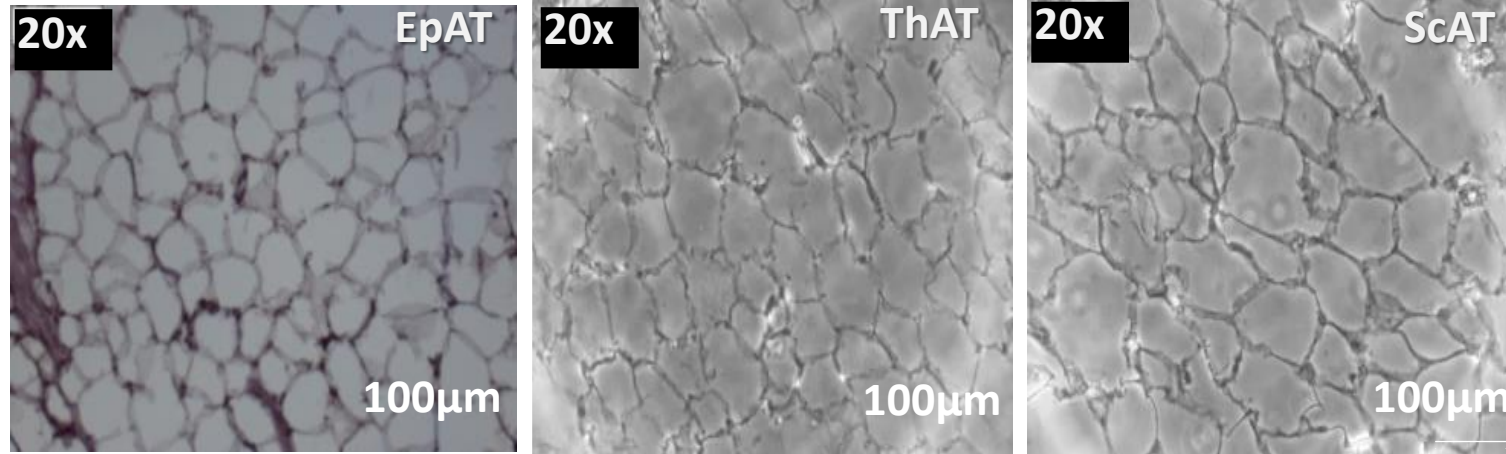
Cirulli et al. Cell Metabolism 2019; 29: 488–500
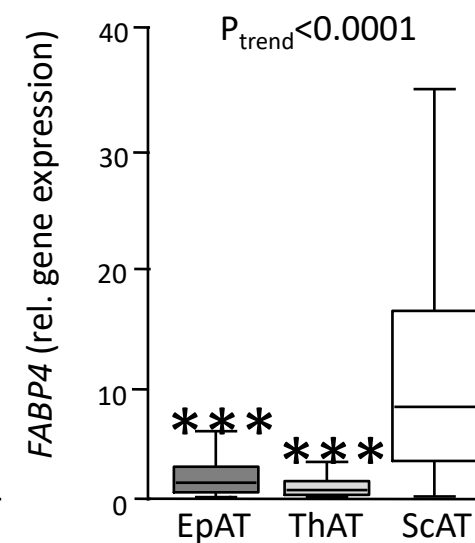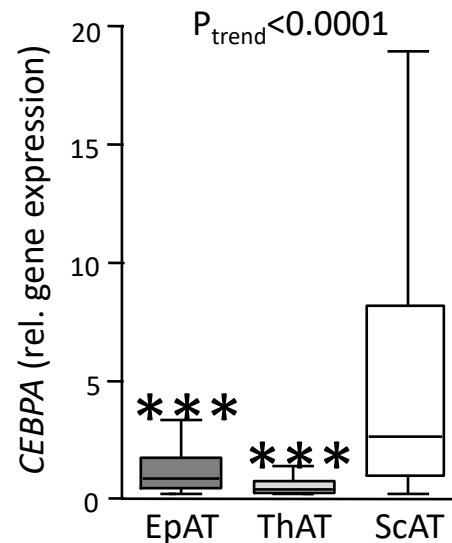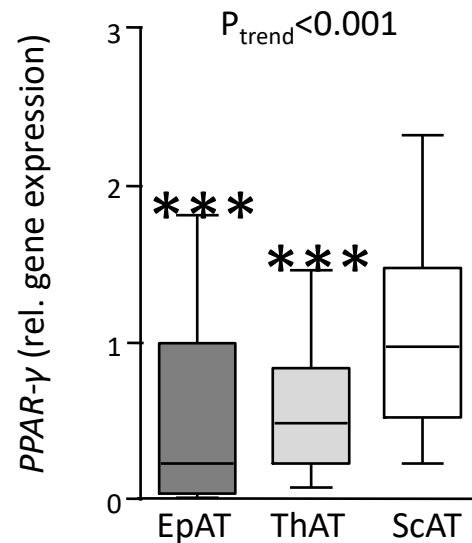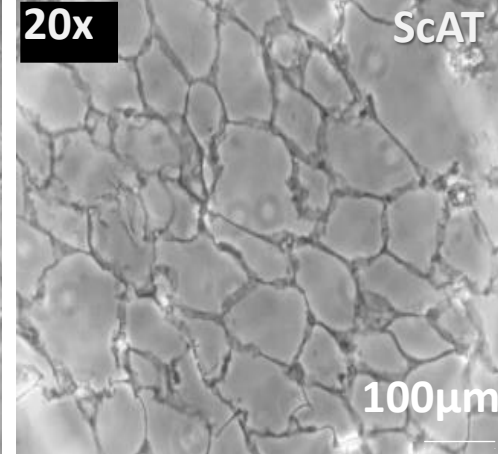
How does adipose tissue communicate with the vascular wall

Akoumianakis & Antoniades Cardiovasc Res 2017

The Oxford Cohort for Heart Vessels and Fat (oxHVF)

DNA

Recruit patients having cardiac surgery

Mononuclear cells
- NADPH oxidase activity
- Biobanking

Imaging (U/S):
- Cardiac (structure/function)
- Vascular (FMD, cIMT, distensinility)
- Adipose tissue expansion
- CCTA scans

Plasma bioresource
- Lipids
- MDA
- Adipokines
- Endothelin
- IGF1
- etc

Blood

Secretome -gene array studies
- ex-vivo culture 4h
- AT culture supernatant bioresource
- AT bioresource
- RNA + cDNA library

Epicardial
Perivascular
Thoracic
Subcutaneous
Hip

Human vessels (IMA, SV, Aortic tissue)

Human fat

Ex vivo models
- Vascular redox state
- NO bioavailability
- RNA + cDNA library
- Tissue bioresource

Femoral

Adipocyte isolation/culture
- Patient-specific cell culture

Vascular cell isolation
- Cell biobanking
- Patient-specific cell culture

Myocardial biopsies

In-hospital clinical outcome

Ex vivo models of human myocardium
- Myocardial redox signalling
- RNA + cDNA library
- Tissue bioresource
- Ex vivo models of human RAA

Appendage
Left ventricle
LA
A

CCTA + CT fat imaging (6 weeks, 5 years)
- Novel tools studying AT inflammation/adipocyte size
- Tools to study PVAT responsiveness to vessel inflam.
- Tools characterizing atherosclerotic plaques

Cardiomyocyte cell culture

10 year follow up for outcome

n=~1600 –actively recruiting

Differences between adipose tissue depots

Differences between adipose tissue depots

Antonopoulos AS et al Science Transl Med 2017
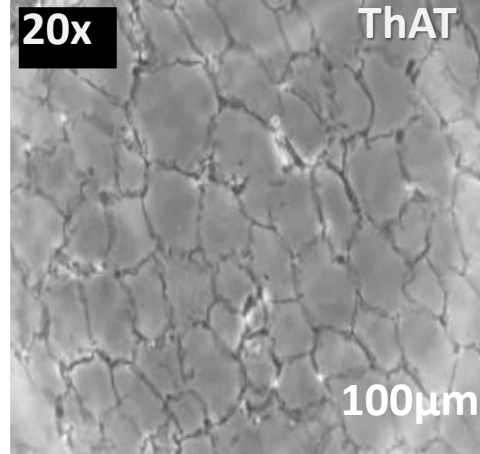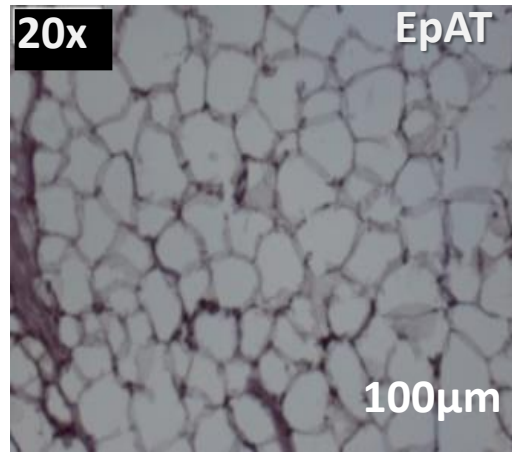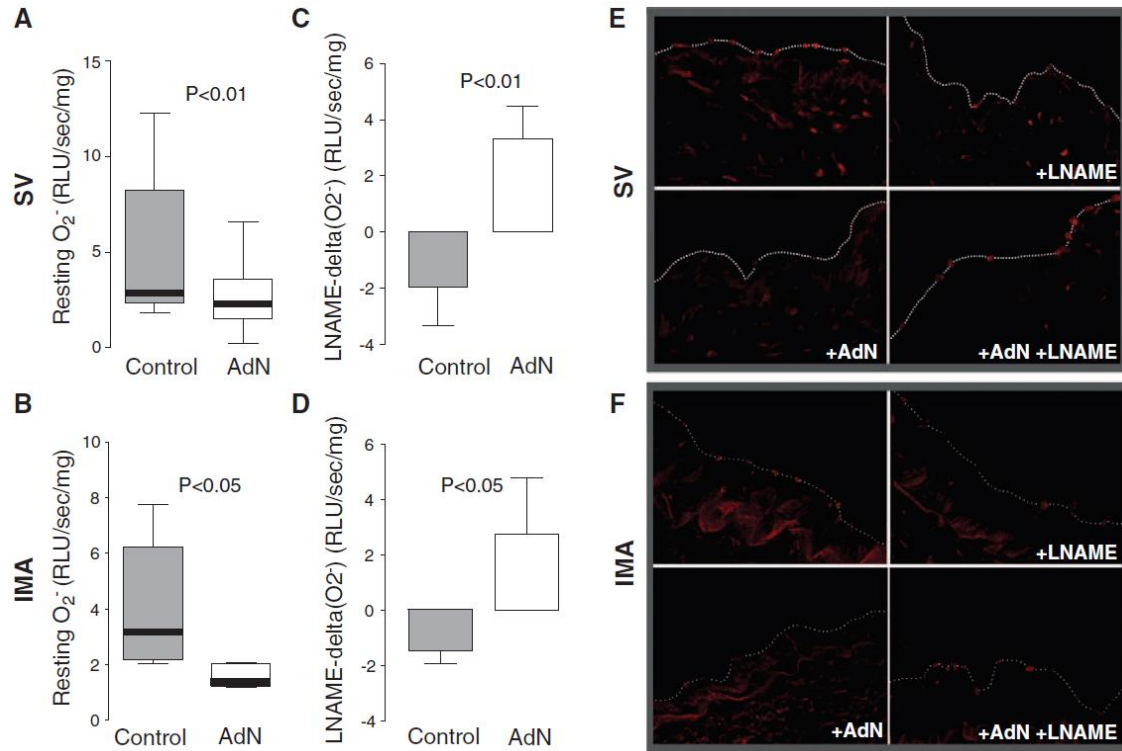
Discovering new <u>endocrine</u> signals from the AT to the vascular wall

