May 20th, 1:15 PM - 2:45 PM

Activated Innate Immunity in Childhood: A Novel Treatment Target

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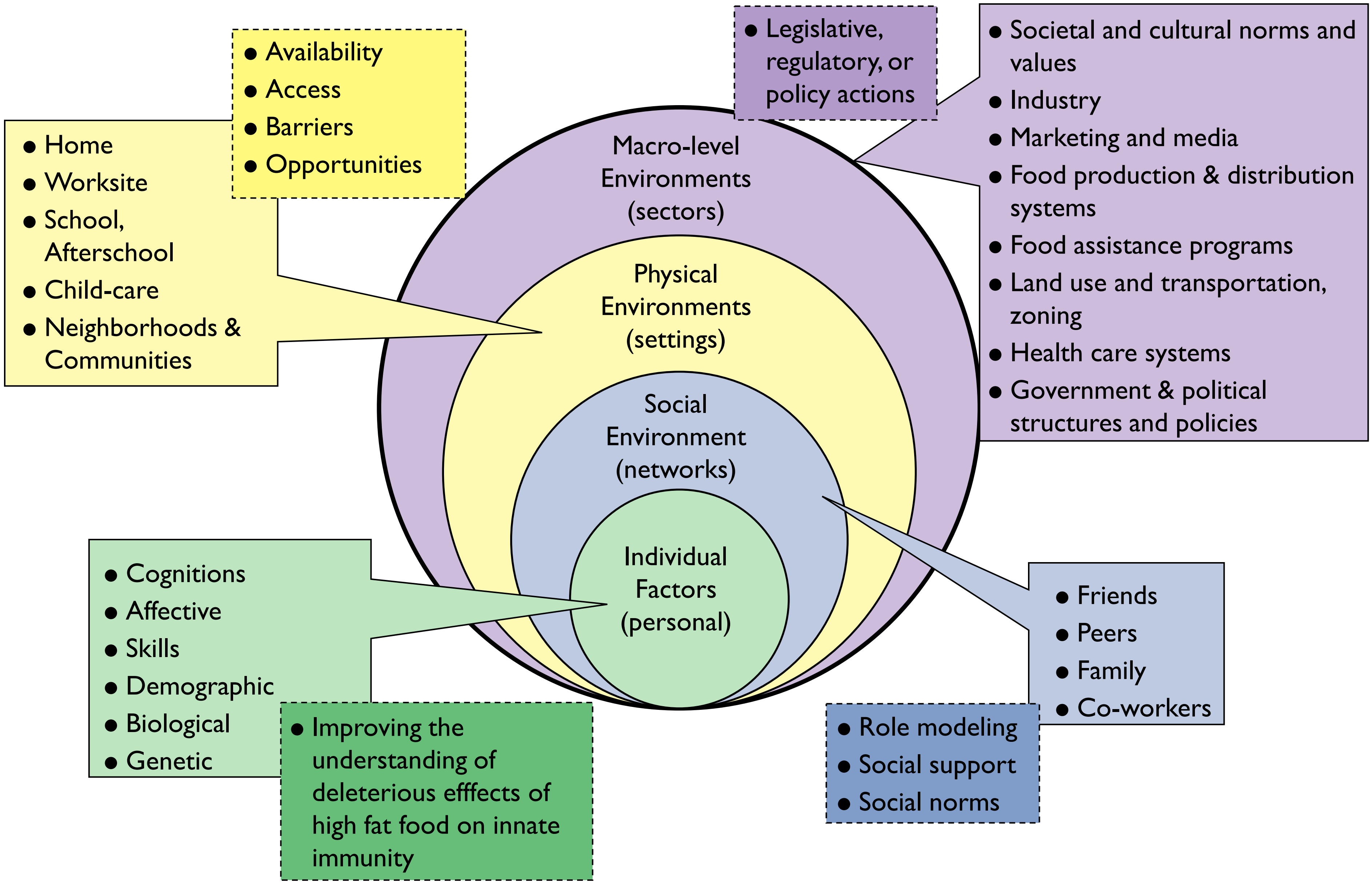
May 20, 2011

DISCLOSURE

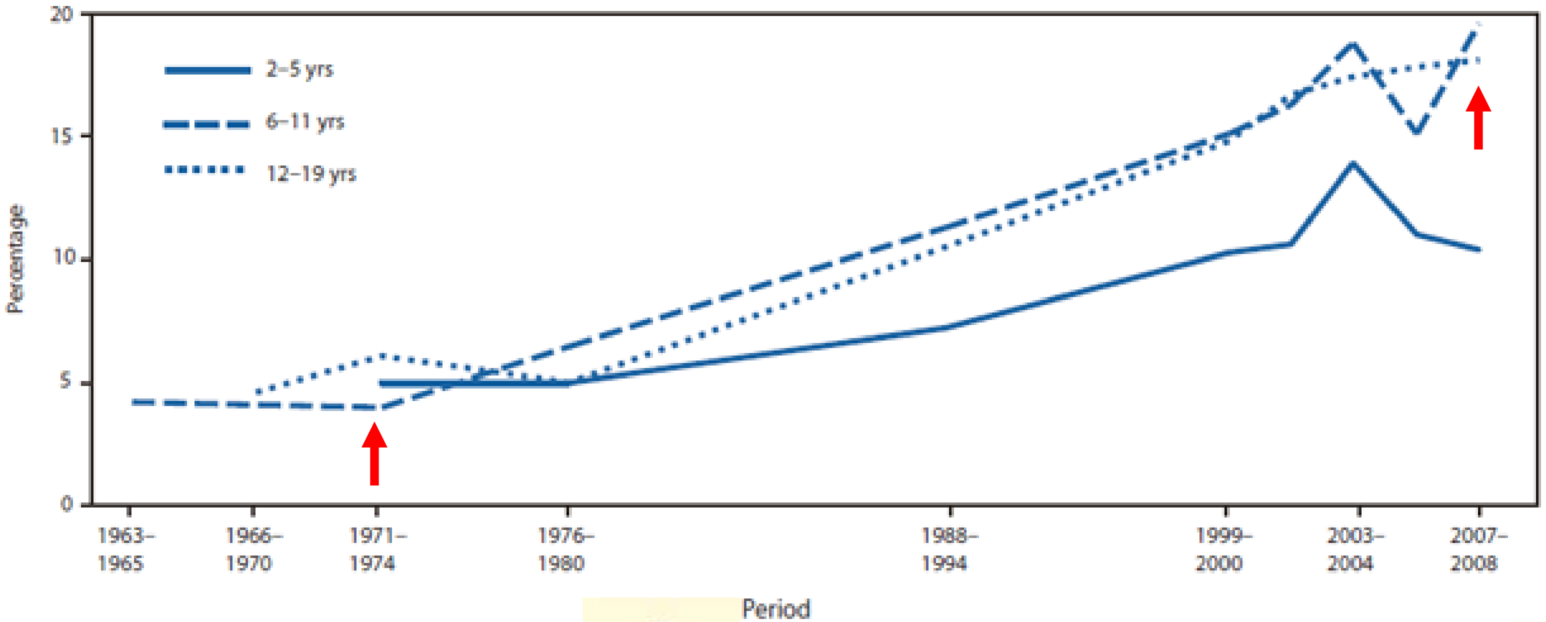
I have no actual or potential conflict of interest in relation to this program or presentation.

