

# **LAB MANUAL**

**Framingham Heart Study  
Cohort / Offspring / Omni**

**MGH DIABETES LAB /  
MGH GCRC CORE LAB**

**Hemoglobin A1c  
Insulin, proinsulin  
Adiponectin, resistin, TNF-alpha  
Urine microalbumin, creatinine, sodium**

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## Cohort Exams 19-20, 22 Hemoglobin A1c

### 1. Funding Source/Lab

Framingham specimens
Grant # FHS contract
Lab: MGH Diabetes Lab
Contact: James Meigs: jmeigs@partners.org

### 2. Method: High-performance liquid chromatography

### 3. Technical Aspects

Method	Nathan DM. Clin Chem 27:1261–1263, 1981; DCCT standardized
Units	% of total hemoglobin
Measuring range	0% - >20%

### 4. FHS Specimen Characteristics

a. fresh whole blood

### 5. QC aspects

CV intra-assay	<2.5%
CV inter-assay	<2.5%

### 6. FHS participant aspect

#### Descriptive Statistics

Exam 19-20 Count: 1274

Mean	SD	minimum	maximum	Median	Q1	Q3
5.65	0.99	3.53	13.66	5.44	5.10	5.91

Exam 22 Count: 824

Mean	SD	minimum	maximum	Median	Q1	Q3
5.85	1.10	4.09	13.86	5.60	5.25	6.10

## Offspring Exam 5

## Plasma Insulin (DPC)

### 1. Funding Source/Lab

Framingham specimens
Grant # FHS contract
Lab: MGH Diabetes Lab
Contact: James Meigs: <a href="mailto:jmeigs@partners.org">jmeigs@partners.org</a>

### 2. Method: Coat-A-Count RIA

### 3. Technical Aspects

Vendor	Diagnostics Products Corp, Los Angeles, CA
Units	uU/mL
Minimum detectable level	1.2 uU/mL

### 4. FHS Specimen Characteristics

- Plasma
- Frozen samples -80°C
- 10 serum samples drawn from FHS employee volunteers were assayed for DPC insulin to generate a regression equation to estimate serum insulin levels from plasma insulin levels for reporting in papers. The serum to plasma insulin correlation was  $r = 0.993$ , and the conversion equation was: serum insulin = (plasma insulin - 17.09)/1.4

### 5. QC aspects

CV intra-assay	5.6%
CV inter-assay	8.8%

### 6. FHS participant aspect

#### Fasting Insulin

Count = offspring: 3629

#### Descriptive Statistics

Offspring:

Mean	SD	minimum	maximum	Median	Q1	Q3
31.45	21.11	11.00	720.00	27.00	23.00	34.05

#### 2 Hour OGTT Insulin

Offspring Count: 3388

#### Descriptive Statistics

Offspring:

Mean	SD	minimum	maximum	Median	Q1	Q3
98.56	71.21	15.00	740.00	79.00	53.00	119.00

## Offspring Exam 5

## Plasma Insulin (Linco)

### 1. Funding Source/Lab

Framingham specimens
Grant # MGH internal funds
Lab : MGH Diabetes Lab
Contact: James Meigs: <a href="mailto:jmeigs@partners.org">jmeigs@partners.org</a>

### 2. Method: Human specific insulin RIA

### 3. Technical Aspects

Vendor	Linco Research Inc, St. Charles, MO
Units	uU/mL
Minimum detectable level	2 uU/mL

### 4. FHS Specimen Characteristics

- Plasma
- Frozen samples -80°C
- Samples analyzed ~9 years after exam 5 DPC assay performed
- For calibration with exam 7 Linco insulin levels only

### 5. QC aspects

CV intra-assay	3.9%
CV inter-assay	Low control 6.1%, high control 4.7%

### 6. FHS participant aspect

Count offspring: 187

Descriptive Statistics

Offspring:

Mean	SD	minimum	maximum	Median	Q1	Q3
51.37	45.49	6.26	184.00	28.6	15.60	85.60

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## Offspring Exam 5

## Plasma Proinsulin (Linco)

### 1. Funding Source/Lab

Framingham specimens
Grant # MGH internal funds
Lab : MGH Diabetes Lab
Contact: James Meigs: jmeigs@partners.org

### 2. Method: Human proinsulin RIA

### 3. Technical Aspects

Vendor	Linco Research Inc, St. Charles, MO
Units	uU/mL
Minimum detectable level	2 uU/mL

### 4. FHS Specimen Characteristics

- Plasma
- Frozen samples -80°C
- For calibration with exam 7 Linco proinsulin levels only