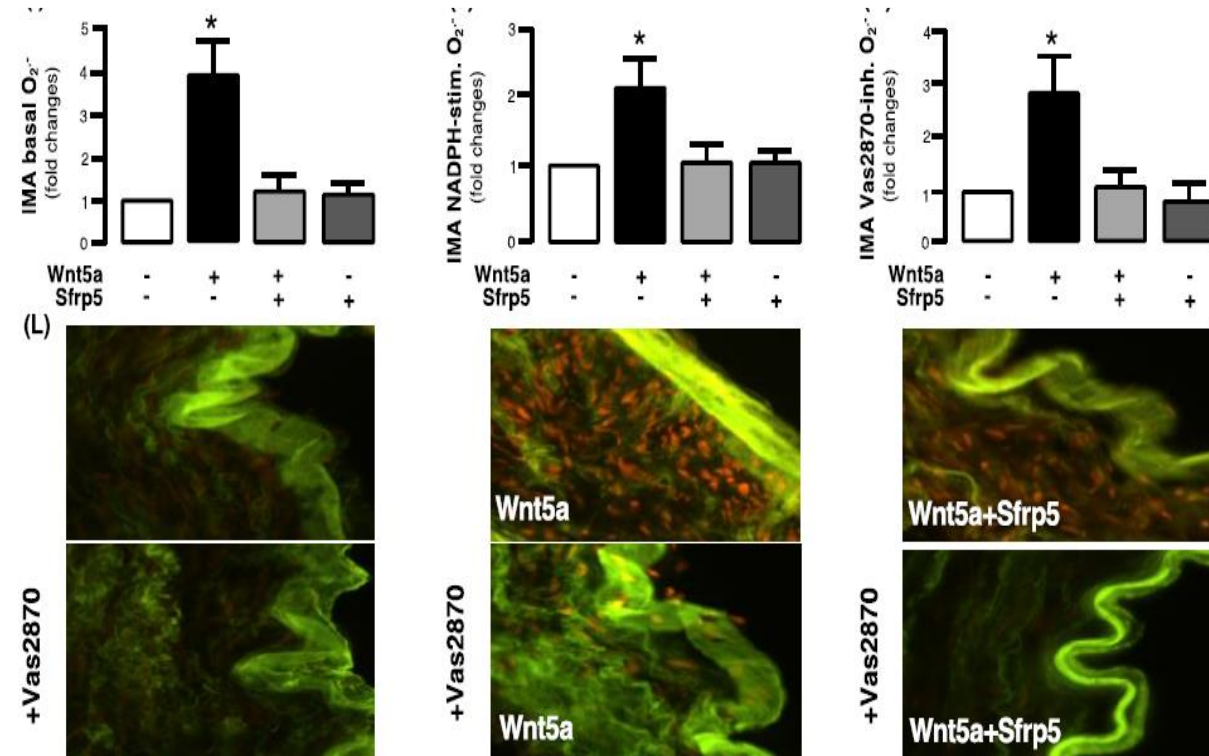# The cross-talk between adipose tissue and cardiovascular system

Adiponectin

Wnt5a/Sfrp5

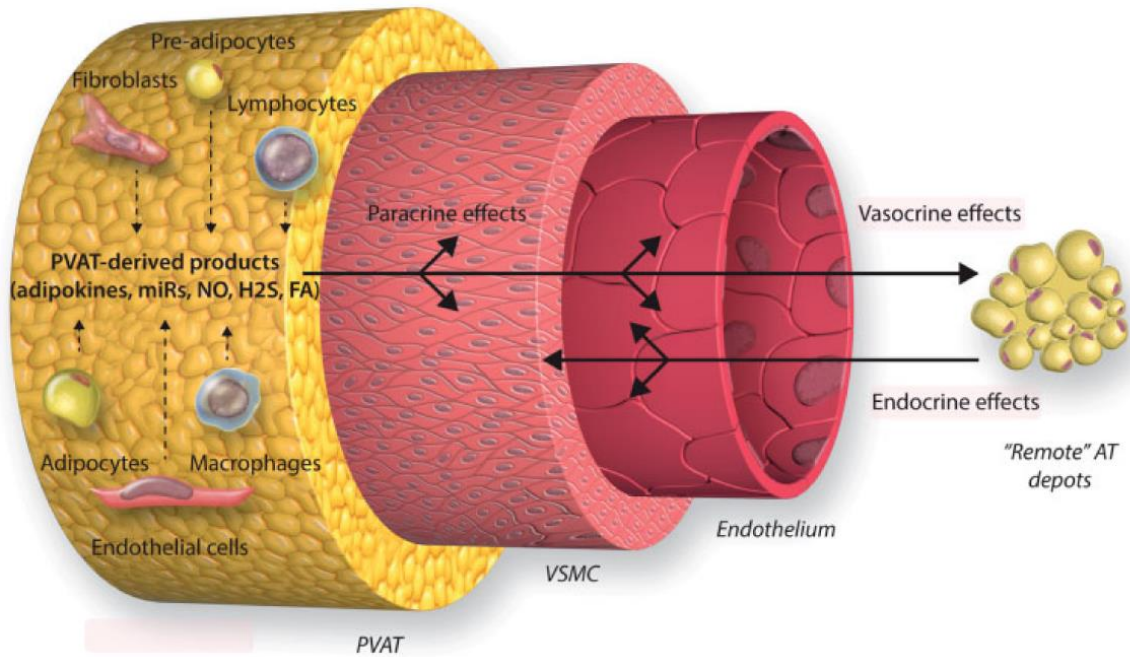Antonopoulos AS & Margaritis M et al. Circulation 2013
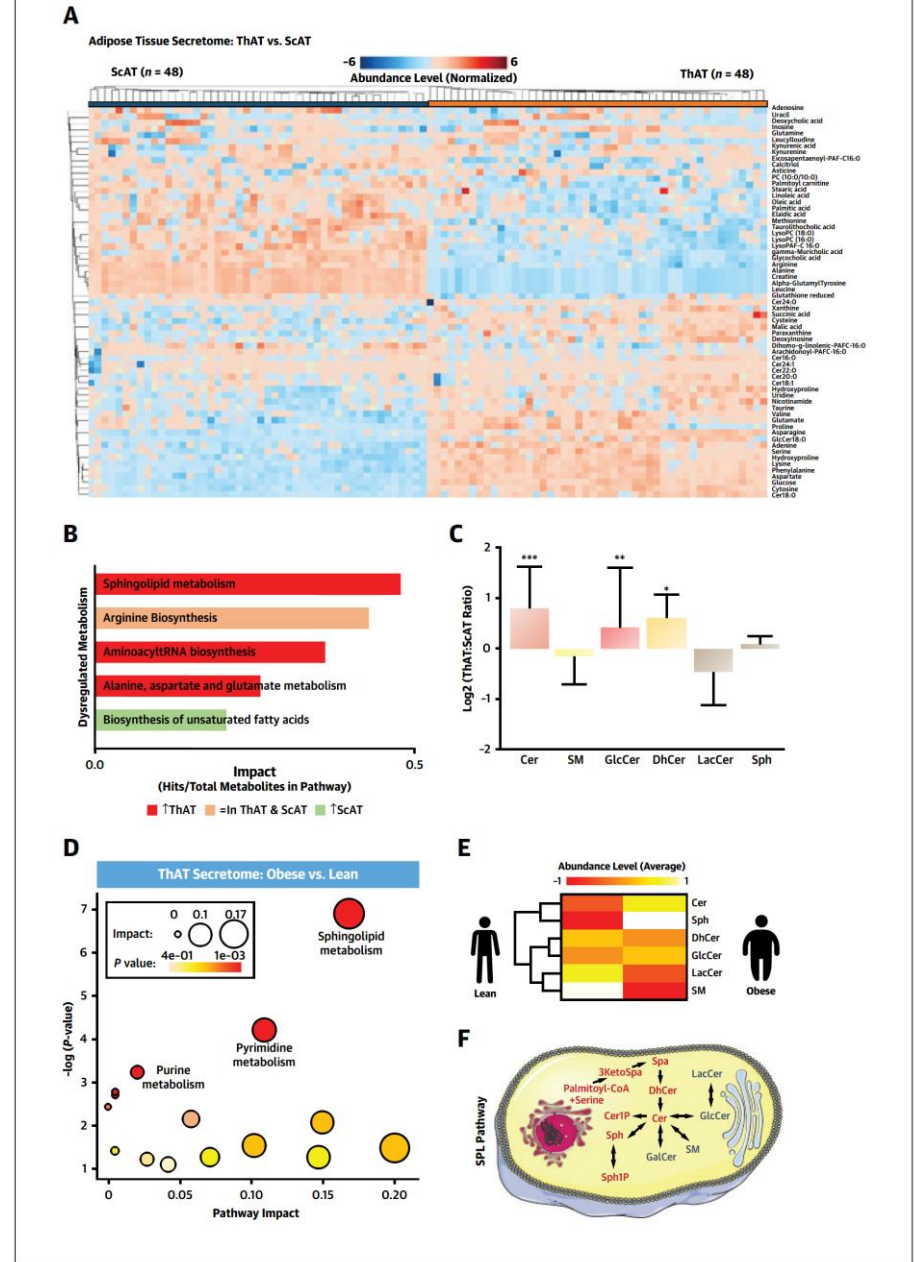Antonopoulos AS et al. Circ Res 2016

Akoumianakis et al Science Transl Med 2020

Discovering new endocrine signals from the AT to the vascular wall?

Akoumianakis & Antoniades Cardiovasc Res 2017

Akawi et al; J Am Coll Cardiol 2021;77:2494–513

Could ceramides drive vascular disease? Which ceramide species?

E

	1	2	3	4	5	
Ponceau						
PERK						<140 kDa
ALIX						<96 kDa
TSG101						<48 kDa
ATP5A						<53 kDa
CD9						<28 kDa
Histone H3						<17 kDa

1 = Media only
2 = Media only supernatant
3 = Adipocytes EV

F 200 nm

G Adipocyte-Derived Ceramides (nmol/l)
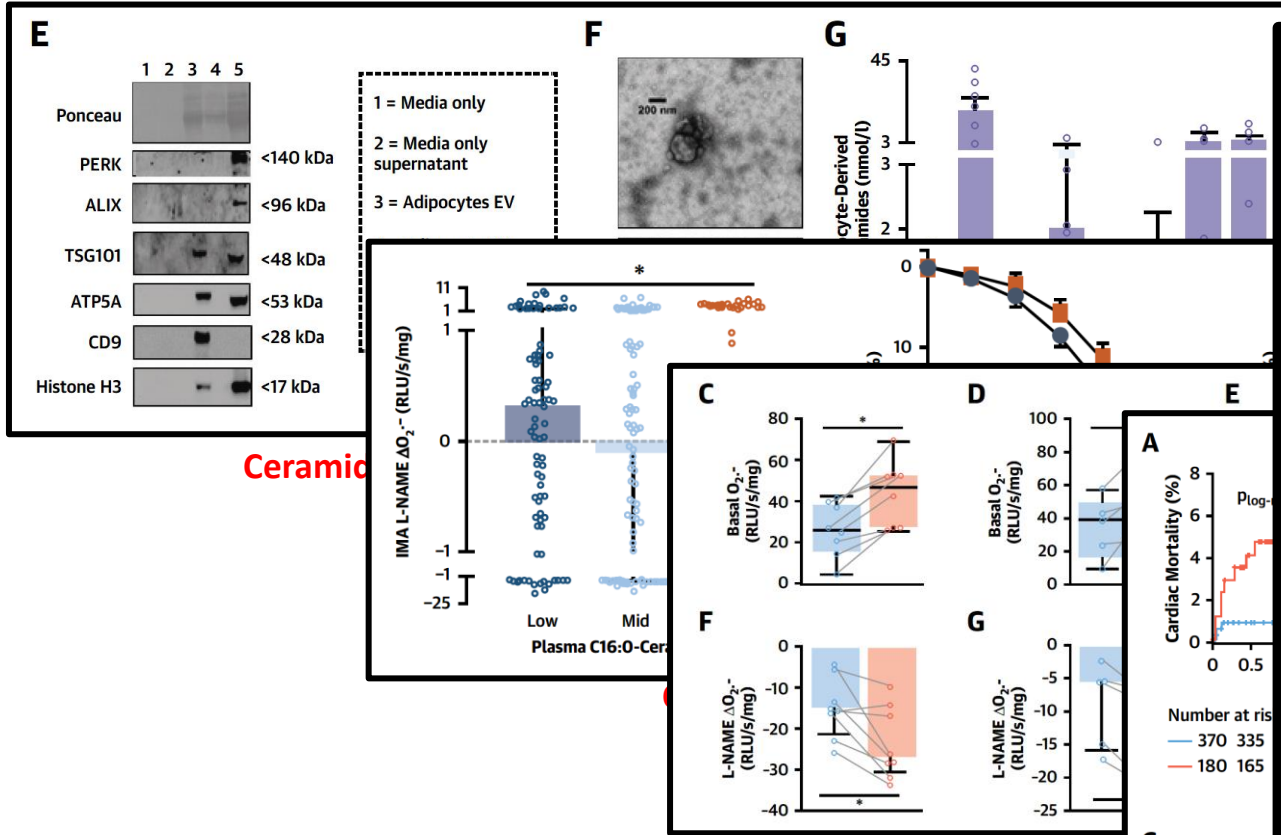
Ceramid...

