Diabetes mellitus

Regulation of glycemia

• humoral
  • principal
    • insulin
    • glucagon

• auxiliary
  • glucocorticoids
  • adrenaline
  • growth hormone

• neural
  • sympathetic
    • hyperglycemia
  • parasympathetic
    • hypoglycemia

Main contra-regulation: insulin/glucagon

Definition of diabetes mellitus (DM)

• DM is a group of metabolic disorders characterized by hyperglycemia resulting from a lack of insulin effect
  • due to either defect in insulin secretion or insulin action

• chronic hyperglycemia leads to long-term cell, tissue & organ damage = diabetic complications
  • retina
  • kidney
  • nerves
Diagnosis of DM

- diabetes
  - classical symptoms + random plasma glycemia ≥11.1 mmol/l (venous plasma)
    - random = any time of the day
    - symptoms include polyuria and polydipsia
  - FPG (fasting plasma glucose) ≥7.0 mmol/l
    - fasting means at least 8 h from the last meal
  - 2-h PG (postprandial glucose) ≥11.1 mmol/l during oGTT
    - oGTT: according to the WHO consists of FPG examination followed by a standard load of 75g of glucose (diluted in water) and examination of glycemia in 60th and 120th minute

- impaired glucose tolerance (IGT)
  - excluded <7.8 mmol/l
  - 2-h PG ≥7.8 - <11.1 mmol/l during oGTT

- impaired fasting glucose (IFG)
  - diabetes excluded by FPG ≤5.6 mmol/l
  - FPG ≥5.6 – <7 mmol/l

Interpretation of glycemia

<table>
<thead>
<tr>
<th>Glycemia (mmol/l)</th>
<th>Normal</th>
<th>IFG</th>
<th>IGT</th>
<th>Diabetes</th>
</tr>
</thead>
<tbody>
<tr>
<td>FPG</td>
<td>3–6</td>
<td>5.6</td>
<td>7.0</td>
<td>&gt; 7.0</td>
</tr>
<tr>
<td>60 min</td>
<td>4–7</td>
<td>5.6</td>
<td>7.8</td>
<td>&gt; 11.1</td>
</tr>
<tr>
<td>120 min</td>
<td>4–7</td>
<td>5.6</td>
<td>7.8</td>
<td>&gt; 11.1</td>
</tr>
</tbody>
</table>

The way glucose enters the cell??

- exocytosis from B-cells of islets of Langerhans into portal circulation
  - 50% degraded during first pass through liver
  - parallel cleavage of the C-peptide
  - total daily production in healthy subject: ~20–40 U
    - 1/2 basal (postabsorptive) secretion
      - pulsatile (5 - 15 min intervals)
    - 1/2 stimulated (postprandial)
      - early phase (ready insulin)
        - Glc/KATP-dependent
      - late phase (synthesis de novo)
        - other secretagogues

- stimulation of secretion
  - <<glucose
  - <<amino acids
  - <GIT hormones (incretins)
  - FFA
    - variable stimulation (length of chain & (un)saturation)!
    - since insulin is acting also as peripheral "satiety" signal, reaching the satiety is delayed after fatty meal
Insulin synthesis

C peptide

- activity
  - certain beneficial vascular effects (nitric oxide)
- mainly diagnostic use
  - equimolar to insulin
  - unlike insulin, C-peptide is not degraded from portal blood in liver
  - the systemic concentration reflects endogenous insulin production

Coupling: glycemia – insulin secretion

Summary: coupling of glycemia → biphasic insulin secretion
**Incretins – enteroinsular axis**

- GIT hormones produced by endocrine cells of small intestine stimulating insulin secretion even before elevation of blood glucose
  - Ins-secretion after oral Glc >> after i.v. Glc
  - Hypoglycemia – if the patient still conscious then better to give Glc per os
  - “forward” regulatory mechanism – anticipation of increase of Glc
- 2 major incretin hormones
  - GIP (glucose-dependent insulinotropic peptide or gastric inhibitory peptide)
  - GLP-1 (glucagon-like peptide-1)
- Treatment of T2DM (delayed effect of Glc on Ins stimulation) by incretin analogues
  - GLP-1 analogue - exenatide (GLP-receptor agonist)
  - DPP-4 inhibitors (dipeptidyl peptidase 4 - proteolytic degradation of incretins) - gliptins
  - Improvement of Glc-stimulated Ins secretion after meal
  - Supression of postprandial glucagon release
  - Delayed gastric emptying
  - Protection of β-cells from apoptosis

**NEFA and insulin secretion**

- NEFA can enter cells (incl. B-cells)
  - Directly by diffusion across the membrane (short-chain FA) → metabolism (oxidation) → ATP ...
- Via receptor (GPR40) → see the figure
- However, long term exposure to NEFA, esp. long-chain saturated (e.g. palmitate), suppress secretion of insulin and damages B-cells

