MONOGENIC DIABETES: FROM BENCH TO BEDSIDE
SUNY BUFFALO-DEPARTMENT OF PEDIATRICS GRAND ROUNDS
CHILDREN'S HOSPITAL JULY 23, 2010

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Disclosure Information
Monogenic Diabetes
Mark A Sperling

• I have the following relationships to disclose:
  • Speakers Bureau at Athena Diagnostics
  • Speakers Bureau at Novo Nordisk

OBJECTIVES
• Distinguish between Type 1 Diabetes Mellitus, T2DM, and monogenic forms of the disease complex.
• Appreciate the diagnostic, therapeutic and prognostic significance of establishing the correct molecular basis of monogenic forms of diabetes.
• Recognize the contribution of genetic defects to the global burden of Diabetes Mellitus—monogenic forms are unusual (about 1%-3%) but not rare.

WHAT IS DIABETES MELLITUS—HOW DO YOU DEFINE IT??

Classification of Diabetes Mellitus –ADA 2009

Natural history of T1D

Type 1 autoimmunity: Type 1 DM is autoimmune-insulin deficiency is the hallmark
Type 2 commonly has associated obesity, is more common in certain ethnic groups such as AA, Native Americans, SE Asians is associated with insulin resistance and substantial insulin secretion in many, but insulin deficiency in others. Other specific types includes MODY, NDM, receptor defects, toxins:** In rare instances, patients may require insulin for survival
Empiric risk of developing Type 1 diabetes

First degree relatives of T1DM probands*: 5-7%
Individuals without relatives with T1DM*: <1%
Children of affected father**: ~6%
Children of affected mother**: ~2%

These estimates are for North American Caucasian* and Scandinavian populations**.

Genome-wide association study and meta-analysis find that over 40 loci affect risk of type 1 diabetes
Jeffrey C Barrett et al. & The Type 1 Diabetes Genetics Consortium11
Nature Genetics 41, 703 – 707:2009

HLA system on 6p21
Insulin gene VNTR
CTLA 4
PTPN22

OTHERS
T1a DM is an autoimmune disease, markers of islet autoimmunity in the majority, affects young people, leads to total insulin deficiency/dependence, hence ketosis prone.

High Risk HLA-DQ Haplotypes

Defined as either DQA1*0501-DQB1*0201 (DR3) and/or DQA1*0301-DQB1*0302 (DR4)
0 High Risk Haplotypes
1 High Risk Haplotype
2 High Risk Haplotypes

Insulin Response to Standard Hyperglycemic Clamp

Healthy, patient population
Adolescents
Pre-Adolescents

Longitudinal Study of Insulin Sensitivity

**Pre-Pubertal vs Pubertal (N=9)**

<table>
<thead>
<tr>
<th>Healthy patient population</th>
</tr>
</thead>
<tbody>
<tr>
<td>P &lt; 0.001</td>
</tr>
<tr>
<td>P &lt; 0.05</td>
</tr>
</tbody>
</table>


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**Glucose and Insulin Responses to OGTT In Pre-Pubertal and Pubertal Subjects**

<table>
<thead>
<tr>
<th>Healthy, patient population</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>PRE-PUBERTAL Glucose RESPONSE</strong></td>
</tr>
<tr>
<td>1.75 g/kg (N = 9)</td>
</tr>
<tr>
<td><strong>PUBERTAL Glucose RESPONSE</strong></td>
</tr>
<tr>
<td>1.75 g/kg (N = 10)</td>
</tr>
</tbody>
</table>

| Fasting Blood Glucose (mg/dL) | 82.0±3.3 | 75.6±4.1 |
| Peak Blood Glucose (mg/dL) | 151.6±18.5 | 143.3±27.5 |
| Area Glucose mg/dL x 4hr | 421.1±17.0 | 409.5±16.0 |
| Area Insulin μU/mL x 4hr | 118.5±3.0 | 115.4±16.5 |