Exogenous C16 drives endoth...

Association of Ceramides with Cardiac and Non-Cardiac Mortality

Metabolite	Cardiac Mortality Adjusted HR (95% CI)*, p	Non-Cardiac Mortality Adjusted HR (95% CI)*, p
Cer16:0	1.39 (1.03-1.89), 0.031	1.00 (0.67-1.51), 0.986
GlcCer16:0	1.60 (1.04-2.44), 0.032	0.60 (0.34-1.06), 0.078
Cer16:0/Cer22:0	2.10 (1.45-3.03), <0.0001	1.18 (0.82-1.70), 0.372
Cer16:0/Cer24:0	1.87 (1.46-2.39), <0.0001	1.29 (0.91-1.83), 0.161

*Presented per-1SD metabolite change, adjusted for age, gender, and smoking

Ceramide C16 is a strong predictor of cardiac mortality

Thoracic Adipose Tissue

Cer16:0

↑Circulating Cer16:0 — Liraglutide ?

Systemic Circulation

↑PP2A activity   ↓BH4 bioavailability

↓eNOS coupling & activity   ↑O$_2^-$

↓Vasorelaxation

Vascular Endothelium

Clinical Associations:
• Increased systemic inflammation
• Increased oxidative stress
• Reduced vascular relaxation
• Increased risk of cardiac death

Adipocytes   Adipocytes-secreted EV   Immune cells   Microvessels

Akawi et al; J Am Coll Cardiol 2021;77:2494–513

Discovering new paracrine signals from PVAT to the vascular wall?

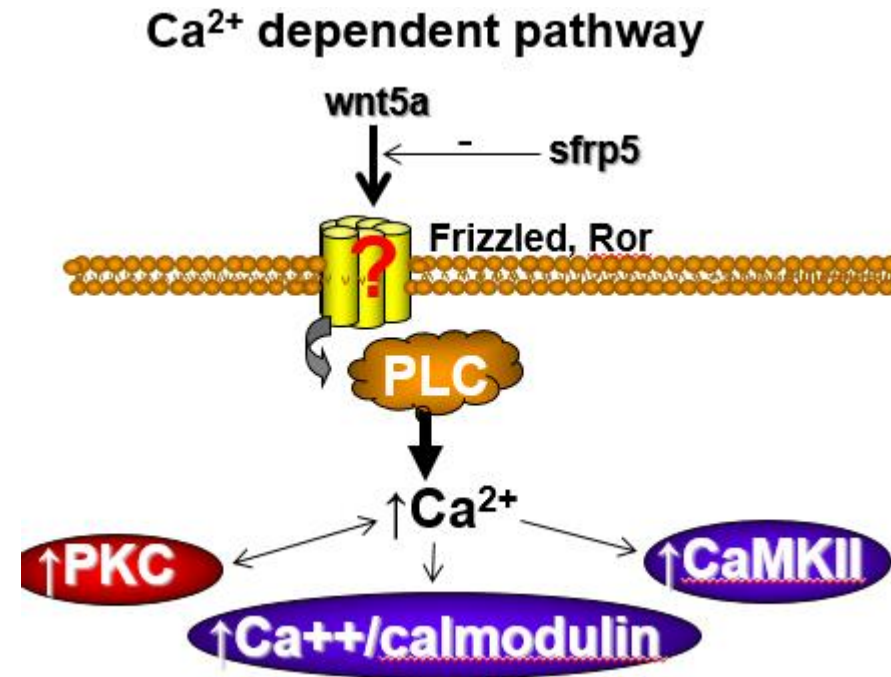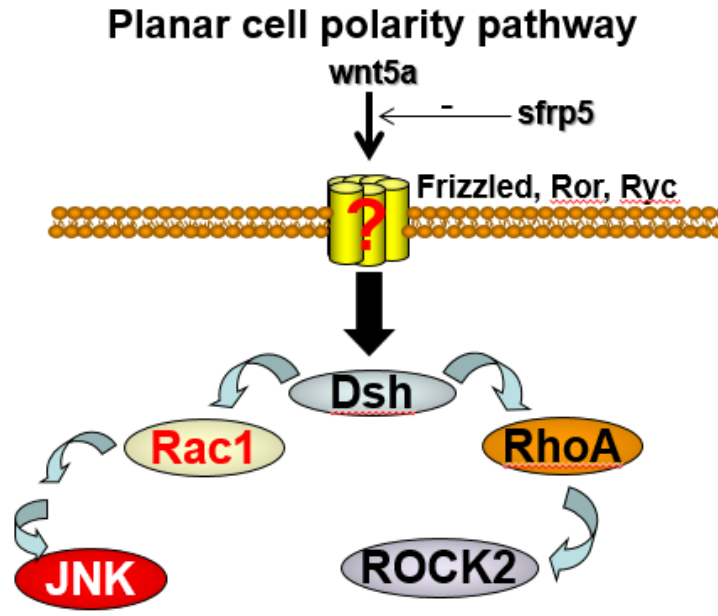# Discovering new paracrine signals from PVAT to the vascular wall?
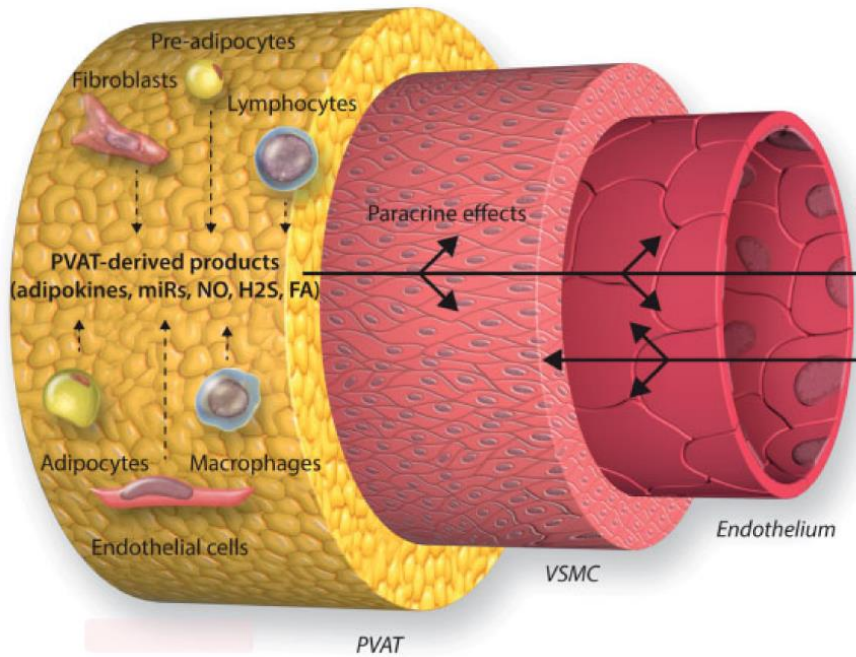
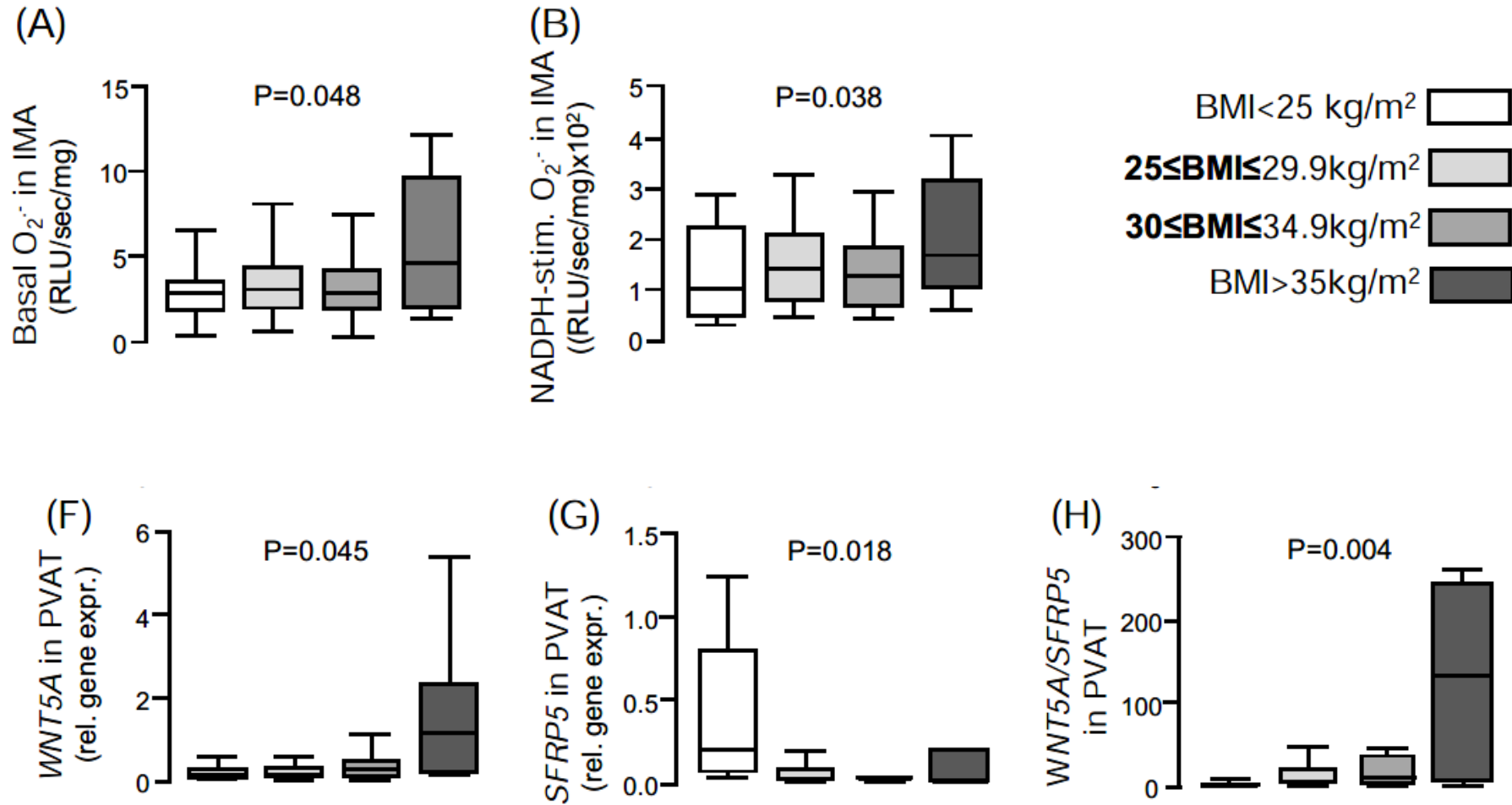Akoumianakis & Antoniades Cardiovasc Res 2017
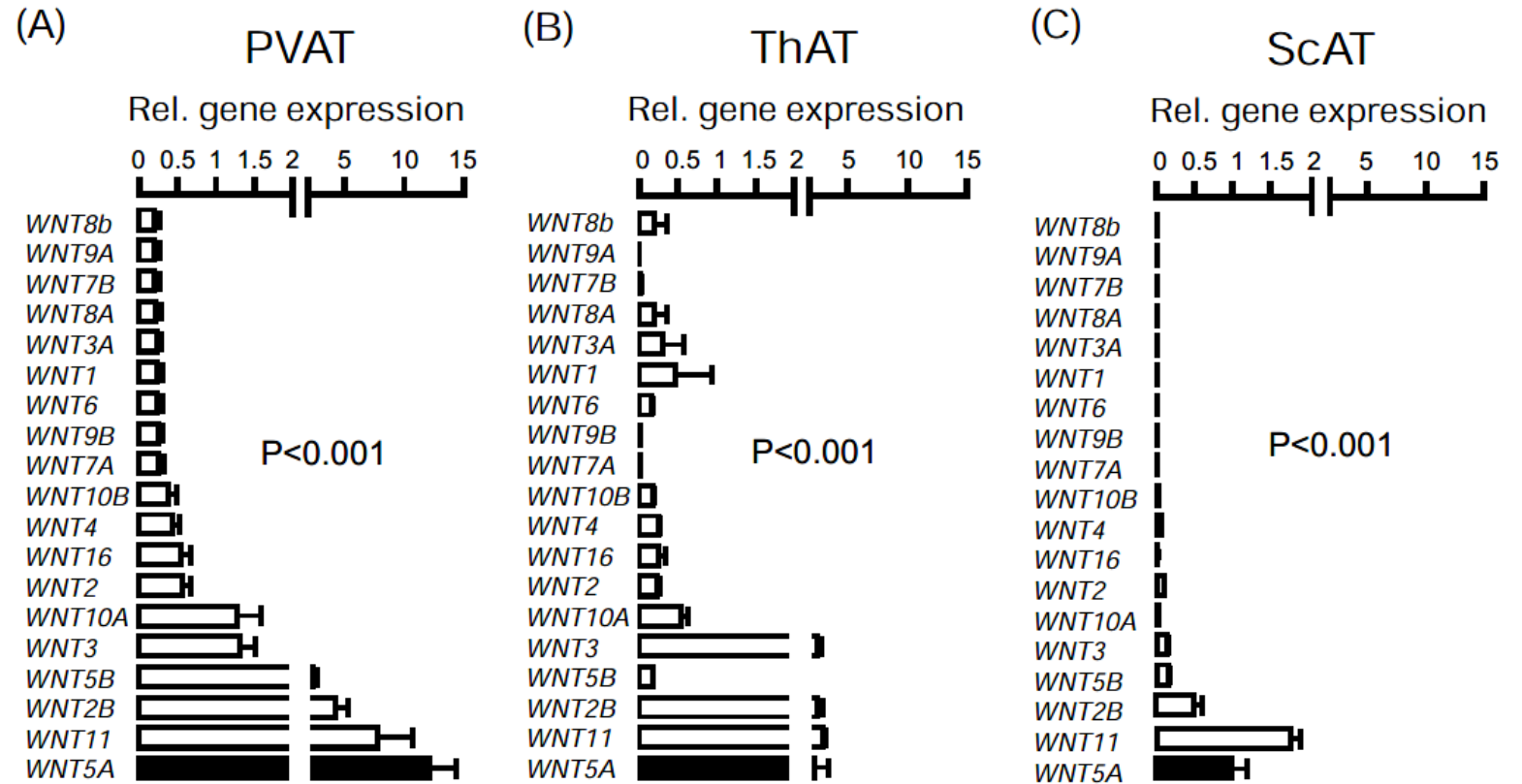
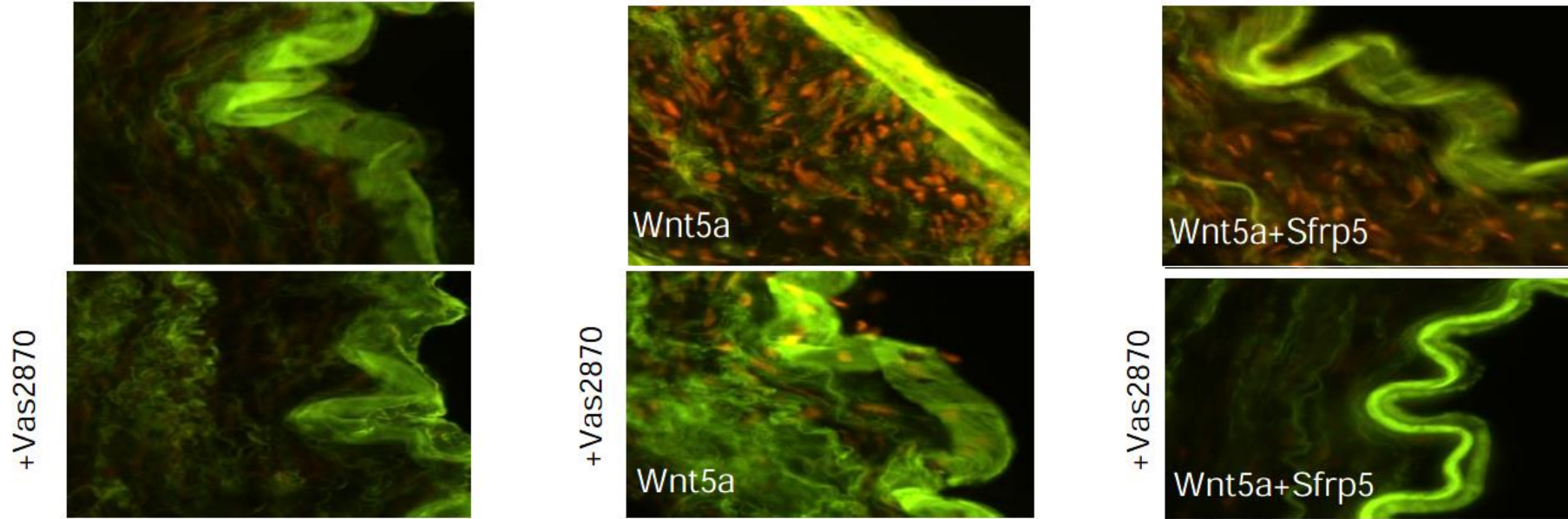Akoumianakis I et al; Science Transl Med 2019; 11(510):5055