### 5. QC aspects

CV intra-assay	3.7%
CV inter-assay	Low control 12.5%, High control 7.8%

### 6. FHS participant aspect

Count of offspring: 7/86

#### Descriptive Statistics

#### Offspring

Mean	SD	minimum	maximum	Median	Q1	Q3
31.77	22.50	3.88	88.70	26.15	10.90	50.20

**1.Funding Source/Lab**

Framingham specimens
Grant # FHS contract
Lab : MGH Diabetes Lab
Contact: James Meigs: jmeigs@partners.org

**2. Method: High-performance liquid chromatography****3.Technical Aspects**

Method	Nathan DM. Clin Chem 27:1261–1263, 1981; DCCT standardized
Units	% of total hemoglobin
Measuring range	0% - >20%

**4.FHS Specimen Characteristics**

a. Fresh whole blood

**5. QC aspects**

CV intra-assay	<2.5%
CV inter-assay	<2.5%

**6. FHS participant aspect**

Count offspring: 12732

Descriptive Statistics

Offspring

Mean	SD	minimum	maximum	Median	Q1	Q3
5.47	1.02	3.07	13.71	5.28	4.92	5.69

## Offspring Exam 7

## Adiponectin

### 1. Funding Source/Lab

Framingham specimens

Grant # Meigs American Diabetes Association Career Development Award; NCRR GCRC M01-RR-01066

Lab : MGH Mallinckrodt General Clinical Research Center Core Lab

Contact: James Meigs: jmeigs@partners.org

### 2. Method: Enzyme-linked Immunosorbent Assay (EIA) (R&D kit # DRP300)

### 3. Technical Aspects

Vendor	R & D Systems, Minneapolis, MN
Units	ng/dL
Minimum detectable level	0.079 – 0.891 ng/dL

### 4. FHS Specimen Characteristics

- a. Plasma
- b. Frozen samples -80°C

### 5. QC aspects

CV intra-assay	Low control 6.16%, high control 5.34%
CV inter-assay	Low control 9.6%, high control 9.0%

### 6. FHS participant aspect

Count offspring: 12773

Descriptive Statistics

Offspring:

Mean	SD	minimum	maximum	Median	Q1	Q3
10.09	6.34	0.70	59.88	8.50	5.50	13.10

**1.Funding Source/Lab**

Framingham specimens

Grant # Meigs American Diabetes Association Career Development Award; NCRR GCRC M01-RR-01066

Lab : MGH Mallinckrodt General Clinical Research Center Core Lab

Contact: James Meigs: jmeigs@partners.org

**2. Method: Enzyme-linked Immunosorbent Assay (ELISA) (R&D kit # DRTSN00)**

**3.Technical Aspects**

Vendor	R & D Systems, Minneapolis, MN
Units	ng/dL
Assay range	0.156 – 10 ng/dL

**4.FHS Specimen Characteristics**

- a. Plasma
- b. Frozen samples -80°C

**5. QC aspects**

CV intra-assay	Low control 7.58%, high control 10.47%
CV inter-assay	Low control 8.3%, high control 7.6%

**6. FHS participant aspect**

Count of offspring: 2790

**Descriptive Statistics**

Offspring:

Mean	SD	minimum	maximum	Median	Q1	Q3
14.27	7.43	1.20	110.012	12.70	10.00	16.40

## Offspring Exam 7

## high sensitivity TNF-alpha

### 1. Funding Source/Lab

Framingham specimens

Grant # Meigs American Diabetes Association Career Development Award; NCRR GCRC M01-RR-01066

Lab : MGH Mallinckrodt General Clinical Research Center Core Lab

Contact: James Meigs: [jmeigs@partners.org](mailto:jmeigs@partners.org)

### 2. Method: Enzyme-linked Immunosorbent Assay (EIA) (R&D kit # HSTA00C)

### 3. Technical Aspects

Vendor	R & D Systems, Minneapolis, MN
Units	pg/dL
Minimum Detectable Level	0.06 – 0.32 pg/mL

### 4. FHS Specimen Characteristics

- a. Plasma
- b. Frozen samples -80°C

### 5. QC aspects

CV intra-assay	Low control 7.57%, high control 5.63%
CV inter-assay	Low control 11.3%, high control 5.4%

### 6. FHS participant aspect

Count offspring: 2537

Descriptive Statistics

Offspring:

Mean	SD	minimum	maximum	Median	Q1	Q3
1.48	1.29	0.28	21.10	1.21	0.93	1.64

## Offspring Exam 7 Plasma Insulin (Linco)

### 1. Funding Source/Lab

Framingham specimens

Grant # FHS Contract

Lab : MGH Diabetes Lab

Contact:

James Meigs: jmeigs@partners.org

### 2. Method: Human specific insulin RIA

### 3. Technical Aspects

Vendor	Linco Research Inc, St. Charles, MO
Units	pmol/L
Minimum detectable level	12 pmol/L