An Ecological Framework : Multiple Influences on Physical Activity and Eating Behaviors



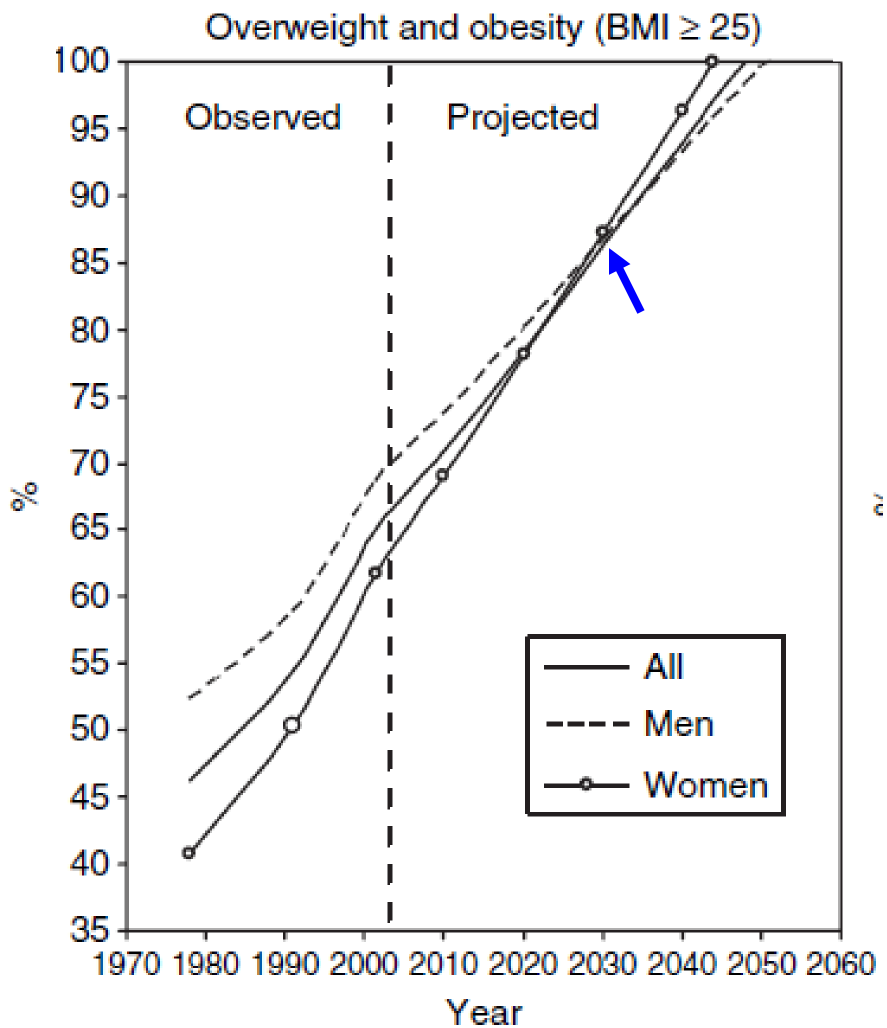
Childhood obesity is an epidemic

Prevalence of obesity among children and adolescents, by age group --- United States, 1963-2008