What is wnt5a?

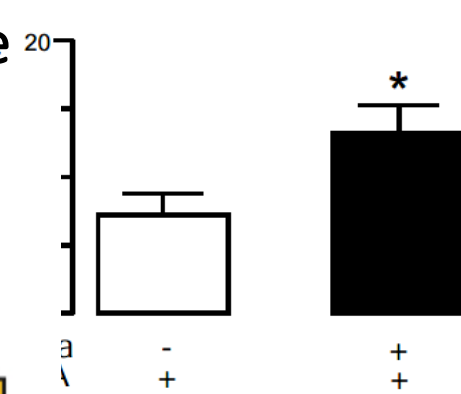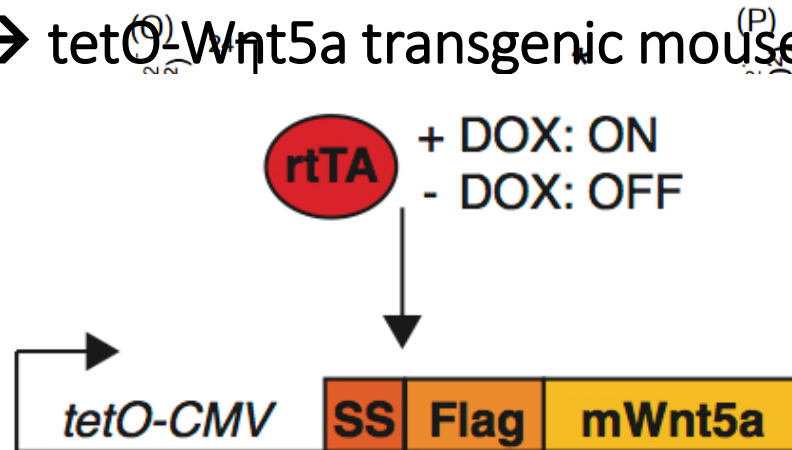# Could wnt5a/sfrp5 mediate the obesity-related vascular oxidative stress?

Human arteries

In vivo mouse model

Is this valid in vivo? → tetO-Wnt5a transgenic mouse

Akoumianakis I et al; Science Transl Med 2019; 11(510):5055

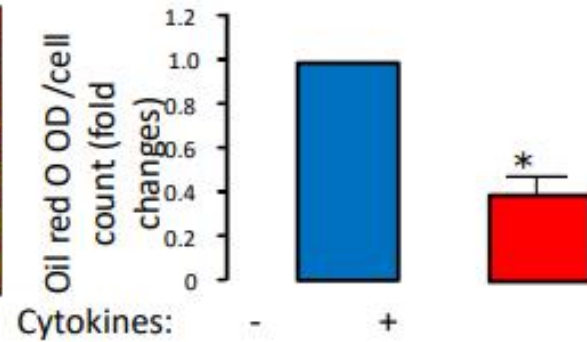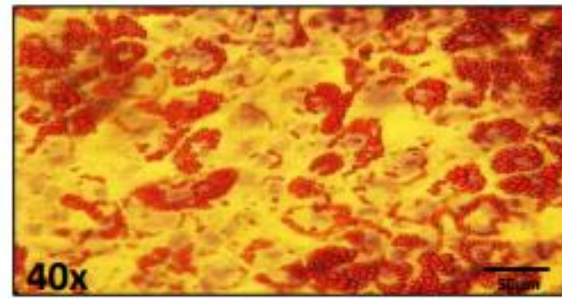# Exploring new concepts:
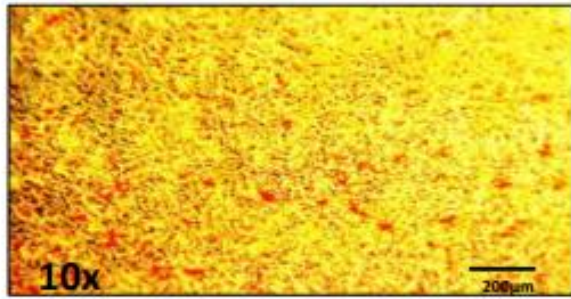# Does PVAT always affect the vascular wall, or….?
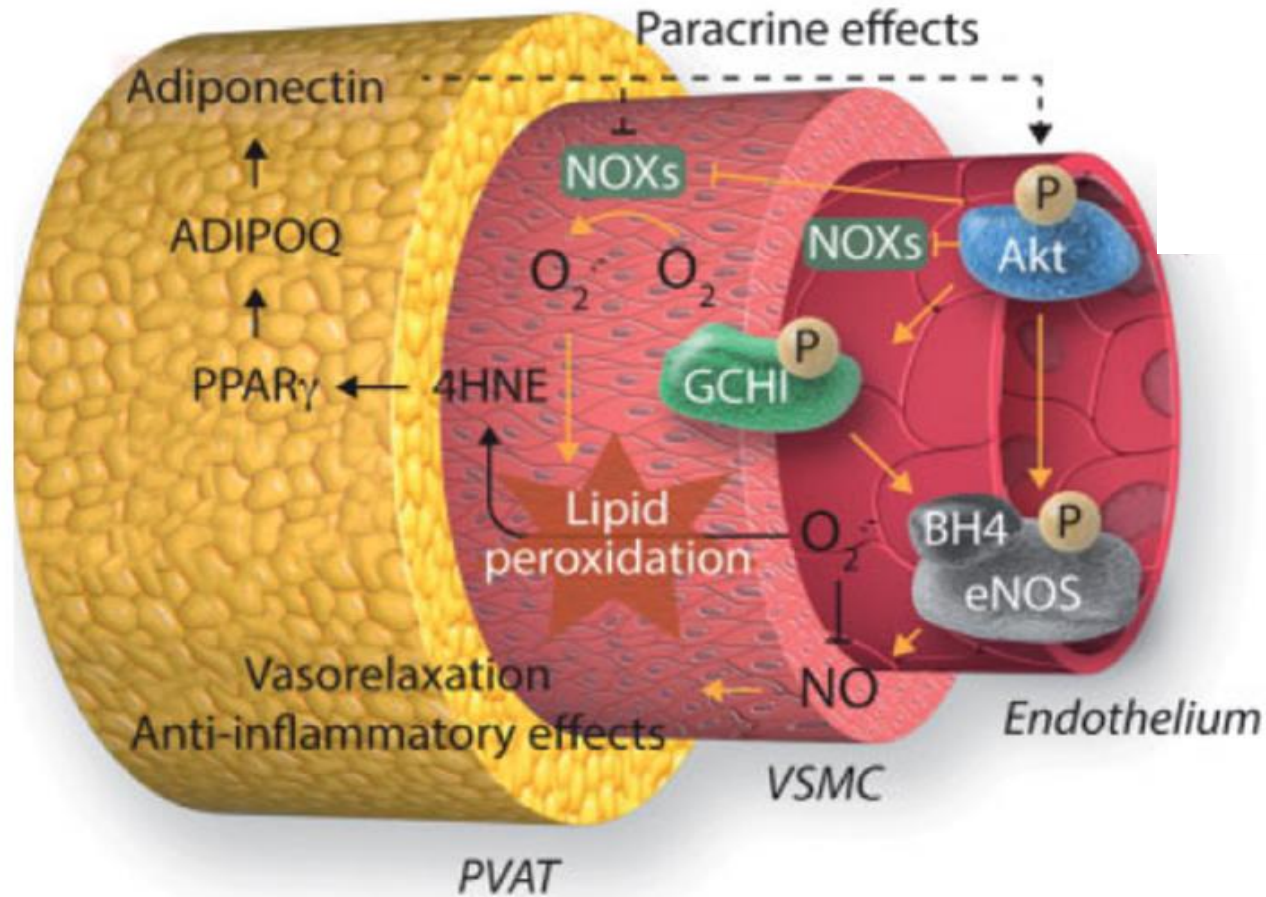
New concept:
The Inside-to-Outside Signalling from the vascular wall to PVAT
(local defence mechanism of human vessels)