### 4. FHS Specimen Characteristics

a. Plasma

b. Frozen samples -80°C

### 5. QC aspects

CV intra-assay	3.9%
CV inter-assay	Low control 6.1%, high control 4.7%

### 6. FHS participant aspect

#### Fasting Insulin

Offspring count: 3261

Descriptive Statistics

Mean	SD	minimum	maximum	Median	Q1	Q3
93.57	66.77	14.04	1296.00	78.00	57.30	109.80

#### 2 Hour OGTT Insulin

Offspring Count: 1113

Descriptive Statistics

Mean	SD	minimum	maximum	Median	Q1	Q3
444.20	399.92	36.06	4500.00	328.20	196.20	552.60

## Offspring Exam 7 Plasma Proinsulin (Linco)

### 1. Funding Source/Lab

Framingham specimens

Grant # FHS Contract

Lab : MGH Diabetes Lab

Contact:

James Meigs: [jmeigs@partners.org](mailto:jmeigs@partners.org)

### 2. Method: Human specific insulin RIA

### 3. Technical Aspects

Vendor	Linco Research Inc, St. Charles, MO
Units	pmol/L
Minimum detectable level	2 pmol/L

### 4. FHS Specimen Characteristics

a. Plasma

b. Frozen samples -80°C

### 5. QC aspects

CV intra-assay	3.7%
CV inter-assay	Low control 12.5%, high control 7.8%

### 6. FHS participant aspect

Offspring count: 3170

Descriptive Statistics

Mean	SD	minimum	maximum	Median	Q1	Q3
16.50	15.73	2.04	199.00	11.80	8.03	5.00

## Offspring Exam 7 Plasma Insulin (DPC)

### 1. Funding Source/Lab

Framingham specimens
Grant # FHS Contract
Lab : MGH Diabetes Lab
Contact: James Meigs: <a href="mailto:jmeigs@partners.org">jmeigs@partners.org</a>

### 2. Method: Human specific insulin RIA

### 3. Technical Aspects

Vendor	Diagnostics Products Corp, Los Angeles, CA
Units	uU/mL
Minimum detectable level	1.2uU/mL

### 4. FHS Specimen Characteristics

- Plasma
- Frozen samples -80°C
- For calibration with exam 5 DPC, or estimation of exam 7 DPC insulin from exam 7 Linco insulin levels only

### 5. QC aspects

CV intra-assay	5.6%
CV inter-assay	8.8%

### 6. FHS participant aspect

#### Fasting Insulin

Offspring count: 17

#### Descriptive Statistics

Mean	SD	minimum	maximum	Median	Q1	Q3
9.52	7.47	1.80	31.20	7.96	5.00	11.00

## Offspring Exam 7 Serum Insulin (Linco)

### 1. Funding Source/Lab

Framingham specimens

Grant # MGH internal funds

Lab : MGH Diabetes Lab

Contact:

James Meigs: [jmeigs@partners.org](mailto:jmeigs@partners.org)

### 2. Method: Human proinsulin RIA

### 3. Technical Aspects

Vendor	Linco Research Inc, St. Charles, MO
Units	pmol/L
Minimum detectable level	12 pmol/L

### 4. FHS Specimen Characteristics

- Serum
- Frozen samples -80°C
- For estimation of exam 7 serum insulin levels from exam 7 plasma insulin levels only

### 5. QC aspects

CV intra-assay	3.9%
CV inter-assay	Low control 6.1%, high control 4.7%

### 6. FHS participant aspect

Offspring count: 97

Descriptive Statistics

Offspring:

Mean	SD	minimum	maximum	Median	Q1	Q3
91.06	55.70	39.36	340.80	77.40	54.06	100.20

**1. Funding Source/Lab**

Framingham specimens
Grant # FHS contract
Lab : MGH Diabetes Lab
Contact: James Meigs: jmeigs@partners.org

**2. Method: High-performance liquid chromatography****3. Technical Aspects**

Method	Nathan DM. Clin Chem 27:1261–1263, 1981; DCCT standardized
Units	% of total hemoglobin
Measuring range	0% - >20%

**4. FHS Specimen Characteristics**

a. Fresh whole blood

**5. QC aspects**

CV intra-assay	<2.5%
CV inter-assay	<2.5%

**6. FHS participant aspect**

Count : offspring: '2992

## Descriptive Statistics

## Offspring

Mean	SD	minimum	maximum	Median	Q1	Q3
5.66	0.98	1.72	14.57	5.47	5.13	5.89

## Exam 7 OGTT Two Hour Glucose

### 1. Funding Source/Lab

Framingham specimens

Grant # FHS contract

Lab : FHS Laboratory

Contact:

James Meigs: jmeigs@partners.org

### 2. Method:

### 3. Technical Aspects

Vendor	A-gent glucose test; Abbott, South Pasadena, CA
Units	mg/dl
Measuring range	0 - > 400 mg/dl