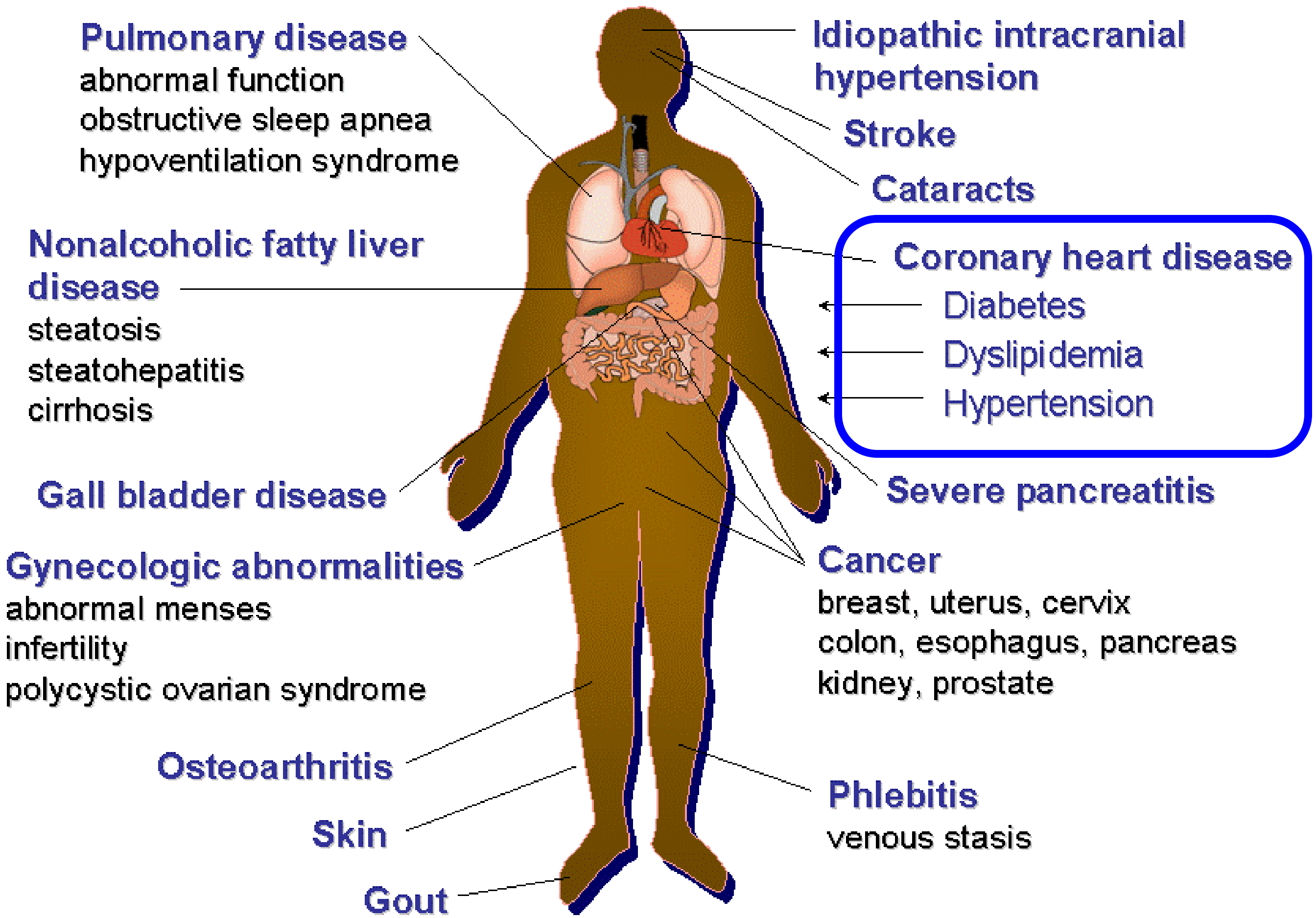
"CDC Grand Rounds: Childhood Obesity in the United States." MMWR: Morbidity and Mortality Weekly Report. January 21 2011. 60(2):42-46.

Based on current trends 86% of U.S. adults will be overweight by 2030



Wang Y, et al. Will all Americans become overweight or obese? estimating the progression and cost of the US obesity epidemic. Obesity. 2008.

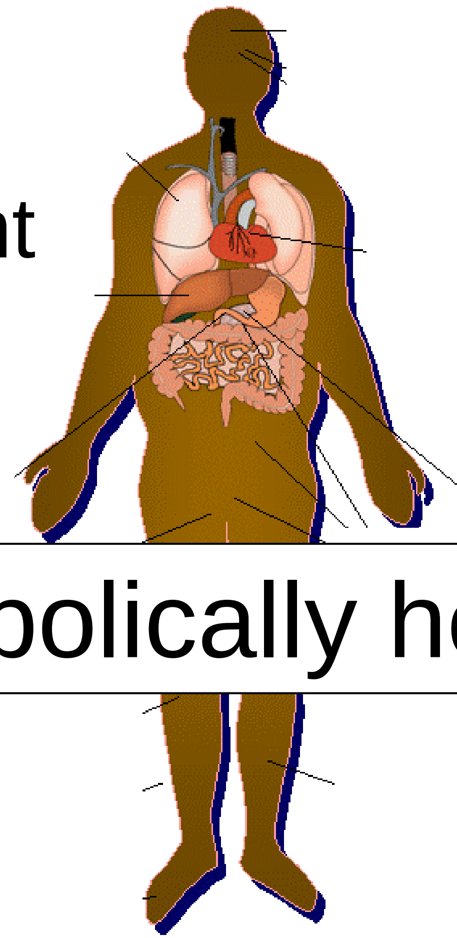
Obesity is a risk factor for numerous medical conditions