Antonopoulos A et al; Circ Res 2016;118:842-55
Margaritis et al; Circulation 2013;127:2209-21
Antonopoulos A et al Diabetes 2015 64:2207-19
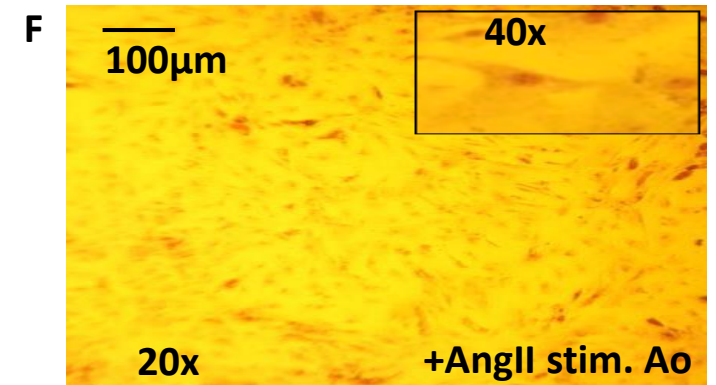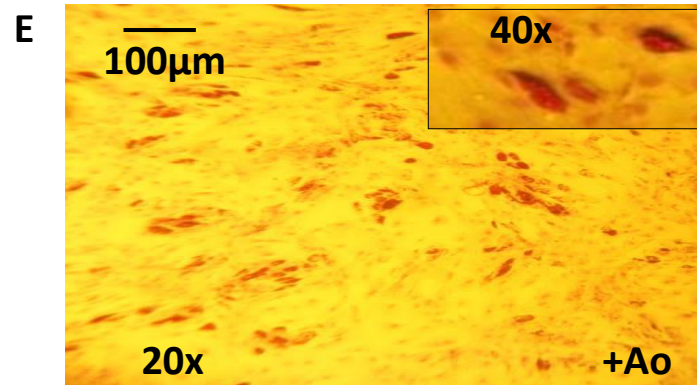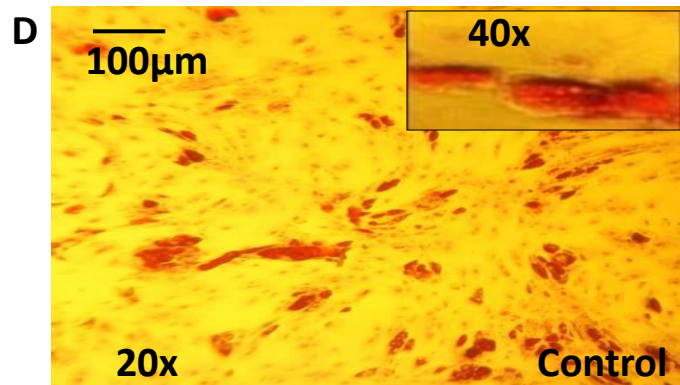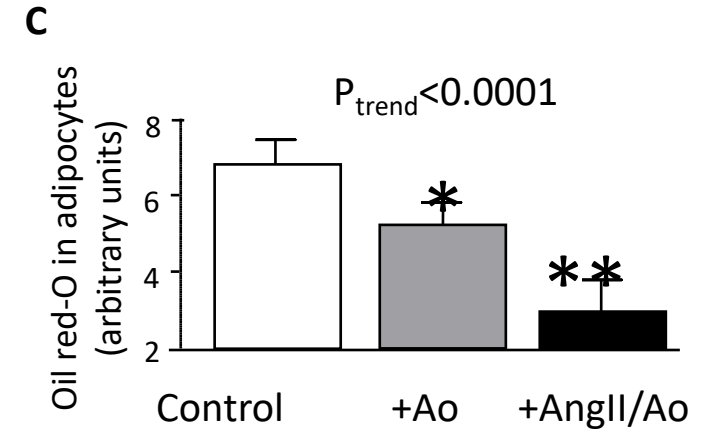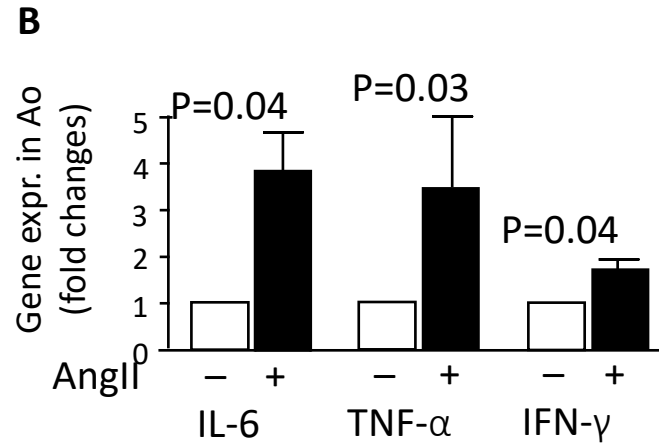Akoumianakis & Antoniades Cardiovasc Res 2017

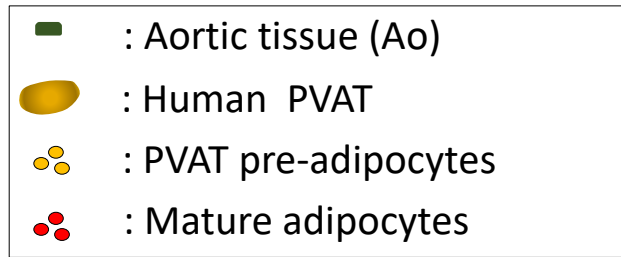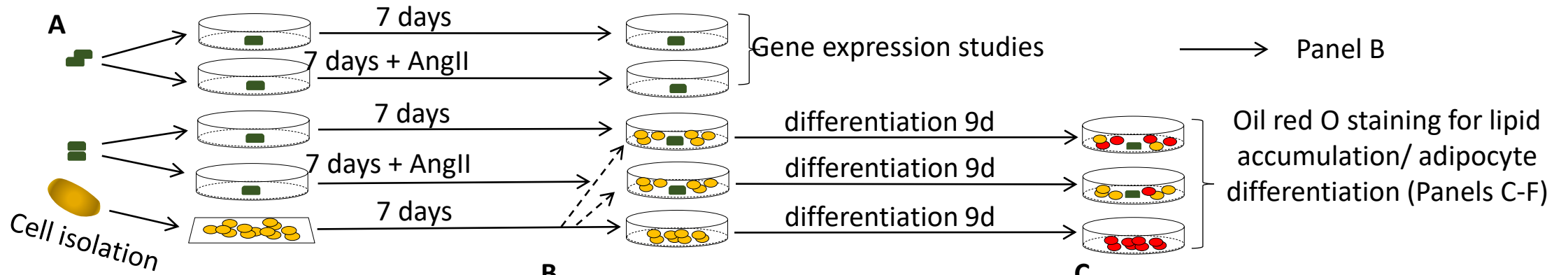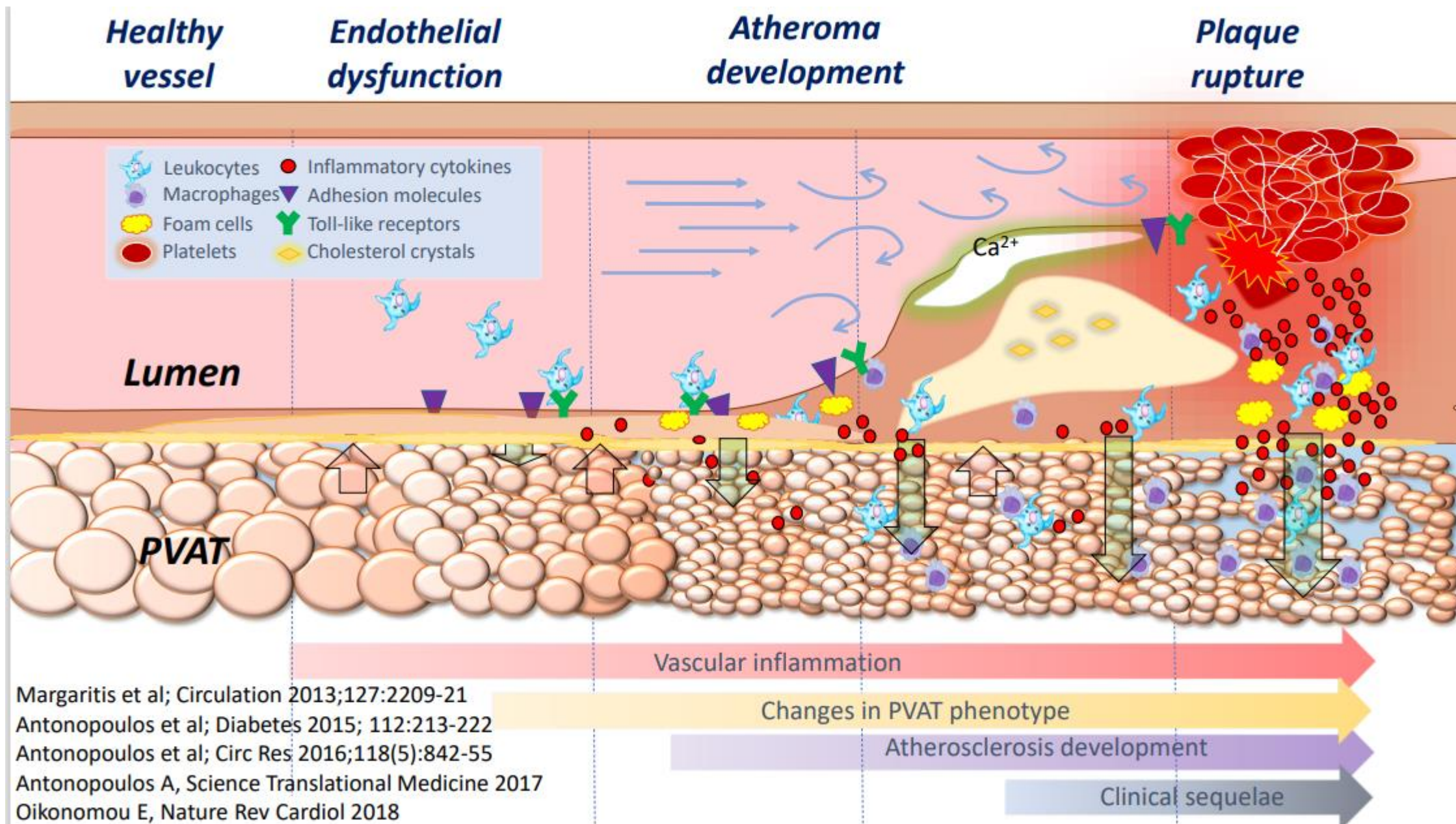# Chasing the chicken and the egg in clinical association studies.....
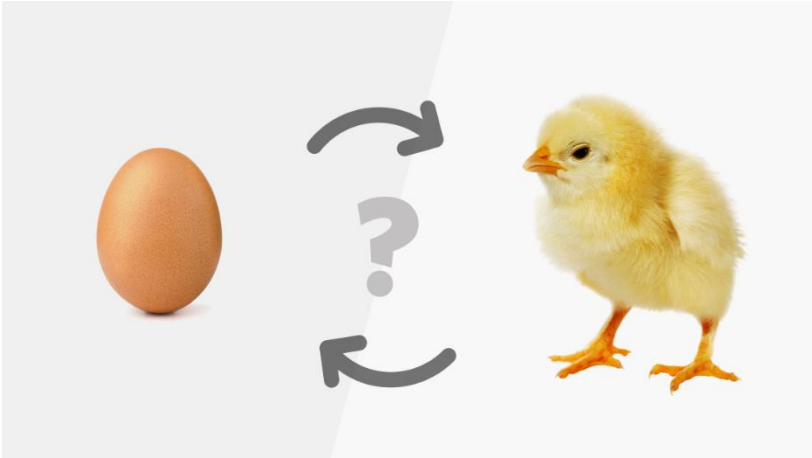
Inflammation inhibits adipocyte differentiation