Bloch C, et al. J Pediatrics. 1987; 110:481-87 ** §Like symbols compared; P < 0.05

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**Cumulative risk of developing clinical Type 1 diabetes in relatives of IDDM probands using Ab markers alone (IAA, GAD65, IA-2, ICA)**

<table>
<thead>
<tr>
<th>Percent IDDM-free</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 Abs</td>
</tr>
<tr>
<td>1 Ab</td>
</tr>
<tr>
<td>2 Abs</td>
</tr>
<tr>
<td>3 Abs</td>
</tr>
<tr>
<td>4 Abs</td>
</tr>
</tbody>
</table>

20 Log Rank
P < 0.00001


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**Update in Type 1 Diabetes**


- Typically, autoantibodies reacting with glutamic acid decarboxylase (GAD65), insulin, and insulinoma antigen-2 are measured. Assays can be set such that each assay has a false-positive rate of ~1%, and together one or more of these autoantibodies are present in approximately 90% of new onset patients with type 1A diabetes. There are undoubtedly additional autoantigens to be defined (Zn Transporter 8). A subset of patients lacking the above 3-4 islet autoantibodies express cytoplasmic islet cell autoantibodies measured with indirect immunofluorescent stains of human pancreas.

NIH Study Section: One of the eligibility criteria for the study for T1D subjects is a positive autoantibody result (GAD65 or ICA512). Dr. X indicated that a negative antibody result could be expected in 15% of subjects.

**DO THE 10%-15% OF CHILDREN WITHOUT ANTIBODIES HAVE T1DM?? IS IT BECAUSE WE HAVEN'T IDENTIFIED THE AB OR BECAUSE THEY DON'T HAVE AUTOIMMUNITY??**

Patterns of glucose and insulin in lean and obese individuals


Balance Between Insulin Sensitivity and Secretion

More Insulin Resistant

Less Insulin Resistant

Gain-of-function mutations
Channel remains open
Decr. insulin secretion
Neonatal DM

KCNJ11

Inhibited by:
ATP, ADP

Activated by:
P2, fatty acid metabolites

Loss-of-function mutations
Channel remains closed
Incr. insulin secretion
Neonatal hyperinsulinemic hypoglycemia

SPERLING MA NEJM 2006;355:507-510
(Editorial)

PANCREATIC ß CELL

Glucose

Glut 2

Insulin Exocytosis

ATP/ADP

Ca++

Insulin Resistant

NGT

IGT

T2D


**Diabetes Mellitus in Mitochondrial Diseases**


**Inherited Defects in Beta Cell Function**

<table>
<thead>
<tr>
<th>Type of defect</th>
<th>Molecule</th>
<th>Mode of inheritance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glucose sensing</td>
<td>Glucokinase</td>
<td>AD (MODY2)</td>
</tr>
<tr>
<td>Coupling</td>
<td>K&lt;sub&gt;a&lt;/sub&gt; Channel</td>
<td>Many mitochondrial</td>
</tr>
</tbody>
</table>

- Insulin production
  - Transcription factors HNF-4 alpha AD (MODY1)
  - For insulin gene HNF-1 alpha AD (MODY3)
  - IPF-1 AD (MODY4)
  - AR (pancreatic agenesis)
  - HNF-1 beta AD (MODY5)
  - Neuro D/beta 2 AD (MODY6)

- Insulin synthesis
  - or processing
  - Insulinopathy AD (hyperproinsulinemia)
  - AD (insulinopathy)