www.doctorsweightsolutions.com; www.cdc.gov

However … not all obese individuals develop complications

51% of overweight adults

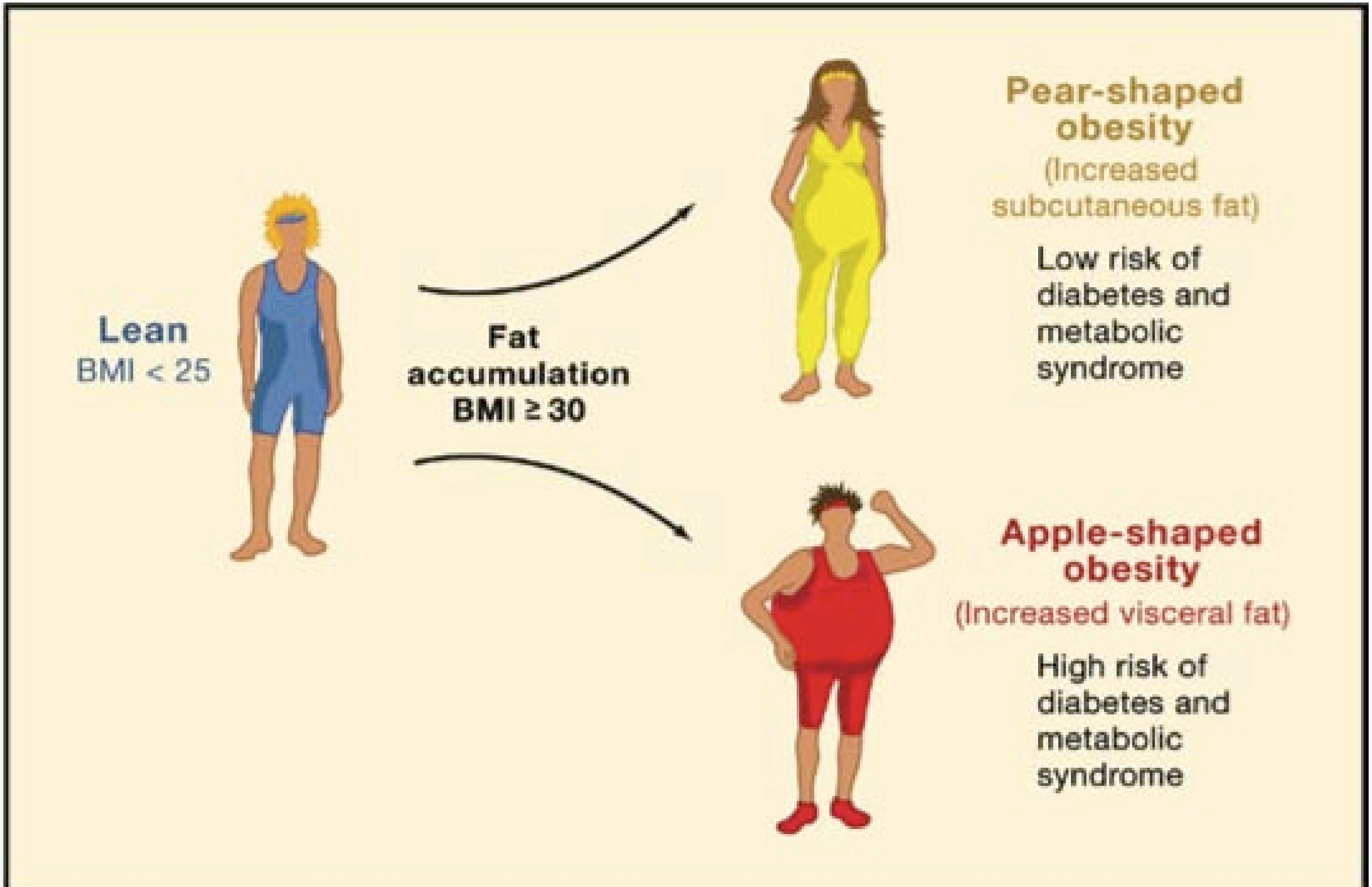


31% of obese adults

Metabolically healthy

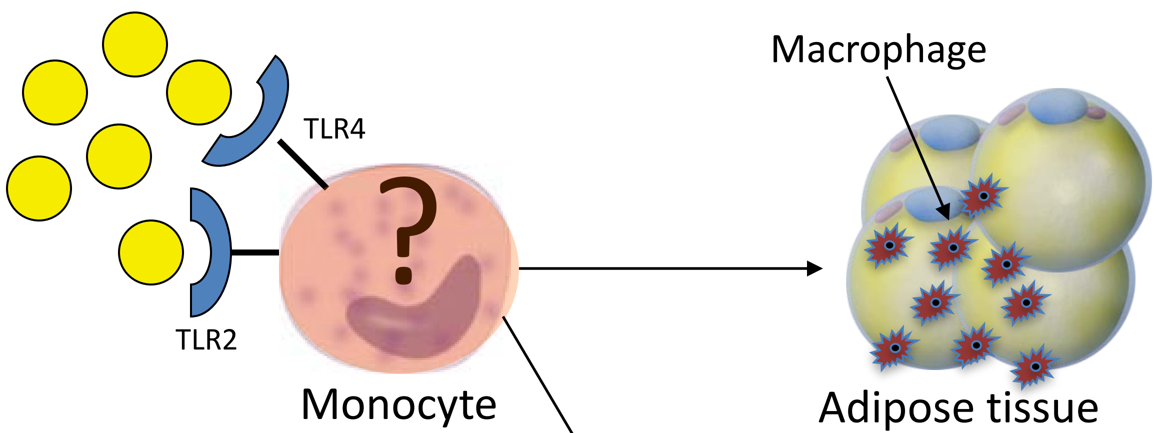
Wildman, et al. The Obese Without Cardiometabolic Risk Factor Clustering and the Normal Weight With Cardiometabolic Risk Factor Clustering, Arch Int Med, Aug 2008.

Increased visceral fat is associated with a high risk of diabetes

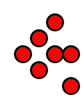


Gesta et al. *Cell* 2007

Monocytes may be a modifiable source of proinflammatory cytokines



IL6, Tnfα → Metabolic disease



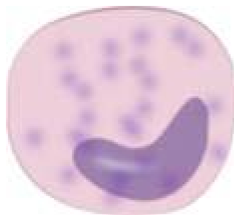
Mononuclear cells from adults with <u>T1DM</u> and <u>T2DM</u> have increased expression of TLR2, TLR4, CCL2 and increased secretion of IL6 and Tnfα

<u>Mice</u> lacking TLR2 or TLR4 are protected from high fat diet induced insulin resistance

Mononuclear cells from <u>obese</u> adults have increased NFκB binding and increased expression of IL6 and Tnfα