Antonopoulos et al., Science Translational Medicine *2017*
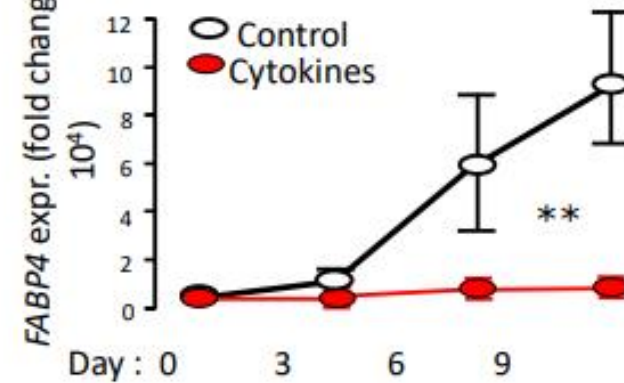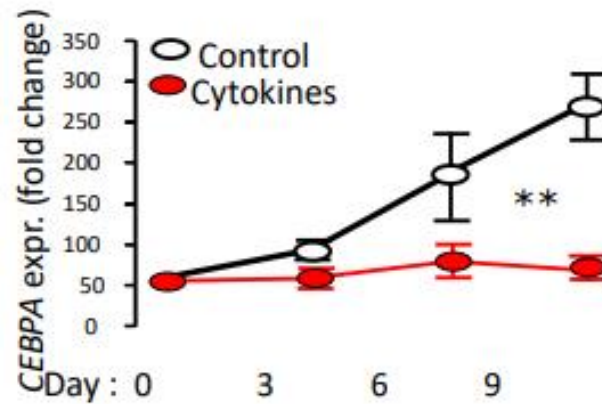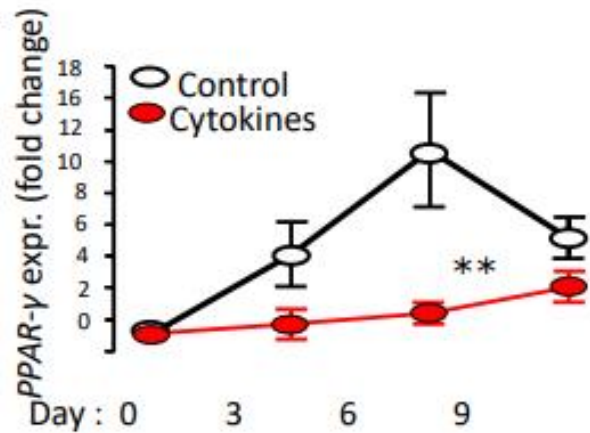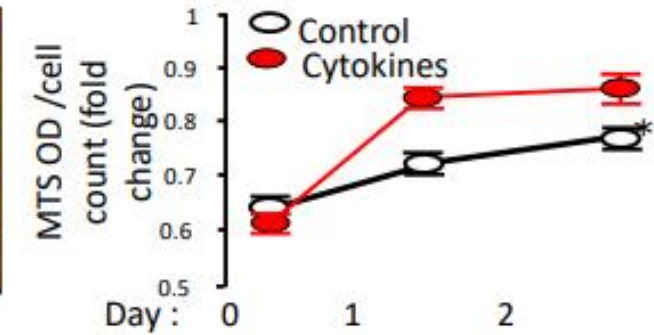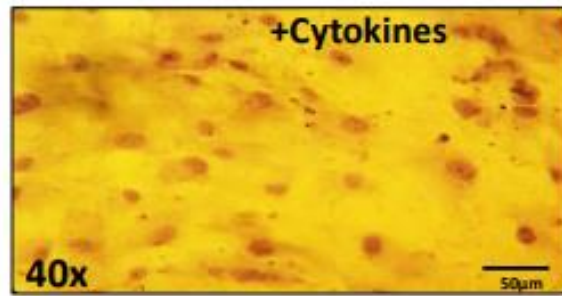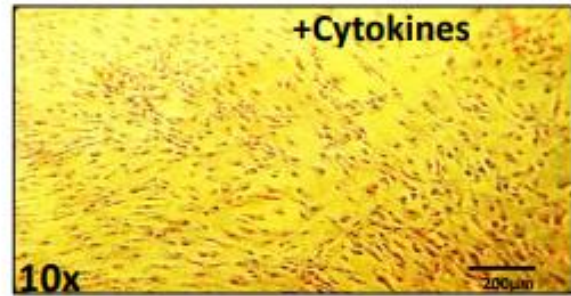
Effects of inflammation on adipocytes

A

7 days → Gene expression studies → Panel B

7 days + AngII → Gene expression studies

Cell isolation

7 days → differentiation 9d → Oil red O staining for lipid accumulation/ adipocyte differentiation (Panels C-F)

7 days + AngII → differentiation 9d

7 days → differentiation 9d

Legend:
- : Aortic tissue (Ao)
- : Human PVAT
- : PVAT pre-adipocytes
- : Mature adipocytes

B

Gene expr. in Ao (fold changes)

P=0.04 (IL-6)
P=0.03 (TNF-α)
P=0.04 (IFN-γ)

AngII: − + − + − +

IL-6   TNF-α   IFN-γ

C

Oil red-O in adipocytes (arbitrary units)

$P_{trend} < 0.0001$

*

**

Control   +Ao   +AngII/Ao

D 100μm 40x 20x Control

E 100μm 40x 20x +Ao
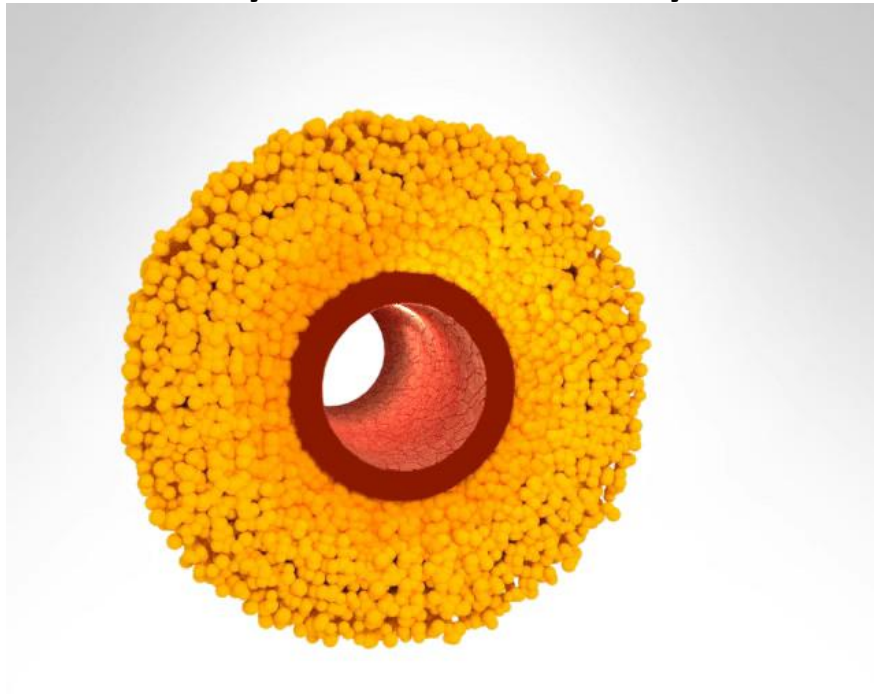
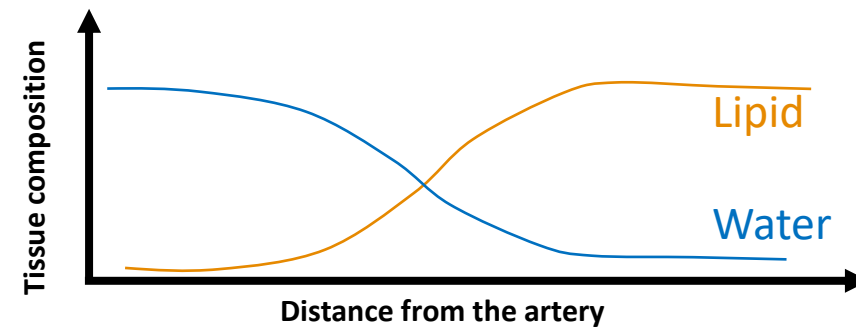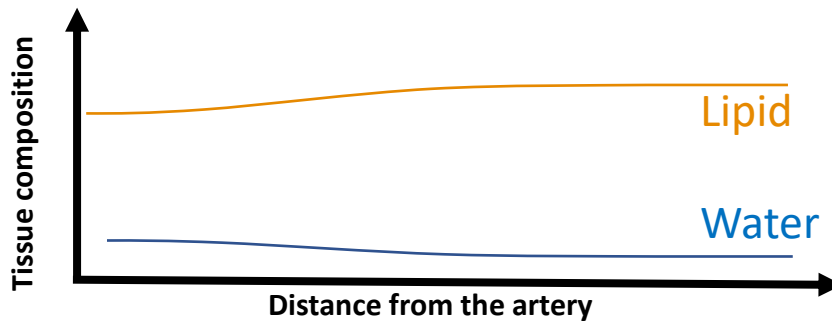**F** 100μm 40x 20x +AngII stim. Ao

