Atherosclerosis in Insulin Resistant Hyperlipidemic Pigs

R01 HL 069364

Timothy C. Nichols, M.D.

David R. Clemmons, M.D.

University of North Carolina, Chapel Hill

Program Goals

1. **Create NL (i.e. <u>N</u>ormal <u>L</u>ipid Chapel Hill pigs with diet-inducible atherosclerosis) and FH (<u>F</u>amilial <u>H</u>ypercholesterolemic) pigs with and without IR (<u>I</u>nsulin <u>R</u>esistance).**

2. **Document the extent and rate of development of atherosclerosis in both strains of IR pigs and compare to IS (<u>I</u>nsulin <u>S</u>ensitive) controls.**

3. **Characterize biochemical changes that occur with disease markers in serum, plasma, or lesions:**
   — **Indices of insulin sensitivity, lipoproteins,**
   — **Proinflammatory cytokines, growth regulatory proteins, etc.**

4. **Establish a colony of well-characterized animals for dissemination to the research community**

Goal 1 - Create 4 pig phenotypes:
NL/IS, NL/IR, FH/IS, FH/IR

Method - Selective breeding based on screening postpubertal pigs for **IR** and **IS** phenotype.

Goal - Postpubertal pigs, insulin (µU/ml) & chol:

	Fast	1 hr	2 hr	Cholesterol
IR	> 25	(1 or 2 hr >80)		NL or FH
IS	< 10	< 20	<20	NL or FH

Goal 1 - Breeders for 4 pig phenotypes
FH/IS, FH/IR,
NL/IS, NL/IR,

Phenotype	Gender + n	Cholesterol (mg/dl)	Serum Insulin level (µU/ml)		
			fasting	1hr	2hr
1. FH/IS	3M/1F	572.5 ± 115.1	11.1 ± 1.3	15.9 ± 3.4	13.0 ± 2.8
2. FH/IR	3M/2F	494 ± 87.7	24.8 ± 9.2	74.5 ± 30.4	52.3 ± 24.4
3. NL/IS	2M/2F	119.5 ± 28.8	7.9 ± 2.6	15.1 ± 2.2	14.5 ± 4.4
4. NL/IR	5M/2F	106.8 ± 27.1	22.2 ± 8.2	95.1 ± 61.6	54.6 ± 29.9

Program Goals

1. **Create NL (i.e. Normal Lipid Chapel Hill pigs with diet-inducible atherosclerosis) and FH (Familial Hypercholesterolemic) pigs with and without IR (Insulin Resistance).**

2. **Document the extent and rate of development of atherosclerosis in both strains of IR pigs and compare to IS (Insulin Sensitive) controls.**

3. **Characterize biochemical changes that occur with disease markers in serum, plasma, or lesions:**

    — **Indices of insulin sensitivity, lipoproteins,**

    — **Proinflammatory cytokines, growth regulatory proteins, etc.**

4. **Establish a colony of well-characterized animals for dissemination to the research community**

# Goal 3 - Characterize biochemical changes that occur with disease markers in serum, plasma, or lesions

Study sample flow chart

Pig ID, age, gender, phenotype

	A	B	C	D	E	F	G	H	I	J	K	L	M	N
1	Timepoint (months)	Baseline	1	2	3	4	5	6	7	8	9	10	11	12
2	DNA sample	x												x
3	weight	x			x			x						x
4	% body fat	x			x			x						x
5	Insulin sensitivity (Bergman)	x			x			x						x
6	Lipoprotein and lipid levels	x	x	x	x	x	x	x	x	x	x	x	x	x
7	IVUS/percutaneous				x			x						x
8	Plasma & Serum Markers	x	x		x			x						x
9	Coronary, carotid, aorta, and iliac atherosclerosis morphometry													x
10	Tissue markers													x

AMDCC Pig Tissue Distribution Plan

| INVESTIGATOR	INSTITUTION	SAMPLE	GOAL
Dale Able Don McClean	Utah	Myocardium	Biochemistry and EM mitochondrial analyses
Tim Kern	Case Western	Eyes	Retinal vessel analyses
Firousz Daneshgari	Cleveland Clinic	Bladder	Physiological analyses
Eva Feldman	Univ of Michigan	Neurological tissues (skin bx x 2, sciatic nerve, other)	Biochemical and microscopic analyses
Eva Feldman	Univ of Michigan	EDTA plasma	Screen for ROS
Charles Jeannette	UNC	Urine and kidneys	Urine protein, glomeruli & other vessel analyses