Devaraj S, et al.. J Clin Endocrinol Metab. 2008; Bradshaw EM, et al. J Immunol. 2009; Cipolletta C, et al. Diabetes. 2005; Devaraj S, et al. Circulation. 2000; Devaraj S, et al. Free Radic Biol Med. 2000; Ghanim H, et al, Circulation. 2004. Caricilli AM. 2008. J Endo

Objectives



Monocyte

Adolescents

- Overweight with metabolic syndrome (Overwt-MetSyn)
- Overweight without metabolic syndrome (Overwt-Healthy)
- Lean

1

Assess inflammatory state

Gene expression

- Toll-like receptors (TLR2, TLR4)
- Cytokines (Tnfα, IL6)

2

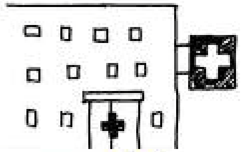
Correlate monocyte inflammation with anthropometric measurements and serum markers

- BMI, Waist circumference
- Glucose, Insulin, Lipid profile
- Tnfα, IL6

Hypotheses:

1. Monocytes from Overwt-MetSyn subjects will have increased gene expression of TLRs and cytokines when compared to Overwt-Healthy and Lean subjects
2. TLR and cytokine expression will show a positive correlation with anthropometric and serum markers of metabolic disease

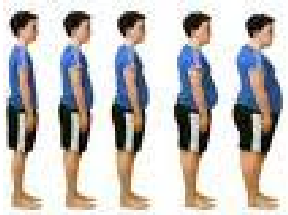
Twenty four adolescents were recruited to participate in this pilot study



UMMS clinics

- Pediatric endocrinology
- Adolescent medicine
- Nutrition

Dorchester Academy – inner city high school



- Lean – BMI < 85% age and gender
- Overweight – BMI > 85% age and gender
- Metabolic syndrome – 3 of the 5 components
  - *Waist circumference > 75% age, gender, ethnicity
  - *Triglycerides > 100
  - *HDL < 50 (girls) and < 40 (boys)
  - *Systolic blood pressure > 95% age, gender
  - *Fasting glucose >100 mg/dl

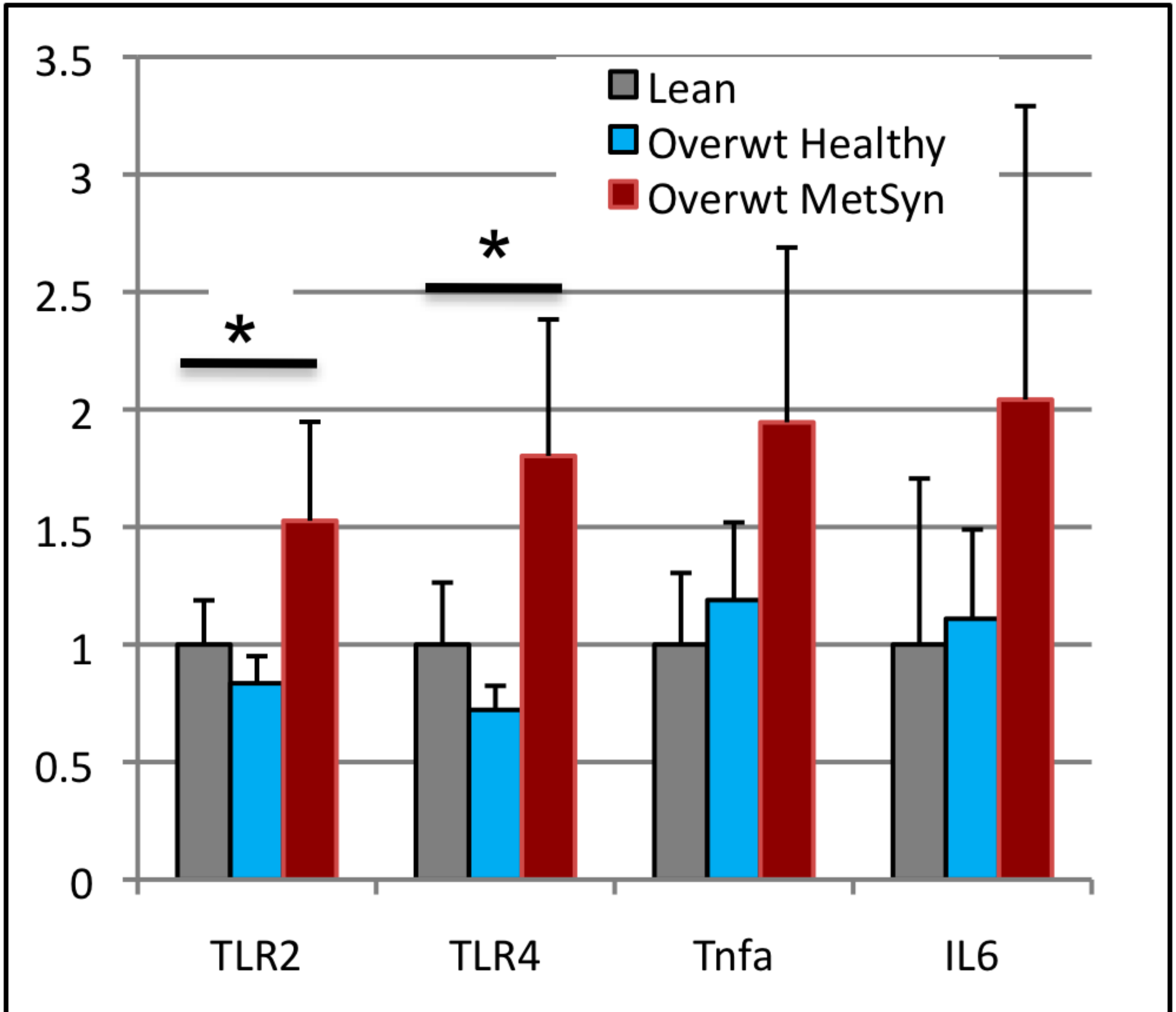
3 groups are similar in age, gender, ethnicity