**Insulin receptor**

**Insulin receptor signal cascade**

- Insulin receptor
  - Tyrosine kinase type (2 α and 2 β subunits)
  - (1) Cascade of phosphorylations (down-stream kinases)
    - Balanced activation or inhibition of hormones
      - Activation of anabolic pathways (i.e. glycogenogenesis, lipogenesis)
      - Inhibition of catabolic pathways (e.g. lipolysis, glycogenolysis) and gluconeogenesis
    - (2) Translocation of GLUT4
Classification of tissues according to insulin action:
- **insulin-sensitive**
  - skeletal and heart muscle
  - adipose tissue
    - glucose uptake facilitated by GLUT4, which becomes integrated into cell membrane after insulin receptor activation
  - liver
    - metabolic actions
- **insulin-insensitive**
  - all others incl. muscle, adipose and liver
    - glucose uptake is realized by facilitated diffusion by GLUT1, 2, 3, 5, ...
    - permanently localized in the cell membrane
    - transport of glucose depends solely on concentration gradient
    - type and density of GLUTs

Diabetes mellitus
- **heterogeneous** syndrome characterized by **hyperglycemia** due to **deficiency of insulin action** as a result of
  - absolute insulin deficiency
    - destruction of the β-cells of the islets of Langerhans
  - relative deficiency of insulin secretion and/or action
    - abnormal molecule of insulin (mutation of insulin gene)
    - defective conversion of preproinsulin to insulin
    - circulating antibodies against insulin or its receptor
  - insulin resistance in peripheral tissues + secondary failure of β-cells of the islets of Langerhans
    - receptor defect
    - post-receptor defect
  - prevalence of DM in general population 5%, over the age of 65 already 25%

Prevalence (%) of diabetes (population 20-79 years)

<table>
<thead>
<tr>
<th>Year</th>
<th>Total Population</th>
<th>Diabetics</th>
<th>Prevalence</th>
</tr>
</thead>
<tbody>
<tr>
<td>2010</td>
<td>4.3 bil.</td>
<td>285 mil.</td>
<td>5%</td>
</tr>
<tr>
<td>2030</td>
<td>5.6 bil.</td>
<td>438 mil.</td>
<td>30%</td>
</tr>
<tr>
<td></td>
<td>(from a total of 7 bil.)</td>
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Classification of DM

1. **Diabetes mellitus type 1 (T1DM)** ~5%
2. **Diabetes mellitus type 2 (T2DM)** ~90%
3. Other specific types:
   a. genetic defects of B-cell
      - monogenic DM (MODY1 - 6)
      - mutation of mitochondrial DNA
   b. genetic defects leading to insulin resistance
      - type A insulin resistance, leprechaunismus, Rabson-Mendenhall syndrome, lipoatrophic DM
   c. diseases of exocrine pancreas
      - pancreatitis, tumor, cystic fibrosis, hemochromatosis
   d. endocrinopathies
      - Cushing syndrome, acromegaly, pheochromocytoma, hyperthyreosis
   e. iatrogenic DM (i.e. drugs and toxins)
   f. other genetic syndromes associated with DM
      - Down, Klinefelter, Turner syndromes, ...
4. **Gestational diabetes mellitus**
**T1DM (formerly IDDM)**

- selective autoimmune destruction of β cells of IofL in genetically predisposed individuals
- genetic susceptibility
  - chromosome 6 – MHC class III
    - DR3-DQ2 and DR4-DQ8
  - chromosome 11 - insulin gene
- cytotoxic autoimmunity mediated by T-lymphocytes
  - there are also antibodies against β cell structures (ICA, GAD, IAA), but they are rather markers of autoimmunity than causal agent

**T1DM**

- autoimmunity has to be triggered by various factors
  - infection
    - viruses
      - rubella, measles, coxsackie B, CMV, EBV, enteroviruses, retro-viruses
    - mechanism is unclear
      - cytolytic (β sequestration of antigens
      - formation of neoantigens
      - molecular mimicry or superantigens
  - environmental factors (according to the epidemiologic evidence)
    - diet – early exposition proteins of cow’s milk
      - bovine insulin
    - vitamin D – reason for northern-southern geographical gradient?
      - toxins (diet, water, bacteria)
      - gluten???
  - manifestation typically in childhood
  - absolute dependence on exogenous supplementation by insulin

**Natural history of T1DM**

[Graph showing the natural history of T1DM]

- insulin sensitivity is a continuous trait with distinct interindividual variability, it can be assessed by:
  - hyperinsulinemic euglycemic clamp
  - calculated indexes (based on relationship between glycemia and insulin during fasting or oGTT) – e.g. HOMA, QUICKI, ...
- main pathophysiologic feature of T2DM is an imbalance between insulin secretion and its effect
  - in the time of clinical manifestation there are both insulin resistance and impairment of insulin secretion
  - what is "chicken" and what is "egg"??
  - insulin resistance
    - genetic predisposition (polygenic) – thrifty phenotype
    - acquired factors
      - competition of Glu with NEFA!! (diet)
      - effect of adipokines from adipose tissue (obesity)
      - ↓ mobilisation of GLUT4 in physical inactivity
      - ↓ down-regulation of Ins receptor due to hyperinsulinemia
  - impairment of secretion
    - inherited factors
      - fewer B-cells (~20-40%)
    - defect of 1. phase of Ins secretion (~80% reduction)
    - defect of 2. phase of Ins secretion (~80% reduction)
  - acquired factors
    - gluc- and lipotoxicity for B-cells
    - 90% of subjects are obese – metabolic syndrome!!!