**MODY: Classification**

- Other Specific Type of Diabetes

**Classification of MODY**

<table>
<thead>
<tr>
<th>Type</th>
<th>Gene</th>
<th>Chromosome</th>
<th>Treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>MODY 1</td>
<td>HNF4-alfa</td>
<td>20q12-q13.1</td>
<td>INSULIN</td>
</tr>
<tr>
<td>MODY 2</td>
<td>Glucokinase</td>
<td>7p11-q31.1</td>
<td>EXERCISE/DIET</td>
</tr>
<tr>
<td>MODY 3</td>
<td>HNF1-alfa</td>
<td>12q24.2</td>
<td>INSULIN/SU</td>
</tr>
<tr>
<td>MODY 4</td>
<td>IPF1</td>
<td>13q12.1</td>
<td>INSULIN</td>
</tr>
<tr>
<td>MODY 5</td>
<td>HNF1-beta</td>
<td>7cen-q21.3</td>
<td>INSULIN/SU</td>
</tr>
<tr>
<td>MODY 6</td>
<td>NeuroD1-beta2</td>
<td>2q32</td>
<td>INSULIN</td>
</tr>
<tr>
<td>MODY 7-9</td>
<td>CEL5a-1; AIP 11 &amp; Other candidates</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

MODY constitutes about 2% +/- of new onset childhood diabetes and should be suspected in those with a family history of multiple affected members in 2-3 generations with onset before age 35; absence of markers of autoimmunity; “mild diabetes” requiring less than 0.5 U/Kg from the outset or unusually prolonged “honeymoon” phase; early appearance of albuminuria (<5 years of DM) despite good metabolic control with or without detectable renal abnormalities.

The Clinical Manifestations, Pathophysiology and Treatment are reviewed in Fajans SS, Bell GI, Polonsky KS. Molecular mechanisms and clinical pathophysiology of maturity-onset diabetes of the young. N Engl J Med. 2001 345:971-80

**Inherited Defects in Beta Cell Function**

- Insulin production
  - for insulin gene
  - HNF-1 alpha AD (MODY1)
  - IPF-1 AD (MODY4)
  - HNF-1 beta AD (MODY5)
  - Neuro D/beta 2 AD (MODY6)
- Insulin synthesis
  - or processing
  - Insulinopathy AD (hyperproinsulinemia)
  - AD (insulinopathy)
CASE HISTORY
Case 1

15-year-old previously healthy W/M, presented with hyperglycemia discovered during a routine physical. Due to strong FH of diabetes, family requested a screening test for him. His random MBG was 230 mg/dl and urine was 1+ for glucose. He denied any symptoms of DM such as weight loss, polyuria, polydipsia, nocturia and/or enuresis. ROS otherwise negative.

Past medical history: non-contributory.

PE: Healthy appearing, robust adolescent.

FH: Brother and Sister Diagnosed with T1DM-Rx with Insulin-Father Dxed with DM at age 32.

Admitted: BG 252mg/dl; insulin 29uU/ml; C-peptide 3.58 ng/ml

CASE 1 CONT.

Father has 2 sisters diagnosed with DM-one T1DM and other T2DM

Father treated with Metformin and glipizide and runs HbA1c of 6-6.5%-no complications after 30 years of DM.

Father recalls that his father died from leukemia but during treatment was found to have "high blood sugar"

MODY3: HNF-1alpha

- Expressed in liver and beta cells
- HNF-4alpha regulates HNF-1alpha
- Link between MODY1 and MODY3
- Progressive insulinopenia with incr. age
- Birth Weight: decr. ~120 gms
- Common cause of MODY; =>120 mutations
- HNF-1alpha: uncommon cause of T2DM

MODY1: HNF-4alpha (Chr.: 20q12-q13.1)

- First TF mutation discovered
- Expressed: liver, kidney, intestine & islets
- Key regulator of hepatic gene expression
- Progressive insulinopenia with incr. age
- Decreased apo C2, apo C3, Lp (a)
- Uncommon cause of MODY; =>8 mutations
- HNF-4alpha: uncommon cause of T2DM
Macrosomia and HHI in patients with heterozygous mutations in HNF-4A gene

- Conclusions: HNF4A plays role in BW and shows a phase of HI and HHI may precede evolution to hypoinsulinemia and MODY later in life

Pearson EW et al PLOS Med 2007;4:e118

MODY2: GLUCOKINASE (Chr. 7p15-p13)