	Overwt MetSyn (n=6)	Overwt Healthy (n=9)	Lean (n=9)
Female, No. (%)	6 (100)	6 (67)	6 (67)
Age (yr)	16.5 (15.4-17.2)	16.8 (15.9-19.8)	16.6 (15.6-18)
Ethnic group, No. (%)			
African American	4 (67)	6 (67)	7 (78)
Caucasian	0	2 (22)	1 (11)
Hispanic	2 (33)	1 (11)	1 (11)
BMI (kg/m2)	39 (30-52)*	32 (26-40)*	21 (18-25)^
BMI %	98 (96-99)*	94 (85-99)*	50 (36-84)^
Waist circumference	**117 (98-142)****	**95 (80-112)***	**74 (61-83)^**
Systolic BP (mm Hg)	122 (102-139)	117 (108-141)	114 (104-129)
Diastolic BP (mm Hg)	75 (61-84)	71 (53-85)	73 (63-81)
White blood cell counts (k/uL)	8 (5-12)*	6 (4-9)	6 (4-7)
Monocytes (%)	8 (4-12)	9 (6-14)	8 (5-11)
Cholesterol (mg/dL)	119 (72-162)	113 (83-155)	109 (73-145)
Triglyceride (mg/dL)	61 (23-125)	48 (24-97)	51 (34-90)
HDL (mg/dL)	40 (32-47)	41 (24-56)	42 (29-56)
LDL (mg/dL)	67 (27-90)	62 (49-100)	57 (32-81)
CRP (pg/mL)	3.5 (1-11)	3.1 (1-16)	1 (1-1)
Fasting glucose (mg/dL)	98 (82-119)	89 (66-109)	95 (84-110)
Fasting insulin (uIU/mL)	13 (2-31)*	6 (2-14)	3 (2-6)^
HOMA-IR	2.9 (0.8-7.1)*	1.3 (0.3-3.3)	0.7 (0.4-1.4)^
TNF Tnfα (pg/mL)	**2.14 (0.75-4.66)****	**0.86 (0-2.5)**	**0.95 (0.3-1.84)**
IL6 (pg/mL)	**2.75 (0.49-3.89)****	**1.46 (0-2.74)***	**0.61 (0-2.2)^**

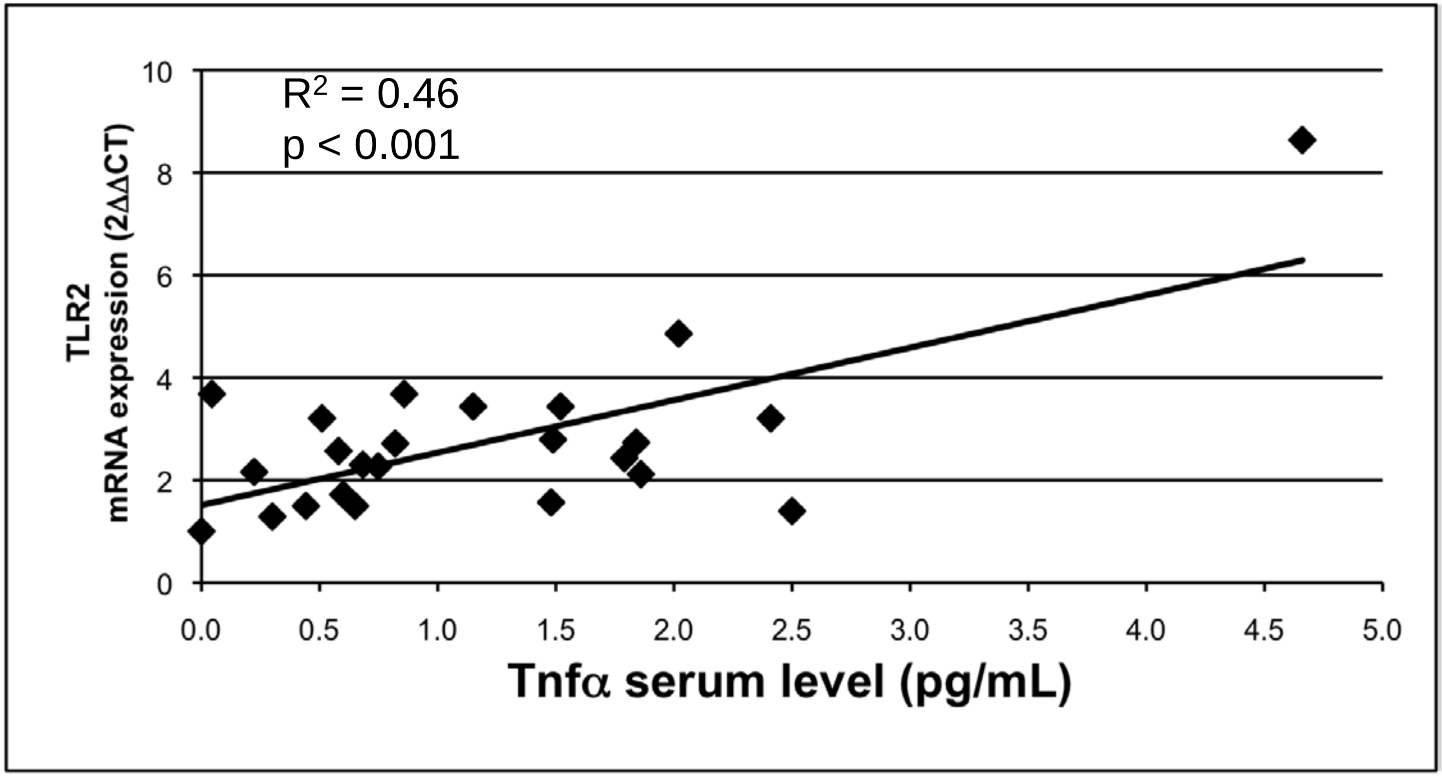
similar

Data presented as mean (range)
* P < 0.05 compared with lean
** P < 0.05 compared with overweight healthy
^ P < 0.05 compared with all overweight (Overweight healthy and Metabolic syndrome)