The role of PVAT in atherogenesis

Perivascular FAI: a "sensor" of vascular inflammation

Healthy, non-inflamed artery

"Healthy," inflamed artery

✓ ↓ **Adipogenesis**
✓ ↑**Lipolysis**
✓ ↑**Oedema**

Tissue composition
Lipid
Water
Distance from the artery

Tissue composition
Lipid
Water
Distance from the artery
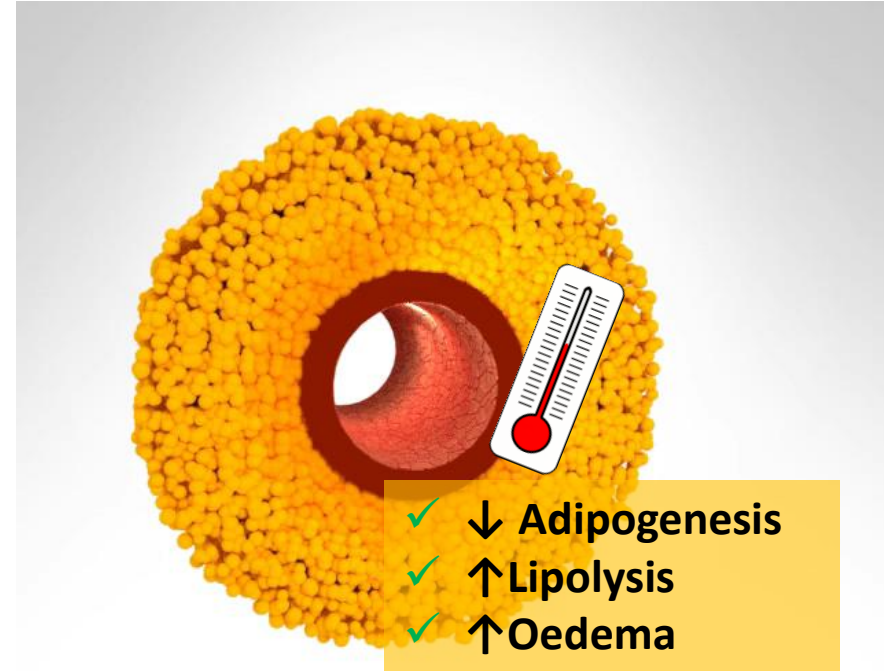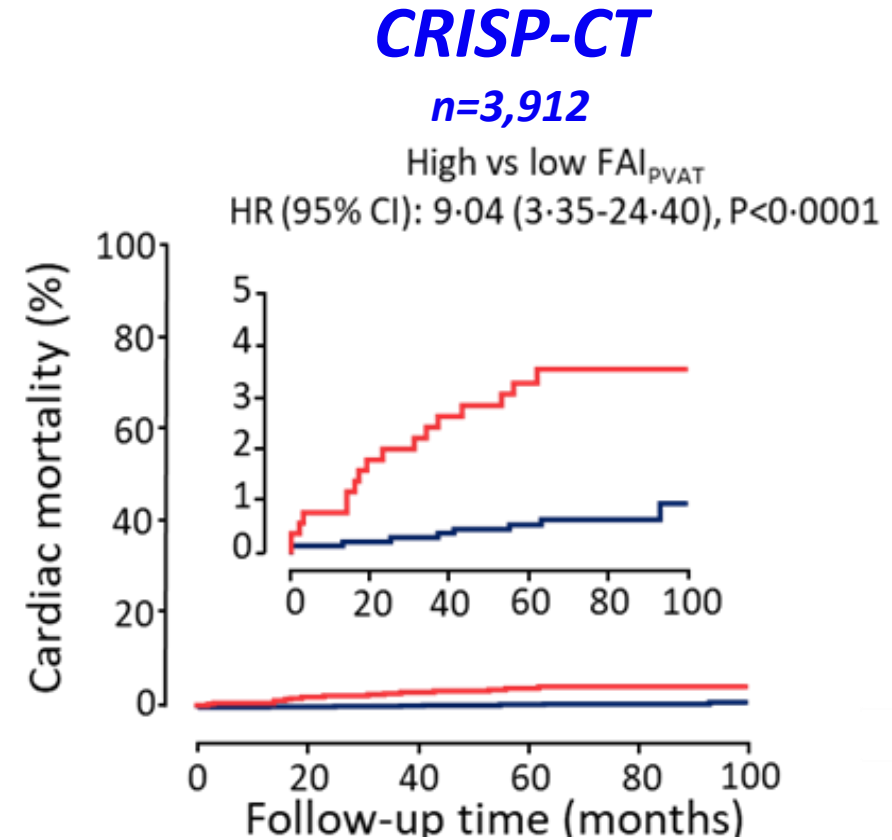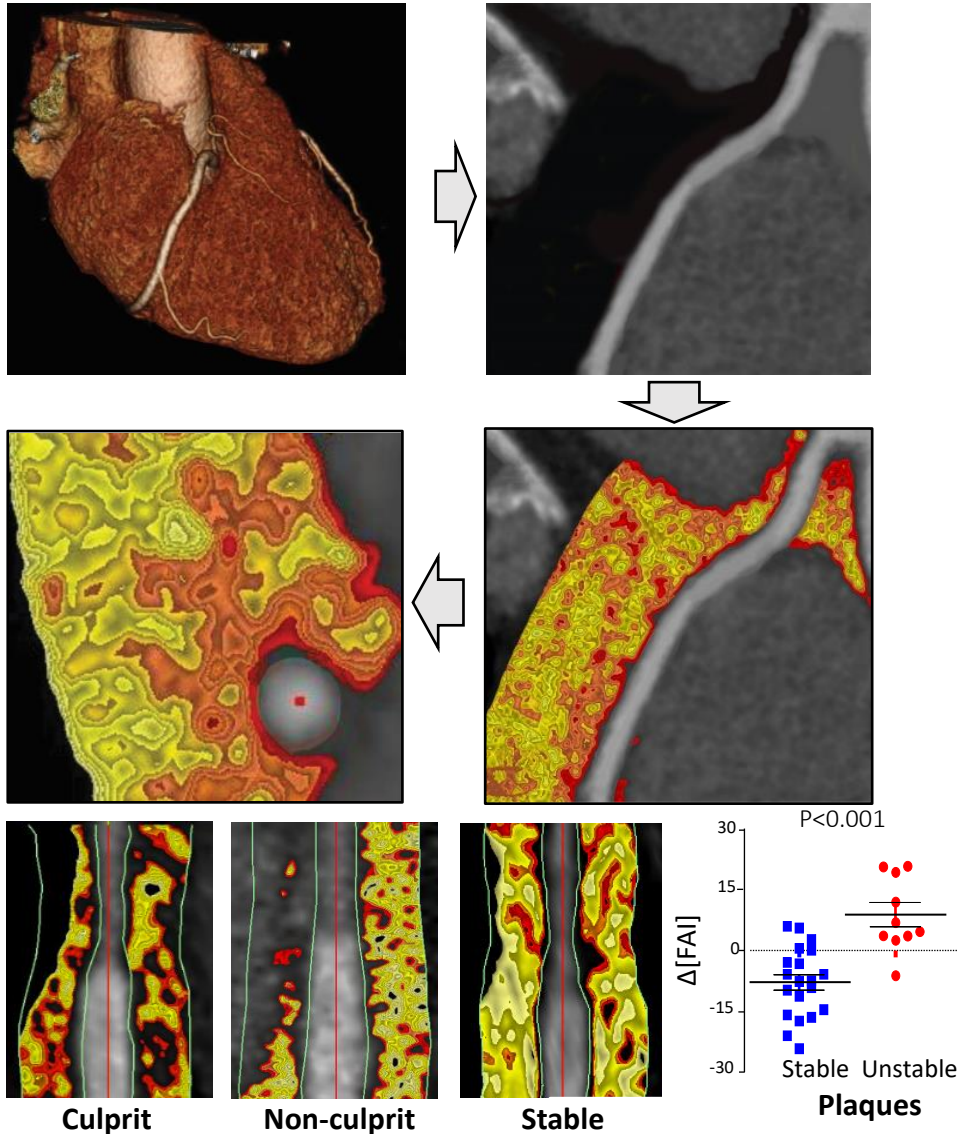
Antonopoulos et al., Science Translational Medicine *2017*
Oikonomou & Antoniades. Nature Rev Cardiol 2018

Margaritis et al; Circulation 2013;127:2209-21
Antonopoulos et al; Diabetes 2015; 112:213-222
Antonopoulos et al; Circ Res 2016;118(5):842-55

Pericoronary fat imaging as a sensor of vascular inflammation

Culprit   Non-culprit   Stable

CRISP-CT

n=3,912

High vs low FAI_PVAT

HR (95% CI): 9·04 (3·35-24·40), P<0·0001

FAI is an independent predictor of cardiac mortality
on top of CV risk factors, calcium score,
extent of coronary atherosclerosis
& presence of high risk plaques on CCTA

Antonopoulos AS et al. Science TM 2017

Oikonomou EK et al. Lancet 2018

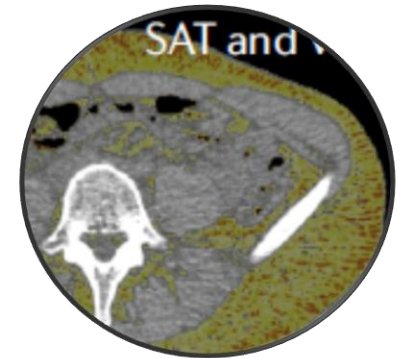# What more can we do to identify dysfunctional adipose tissue?

Coupling imaging with biology
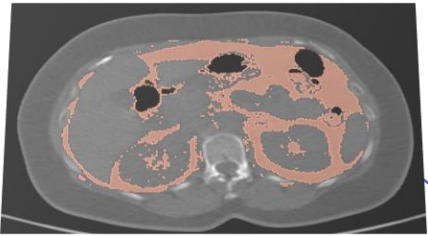
AT proteome/secretome

&
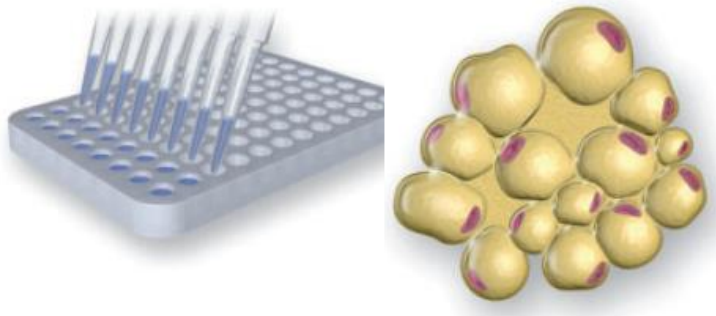
AT imaging

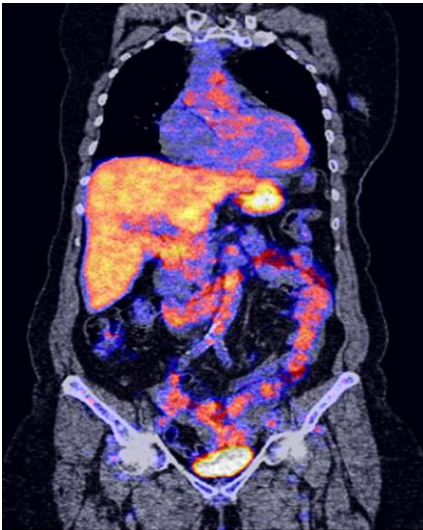AT imaging and adipokine profile determine the levels of arterial inflammation
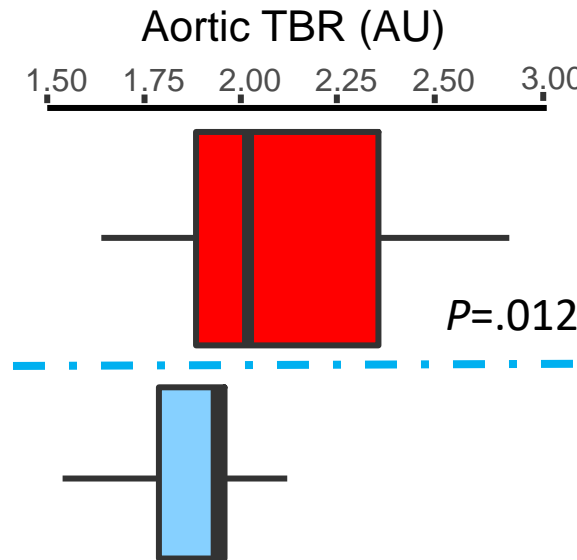
Plasma

CT

18-FDG PET

Adipokine profile
VAT volume
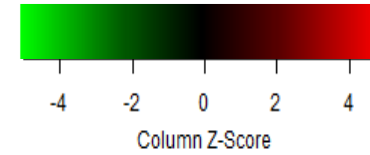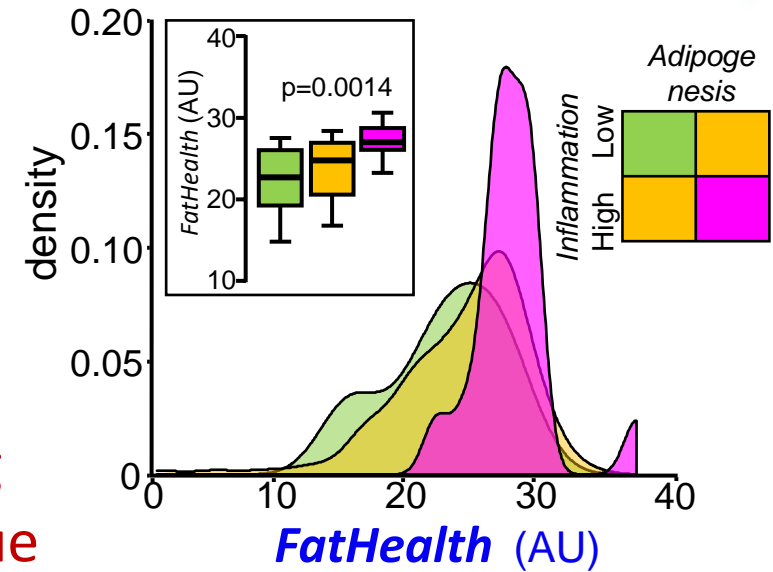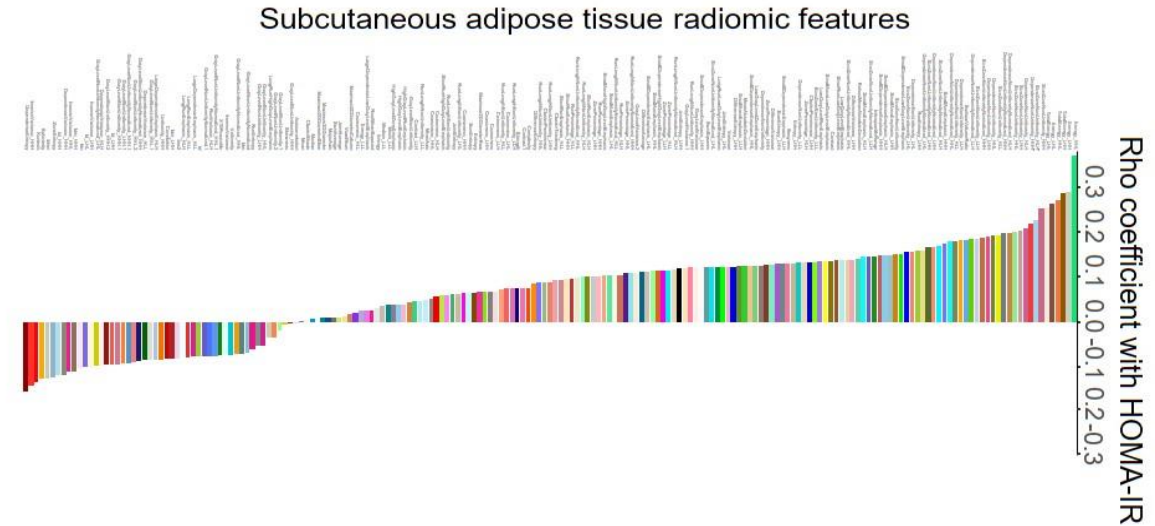VAT & aortic inflammation