- Decreased beta cell sensitivity to glucose: insulin is released at increased glucose concentrations
- Common cause of MODY; >120 mutations
- First molecular cause identified
- GCK: uncommon cause of type 2 diabetes (true for all forms of MODY)

MODY5: HNF-1beta

- Japanese MODY families lacking HNF-4alpha, HNF-1alpha, IPF-1 mutations: discovered linkage to HNF-1beta in 2 families
- HNF-1beta regulates HNF-4alpha
- Link between MODY5 and MODY1
- Homo or heterodimer with HNF-1alpha
- Renal cysts, vaginal/uterine malformations, abnormal liver function, nondiabetic renal disease
- Mutations: R177X; A263fsinsGG
Proteinuria in an Adolescent with Diabetes

- A 17-year 4-month-old female considered to have type 1 diabetes since February 2003.
- She has maintained meticulous metabolic control with all her HbA1Cs in the better than 7% range.
  - September 2008: 6%.
  - March 2009: 5.8%.

Proteinuria in an Adolescent with Diabetes©

- The early development of albuminuria within 3 years of diagnosis, despite excellent metabolic control as judged by consistent HbA1c values of less than 6.5%, led to suspicion of MODY5 and the discovery of a novel change in intron 2 of the TCF2 gene, at a location that possibly alters a splice site resulting in an altered protein product. Significance under investigation.

MODY7

- several genes proposed
  - Islet-1 (ISL-1); 5q11-q13: Lim domain homeobox gene; TF: regulates insulin expression
  - Carboxyl ester lipase (CEL); 9q34.2: bile salt-stimulated lipase
  - Kruppel-like factor 11 (KLF11); 2p25: SP1-like zinc finger transcription factor

Results of 2.5 year prospective surveillance for monogenic diabetes at Children’s Hospital Pittsburgh 1/07-7/09

- 5 families with MODY 3
- 9 families with MODY 2
- 2 families with MODY 5
- TOTAL OF 16 FAMILIES

MODY: maturity onset diabetes of youth/monogenic diabetes of youth

- autosomal dominant (single gene)
- onset before ~25 to ~35 y/o
- nonketotic, non-insulin dependent
- usually lean
- generally Caucasian/Caucasoid
MODY: DIFFERS FROM T1DM
- MODY:
  - nonketotic
  - lacks islet autoantibodies (ICA, GADA, IA-2A)
  - No incr. frequency of high-risk alleles for T1DM (DR3, DR4, DQB1*0201, DQB1*0302)

MODY: DIFFERS FROM T2DM
- MODY:
  - usually lean
  - insulinopenic diabetes
  - normal insulin sensitivity

FIG. 1. Chronological accounting of the discovery of type 2 diabetes-associated genes, plotted by year of definitive publication and approximate effect size

NEONATAL DIABETES MELLITUS: CASE HISTORY A
- Born to healthy 30 yr old mother @ 35-5/7 W-
- B. weight 2045 gm=IUGR diagnosed by U/S in-utero
- Developed hyperglycemia in first 36 hrs of life-
- Persistence required insulin-MRI showed pancreas
- By age 48d WT 2.82 KG- Breast feeding.
- Parents trained to monitor and use insulin pump
- At age 80 d 2.3 U/day=0.5 U/kg/d 45% basal-WT 4.7 KG-
- Length 54.5 CM-No dysmorphic features
- Insulin d/c at age 4 months—
- Genetic analysis showed uniparental disomy of father's chromosome 6
  - dx: classical transient form of NDM

NEONATAL DIABETES MELLITUS: CASE HISTORY B
- Born FT, unremarkable prenatal course, birth weight: 5 lbs, 8 oz
- At 3 weeks of age: started having episodes of vomiting and multiple formula changes.
- At 9 weeks of age: increased episodes of vomiting, decreased PO intake, lethargy, respiratory distress — presented in shock, DKA:
  - Glucose 1007 mg/dl, pH: 6.9, K: 6.9 meq/l, Na 166 meq/l, BUN 62 mg/dl and creatinine 1.3 mg/dl
  - Base deficit - 29, HbA1 15.8%, HbA1c 11% (post transfusion).
  - Initially intubated, mechanically ventilated for 3 days, Rx of DKA and hypernatremic dehydration.
  - Subcutaneous insulin then maintained on insulin CSII requiring about 0.8 units/kg/day of insulin.