Monocytes from Overwt-MetSyn subjects display increased expression of inflammatory genes



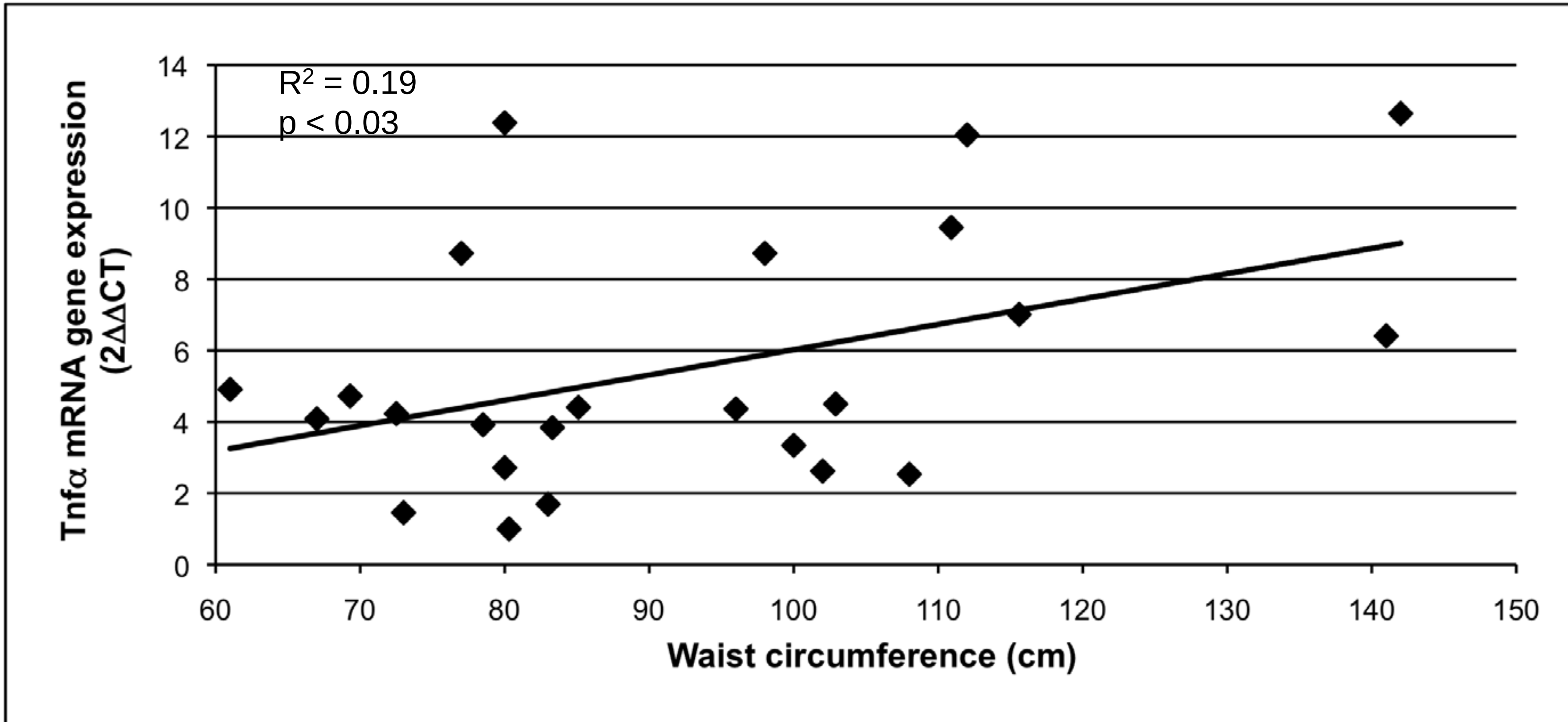
TLR expression in monocytes correlates with circulating cytokines



TLR4 expression

	R^2	*p* -value
Tnfα serum	0.2	0.02
IL6 serum	0.2	0.02

Cytokine expression in monocytes correlates with BMI and central obesity



Tnfα expression

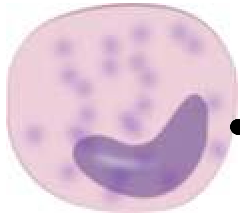
	R^2	*p* -value
BMI	0.2	0.05

IL6 expression

	R^2	*p* -value
WBC count	0.2	0.01

Conclusions

Adolescents
- Overwt-MetSyn, Overwt-Healthy, Lean



Monocyte

Yes

Yes

Assess inflammatory state

Gene expression
- TLR2, TLR4, Tnfα, IL6

Correlate monocyte gene expression with anthropometric measurements and serum markers
- BMI, Waist circumference, Tnfα, IL6

1. Monocytes from Overwt-MetSyn subjects display increased gene expression of TLRs and cytokines
2. TLR expression shows a positive correlation with circulating cytokines; cytokine expression correlates with BMI and waist circumference

Ongoing research
1. Recruit more subjects
2. Assess TLR protein expression, surface markers
3. Measure secreted cytokines (Tnfα, IL6) from cultured monocytes at baseline and in response to TLR ligands and dietary lipids

Innate immunity plays a key role in metabolic disease and may be a useful biomarker

Obese adolescent → Obese adult

↑Tnfα, IL6 serum levels → Type 2 diabetes mellitus

↑TLR2, TLR4 gene expression in MetSyn

↑Tnfα, IL6 gene expression (trend) in MetSyn

<u>Questions</u>

1. Impact of dietary changes on monocyte
2. Improvement in monocyte inflammation
loss and/or exercise?
3. Reversal of monocyte inflammation with pharmacotherapy or nutritional supplements?