Bergman FSIGT on 52F FH/IS

Baseline S_i = 4.7
**Wt 445 lbs**
Chol 376 mg/dl
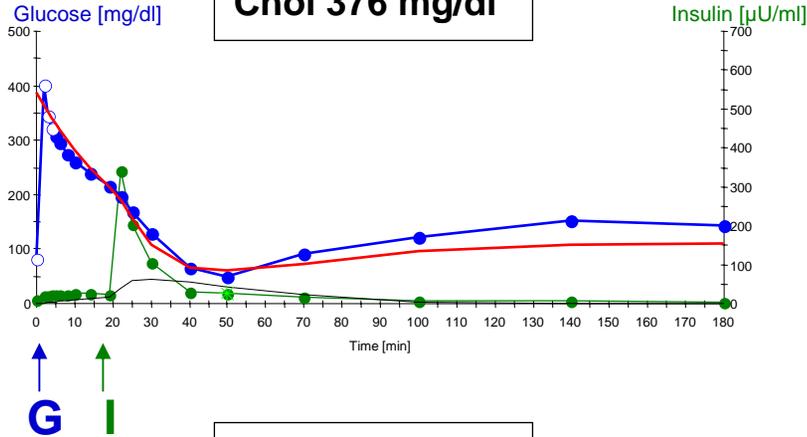
3 month S_i = 4.4
Wt 489 lbs
Chol 340 mg/dl
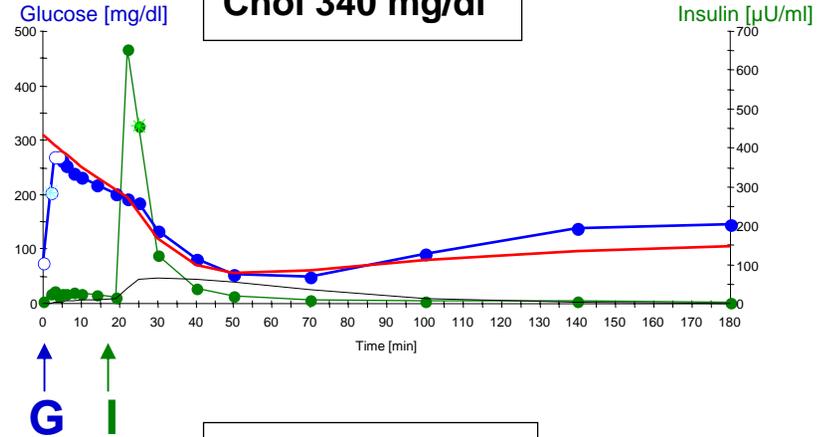
6 month S_i = 4.5
Wt 469 lbs.
Chol 301 mg/dl
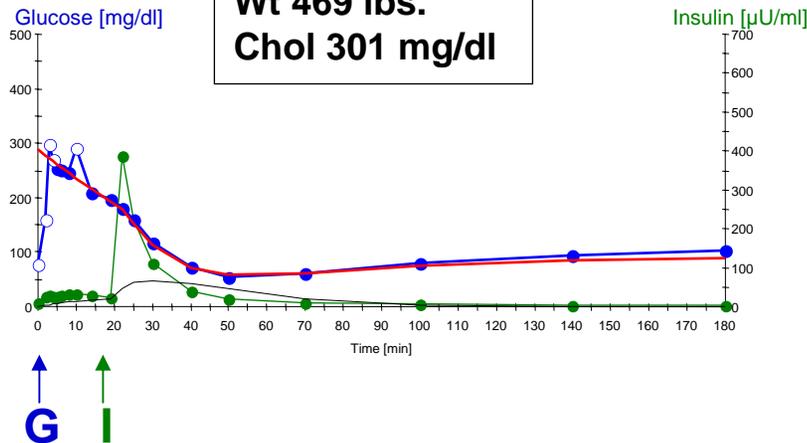
12 month S_i = 4.2
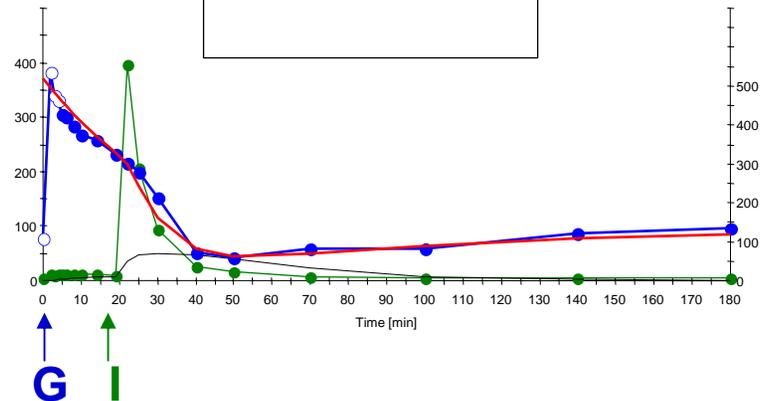
G = glucose, 0.3 g/kg/iv   I = insulin, 0.03 U/kg/iv