Column Z-Score
-4 -2 0 2 4

Cluster A

Cluster B

VAT TBR   VAT volume   Plasma AdN

Aortic TBR (AU)
1.50 1.75 2.00 2.25 2.50 3.00

P=.012
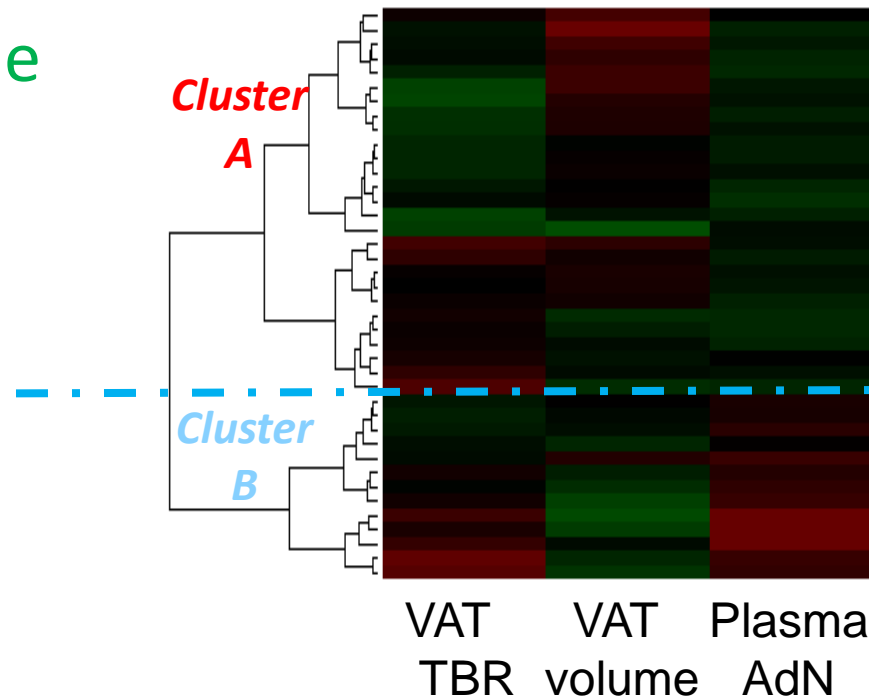
Big-data extraction from medical images and machine learning for dysfunctional adipose tissue

Distance

Subcutaneous adipose tissue radiomic features

Rho coefficient with HOMA-IR
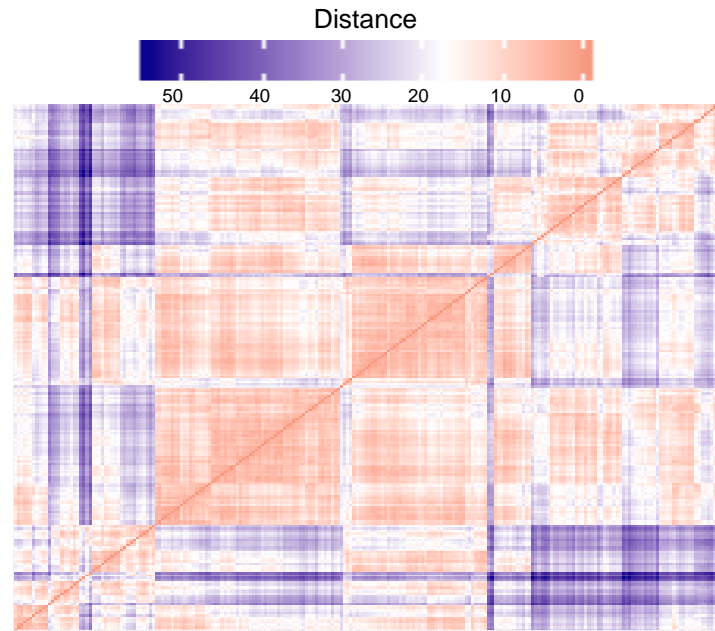
Imaging features & machine learning
to identify dysfunctional adipose tissue

density

FatHealth (AU)

p=0.0014

FatHealth (AU)

Adipogenesis

Inflammation
Low High

Antonopoulos AS et al. (WO2020058712A1)

The **FatHealth** Project - subcutaneous fat radiomics to predict DM

Antonopoulos AS et al. (WO2020058712A1)

ESC
European Society
of Cardiology
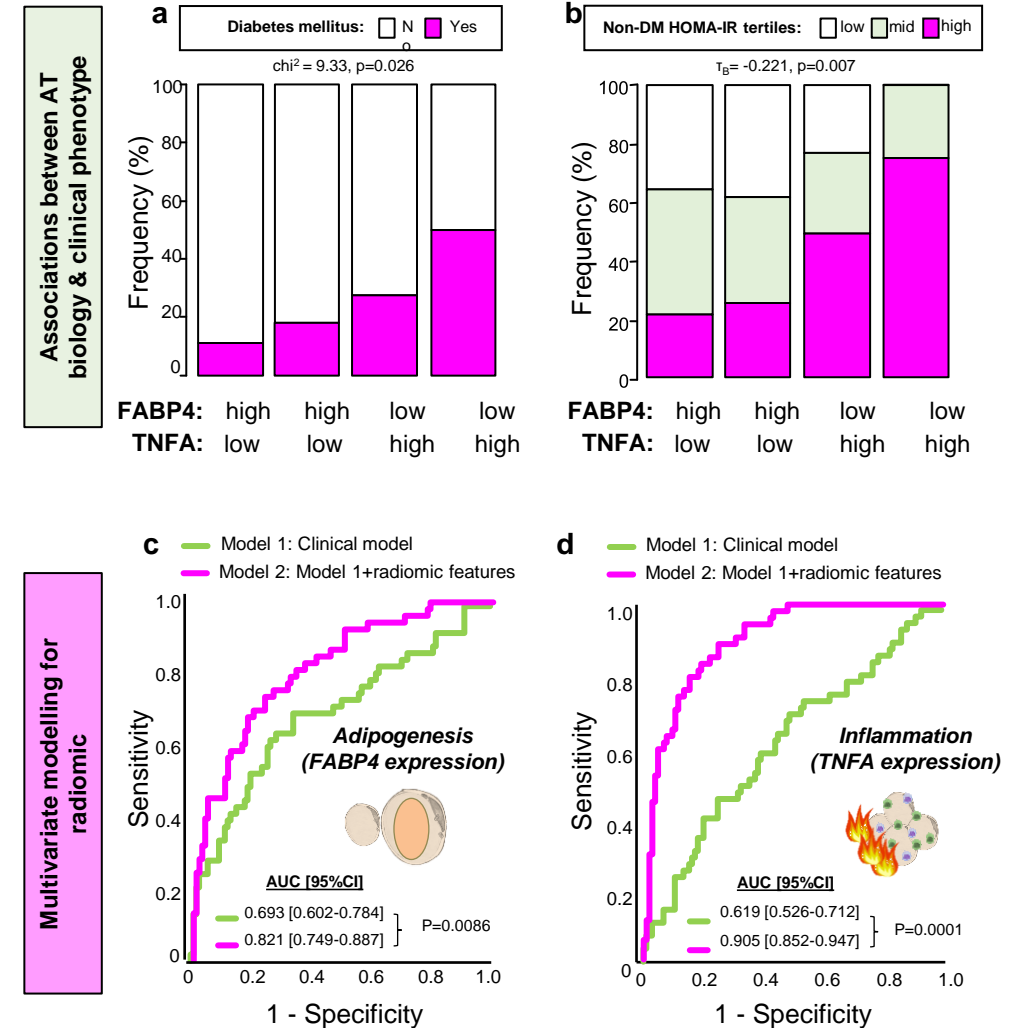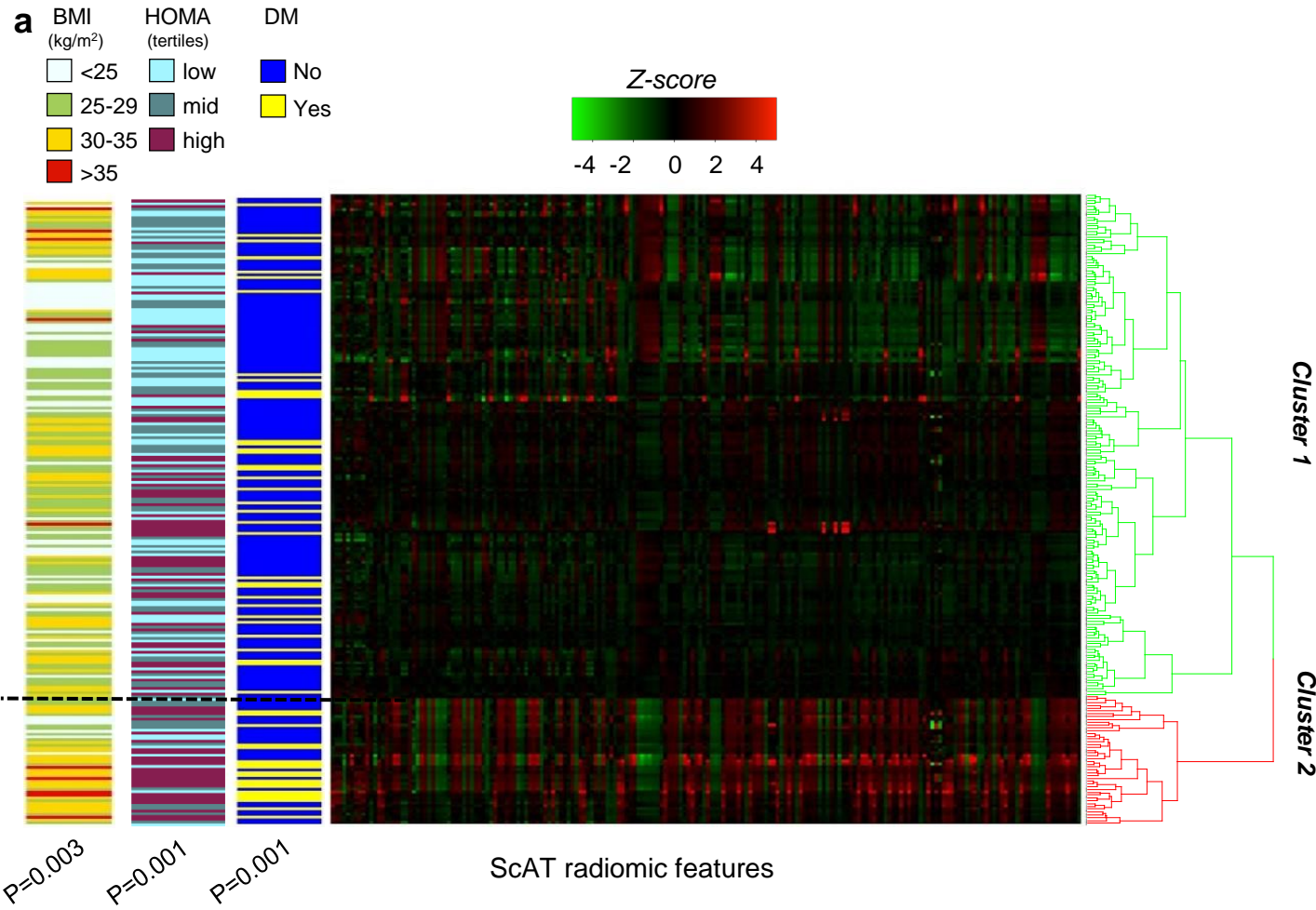
How to assess cardiometabolic risk in obesity?

Obesity diagnosis

Established anthropometric obesity indices

- BMI
- WC

Waist
Hip

Height

Consider secondary indices too

- BF%
- SAD
- Skinfolds

Antonopoulos AS & Tousoulis D. Cardiov Res 2017