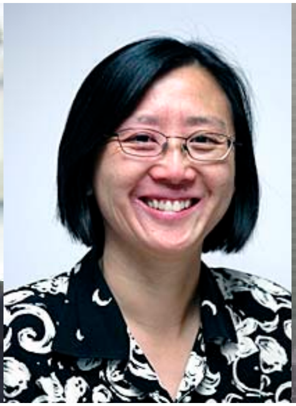
"It is unreasonable to expect that people will change their behavior so easily when so many forces in the social, cultural, and physical environment conspire against change"

Institute of Medicine, 2000

Gordon-Larsen P, et al. JAMA. 2010; Pradhan, et al. JAMA. 2001; Spranger J, et al. Diabetes. 2003.

Acknowledgments



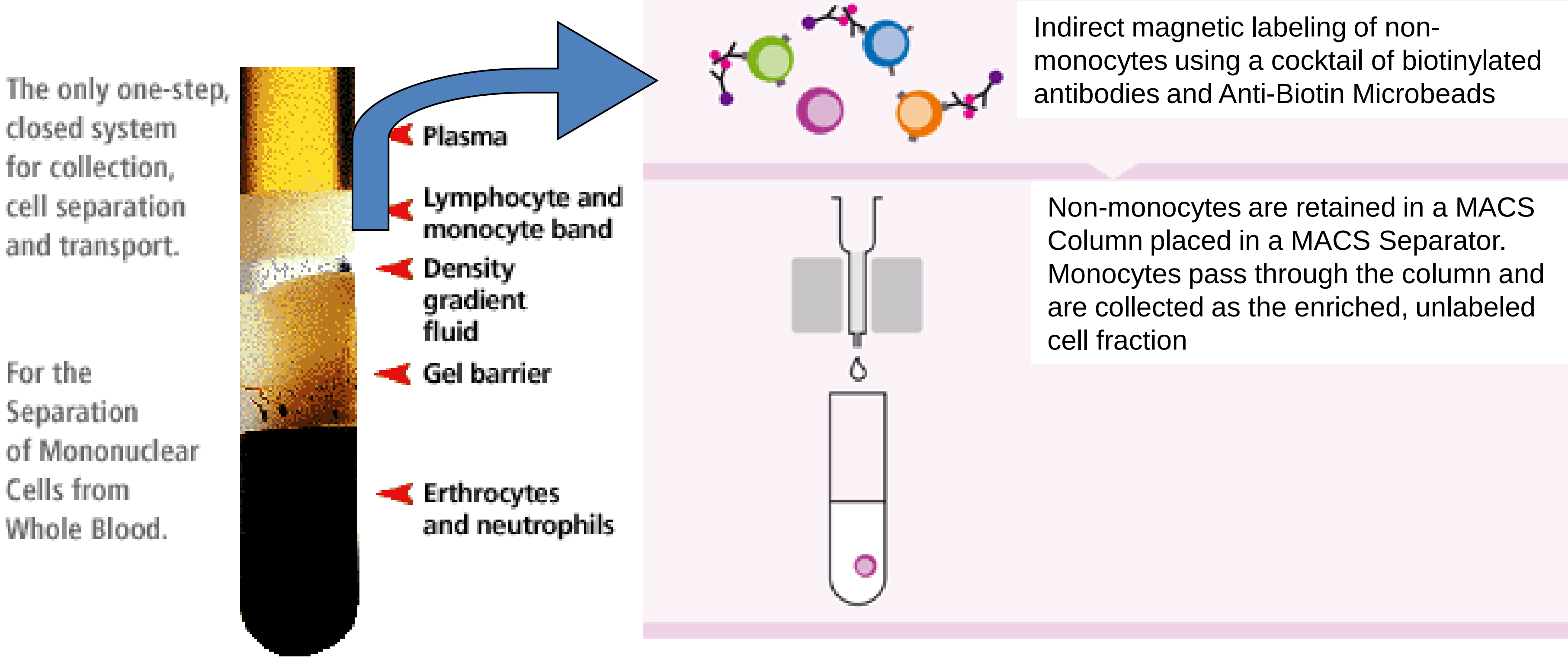



- Mentorship Team
  - Jason Kim, PhD
  - Michael P. Czech, PhD
  - Mary Lee, MD
  - Katherine Luzuriaga, MD
- Collaborators
  - Laura Hayman, PhD (UMass Boston)
  - Jean Wiecha, PhD (UMass Boston)
  - Albert Kim (UMass Boston)
- Study Nurse
  - Carol Ciccarelli, Rn
- Czech Lab Members

- Funding Support
  - CTSA K12 training grant
  - CTSA Life Sciences Moment Fund
  - Diabetes and Endocrinology Research Center Pilot and Feasibility grant

Monocytes were isolated from whole blood with indirect magnetic labelling

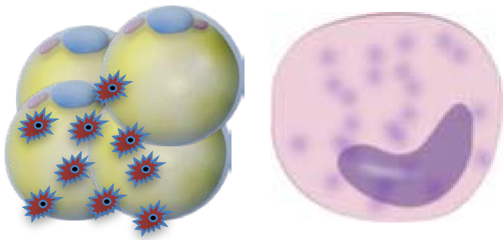
- 25 ml whole blood collected in the fasting state



http://www.bd.com/vacutainer/products/molecular/heparin/; https://www.miltenyibiotec.com

Topics to be covered in this presentation

Obesity and metabolic syndrome (MetSyn)



Immunologic pathways that may contribute to MetSyn

Potential impact on progression of metabolic disease