Bergman FSIGT on 69F NL/IS

Baseline S$_i$ = 4
Wt. 414 lbs.
chol 60 mg/dl

3 month S$_i$ = 4.3
Wt. 385 lbs.
Chol 424 mg/dl

6 month S$_i$ = 3.7
Wt. 438 lbs.
327 mg/dl.
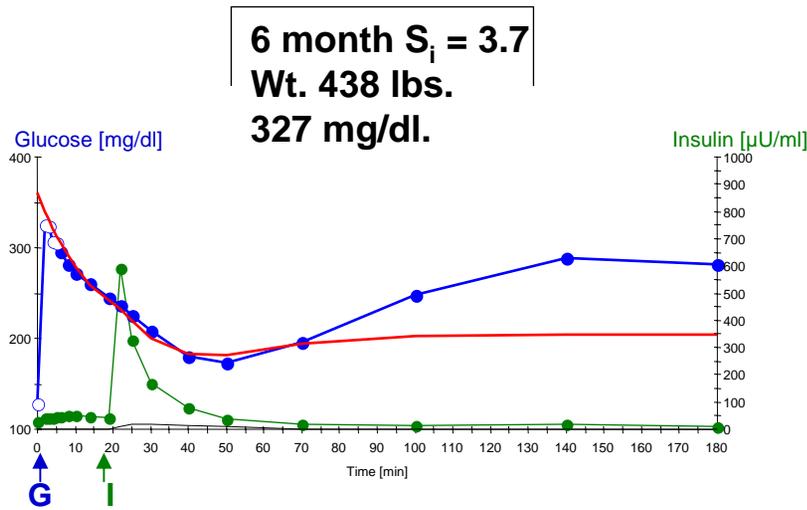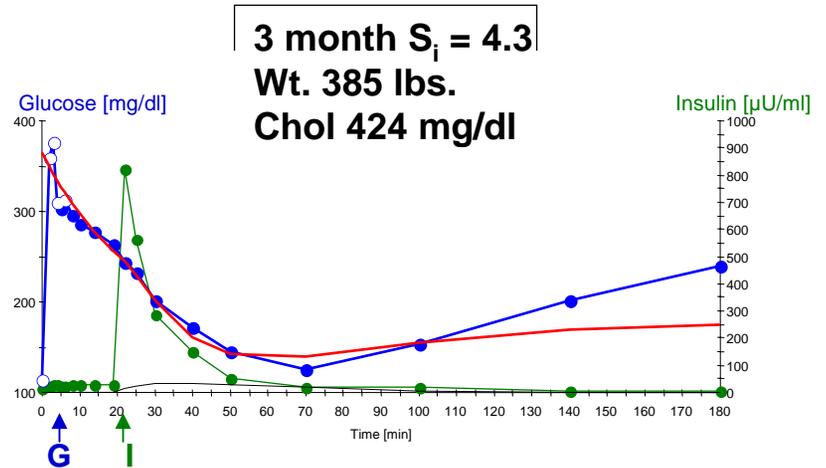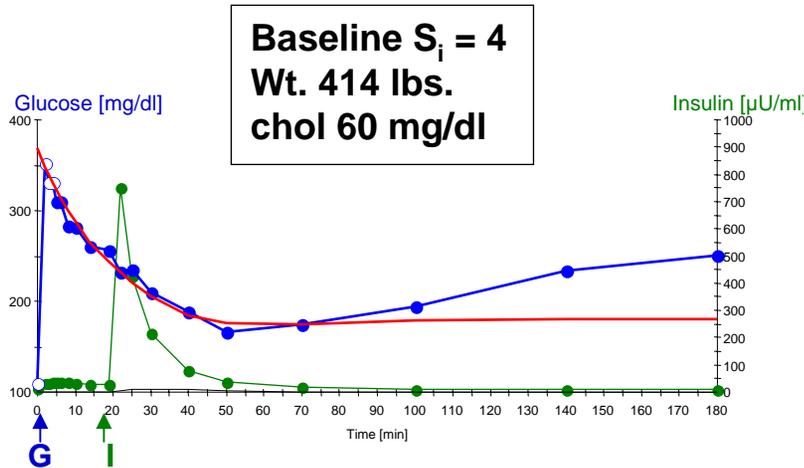
G = glucose, 0.3 g/kg/iv   I = insulin, 0.03 U/kg/iv

Program Goals

1. **Create NL (i.e. <u>N</u>ormal <u>L</u>ipid Chapel Hill pigs with diet-inducible atherosclerosis) and FH (<u>F</u>amilial <u>H</u>ypercholesterolemic) pigs with and without IR (<u>I</u>nsulin <u>R</u>esistance).**

2. **Document the extent and rate of development of atherosclerosis in both strains of IR pigs and compare to IS (<u>I</u>nsulin <u>S</u>ensitive) controls.**

3. **Characterize biochemical changes that occur with disease markers in serum, plasma, or lesions:**
   — **Indices of insulin sensitivity, lipoproteins,**
   — **Proinflammatory cytokines, growth regulatory proteins, etc.**

4. **Establish a colony of well-characterized animals for dissemination to the research community**

Plans for Year 4

1. Continued selective breeding for increased IR.

2. Continued breeding for smaller pigs - Ossabaw strain

3. Complete new pig housing.

4. Continue atherosclerosis study.

5. Monitoring insulin sensitivity in experimental pigs.

6. Validate NMR lipoprotein analyses in all phenotypes

7. Begin validation of pig gene microarray analyses for monitoring originally proposed genes and potentially identification of new genes of interest.

8. Distribute tissues to AMDCC members.