NEONATAL DIABETES MELLITUS: CASE HISTORY C
- Born prematurely and with IU agr
- Multiple congenital anomalies—truncus arteriosus with Ebstein anomaly, cholestasis/biliary atresia, NEC
- Glucose intolerance/diabetes recognized by day 3—IV insulin and glucose given as treatment
- Infant died—parents refused genetic analysis
- Likely permanent neonatal diabetes mellitus with unknown genetic basis
NEONATAL DIABETES MELLITUS: KEY FEATURES

- MOST ARE IUGR/SMALL REFLECTING ROLE OF INSULIN AS IN-UTERO GROWTH FACTOR
- COURSE IS VARIABLE- GLUCOSURIA/POLYURIA, DEHYDRATION, FTT, DKA MILD-SEVERE, INSULIN/IGF-1 LOW
- DYSMORPHIC FEATURES IN SOME INCLUDING: MR, MUSCLE WEAKNESS, EPILEPSY- THESE ARE ONLY FOUND IN THE PERMANENT FORMS.
- Rx WITH INSULIN RESULTS IN DRAMATIC CATCH-UP GROWTH- AFTER SEVERAL WEEKS TO MONTHS INSULIN MAY BE WITHDRAWN IN ABOUT 1/2 OF CASES (TRANSIENT NEONATAL DM) WITH RECURRENCE IN ABOUT 25% SEVERAL YEARS LATER. ABOUT ONE HALF HAVE PERMANENT DM FROM OUTSET.

SUMMARY

- THE SAME GENES THAT ARE RESPONSIBLE FOR NEONATAL DIABETES AND MODY CONTRIBUTE TO TYPE 2 DM AND SOME CASES OF APPARENT T1DM
- CORRECT DIAGNOSIS PROFONDLY AFFECTS TREATMENT, GENETIC COUNSELING AND PROGNOSIS.
- PROBABLY 2% +/- OF ALL CHILDREN DIAGNOSED WITH DM MAY HAVE A MONOGENIC FORM

KEY POINTS

- Most neonatal patients with mutations in the potassium-sensitive ATP channel subunits Kir6.2 and sulfonylurea receptor 1 will be best treated with high-dose sulfonylureas rather than insulin injections, despite seeming insulin dependent
- Patients with glucokinase mutations have stable, mild, regulated hyperglycemia throughout life and do not need pharmacological treatment except possibly during pregnancy
- Patients with mutations in HNF1A have hyperglycemia that deteriorates with age and that can be severe; these patients, like patients with mutations in HNF4A, are sensitive to the hypoglycemic effects of sulfonylureas

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SHADI TABBA
KARA HUGHAN
JAIME HAIDET

RESOURCES:

www.diabetesgenes.org
www.kovlerdiabetescenter.org/registry
www.mody.no
www.genetests.org

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References

Sperling MA: The genetic basis of neonatal diabetes mellitus
Ped. Endo reviews 2006;Suppl :71-75

Sperling MA: ATP-sensitive potassium channels--neonatal
2006;355:507-10

Sperling MA: Neonatal diabetes mellitus: from understudy to
center stage.
Current Opinion in Pediatrics 2005;17:512-18

Murphy et al. Clinical implications of a molecular genetic
classification of monogenic beta-cell diabetes
Nat Clin Pract Endocrinol Metab. 2008;4:204

McCarthy MI, Hattersley A: Learning from molecular
genetics: novel insights arising from the definition of
genes for monogenic and type 2 diabetes. Diabetes. 2008
Nov;57(11):2889-98.