### 4. FHS Specimen Characteristics

a. Fresh plasma

b. hexokinase reagent kit

c. All subjects with IGT (fasting plasma glucose 110 – 125 mg/dl OR 2hr OGTT plasma glucose 140-199 mg/dl) and a subset of subjects with NGT (fasting plasma glucose <110 mg/dl AND 2hr OGTT plasma glucose <140 mg/dl) at exam 5 (sex block-randomly selected from 5 quintile strata of FBS) were offered a morning exam at cycle 7 including an oral glucose tolerance test. This sampling design produced a random sample of about 200 men/women in each quintile cell of fasting plasma glucose and a random sample of 200 men/women with IGT.

### 5. QC aspects

CV intra-assay	<3%
CV inter-assay	<3%

### 6. FHS participant aspect

Offspring Count: 1112

Descriptive Statistics

Mean	SD	minimum	maximum	Median	Q1	Q3
130.48	49.54	44.00	450.00	119.00	97.00	152.00

## Offspring Exam 6

## Urine Microalbumin

### 1. Funding Source/Lab

Framingham specimens

Grant # Meigs American Diabetes Association Research Grant; reagents donated by Roche Diagnostics Inc.

Lab : ██████████ Children's Hospital

Contact: James Meigs: jmeigs@partners.org

### 2. Method: Tina-quant Albumin assay

### 3. Technical Aspects

Vendor	Roche Diagnostics, Indianapolis, IN
Units	mg/L
Measuring range	3-4429 mg/dL

### 4. FHS Specimen Characteristics

- Spot urine
- frozen specimens -20°C
- for calculation of urine albumin / creatinine ratio

### 5. QC aspects

CV intra-assay	3.0%
CV inter-assay	Low control 7.2%, High control 2.1%

### 6. FHS participant aspect

Count offspring: 2993

Descriptive Statistics

Offspring (Exam 6):

Mean	SD	minimum	maximum	Median	Q1	Q3
26.56	158.89	0	4223.00	5.40	3.00	11.20

## Offspring Exam 6

## Urine Creatinine

### 1. Funding Source/Lab

Framingham specimens

Grant # Meigs American Diabetes Association Research Grant; reagents donated by Roche Diagnostics Inc.

Lab : ██████████ Children's Hospital

Contact: James Meigs: jmeigs@partners.org

### 2. Method: modified Jaffe

### 3. Technical Aspects

Vendor	Roche Diagnostics, Indianapolis, IN
Units	g/dL
Measuring range	0.1-250 g/dL

### 4. FHS Specimen Characteristics

- a. Spot urine
- b. Frozen specimen -20°C

### 5. QC aspects

CV intra-assay	2.0%
CV inter-assay	Low control 2.3%, High control 1.9%

### 6. FHS participant aspect

Count      offspring:    /2993

Descriptive Statistics

Offspring (Exam 6):

Mean	SD	minimum	maximum	Median	Q1	Q3
1.15	0.71	0.05	6.26	1.07	0.63	1.53

## Offspring Exam 6

## Urine Sodium

### 1. Funding Source/Lab

Framingham specimens

Grant # Meigs American Diabetes Association Research Grant; reagents donated by Roche Diagnostics Inc.

Lab : ██████████ Children's Hospital

Contact: James Meigs: jmeigs@partners.org

### 2. Method: automated ion electrode (Boehringer Mannheim)

### 3. Technical Aspects

Vendor	Roche Diagnostics, Indianapolis, IN
Units	mmol/L
Measuring range	10-250 mmol/L

### 4. FHS Specimen Characteristics

- a. Spot urine
- b. Frozen specimen -20°C

### 5. QC aspects

CV intra-assay	2.4%
CV inter-assay	2.8%

### 6. FHS participant aspect

Count offspring: 2993

Descriptive Statistics

Offspring (Exam 6):

Mean	SD	minimum	maximum	Median	Q1	Q3
97.80	47.52	6.00	271.00	95.00	62.00	131.00

## Offspring Exam 6

## urine albumin / creatinine ratio

### 1. Funding Source/Lab

Framingham specimens

Grant # Meigs American Diabetes Association Research Grant; reagents donated by Roche Diagnostics Inc.

Lab : ██████████ Children's Hospital

Contact: James Meigs: [jmeigs@partners.org](mailto:jmeigs@partners.org)

### 2. Characteristics

a. Derived variable urine microalbumin/creatinine ratio (UACR) (units = g/mg)

### 3. FHS participant aspect

Count offspring: /2993

Descriptive Statistics

Offspring (Exam 6):

Mean	SD	minimum	maximum	Median	Q1	Q3
31.37	219.38	0.02	6789.39	6.41	2.82	15.42

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