**T2DM (formerly NIDDM)**

- insulin sensitivity is a continuous trait with distinct interindividual variability, it can be assessed by:
  - hyperinsulinemic euglycemic clamp
  - calculated indexes (based on relationship between glycemia and insulin during fasting or oGTT) – e.g. HOMA, QUICKI, ...
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Pathogenesis of T2DM

Secondary failure of β cells

- hyperglycemia induces:
  - oxidative stress
  - endoplasmic reticulum (ER) stress
- high concentration of NEFA causes lipotoxicity
  - short term increase of NEFA stimulates secretion of insulin
  - long term exposure to NEFA, esp. long-chain saturated (e.g. palmitate), suppress secretion of insulin and damages B-cells
- ↑ ceramide → apoptosis

Natural history of T2DM

ER stress → Unfolded protein response

- ER stress → unfolded protein response (UPR) is activated in response to an accumulation of unfolded or misfolded proteins in the lumen of ER
  - incl. insulin in β-cells
- UPR has two primary aims:
  - initially to restore normal function of the cell by halting protein translation and activate the signaling pathways that lead to increasing the production of molecular chaperones involved in protein folding
  - if these objectives are not achieved within a certain time lapse or the disruption is prolonged, the UPR aims to apoptosis
Insulin- and “sport”-dependent translocation of GLUT4

• 2 intracellular “pools” of GLUT4
  • insulin-dependent (see cascade of Ins-receptor)
  • Ca²⁺ / NO / AMPK?-dependent
    • this mechanism is responsible for improvement of insulin sensitivity in physically active subjects

Main characteristics of T1DM and T2DM

<table>
<thead>
<tr>
<th></th>
<th>T1DM</th>
<th>T2DM</th>
</tr>
</thead>
<tbody>
<tr>
<td>onset</td>
<td>childhood</td>
<td>adults</td>
</tr>
<tr>
<td>genetic disposition</td>
<td>yes (oligogenic)</td>
<td>yes (polygenic)</td>
</tr>
<tr>
<td>clinical manifestation</td>
<td>often acute</td>
<td>mild or none</td>
</tr>
<tr>
<td>autoimmunity</td>
<td></td>
<td></td>
</tr>
<tr>
<td>insulin resistance</td>
<td>yes</td>
<td>No</td>
</tr>
<tr>
<td>dependence on insulin</td>
<td>yes</td>
<td>No</td>
</tr>
<tr>
<td>obesity</td>
<td>no</td>
<td>yes</td>
</tr>
</tbody>
</table>

Other types of DM

• LADA (Latent Autoimmune Diabetes in Adults) = slow-onset T1DM
  • diagnosta > 30 let věku, zpravidla mylně jako T2DM
  • zpočátku dobře kontrolovatelná dietou a PAD, nevede k diabetické ketoacidóze
  • postupně však závislost na inzulinu (měsíce – 1 rόk)
  • pozitivní protitátky (autoimunity), nízký C-peptid
  • negativní rodinná anamnéza T2DM

• MODY (Maturity-onset diabetes of the young) – cca 5% T2DM
  • skupina monogenich diabetů s familiárním výskytem a dobře definovaným mendelisčtěm způsobem dědictví (zpravidla AD), časnou manifestaci (dětství, adoleseční či časná dospělost) a bez vztahu k obezitě
  • cca 6 typů (MODY1-6)
  • patofyziology: geneticky podmíněná dysfunkce β-buněk, ale dlouhodobě měřitelný C-peptid bez známek autoimunity
  • dvě podskupiny
    • MODY v důsledku mutací v glukokináze (MODY2)
      - glukokináza = „glukózový sensor“ (vzrůstá uvolňování a produkce inzulinu)
      - forma bez výrazného ztráty žlázy komplikací
    • MODY v důsledku mutací v transkripčních faktorech (ostatních 5 typů)
      - silné defekty β-buněk pravděpodobně vedoucí k diabetu se závažnými pozdními následky
      - postižená glukóza stimuluje tvorbu a uvolnění inzulinu a příbuzace a diferenciace β-buněk

<table>
<thead>
<tr>
<th>MODY</th>
<th>locus</th>
<th>gen</th>
<th>produkt</th>
<th>prim. defekt</th>
<th>závaznost</th>
<th>komplikace</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>20q</td>
<td>GCK</td>
<td>glukokináza</td>
<td>pancreas/játra</td>
<td>mírná</td>
<td>vzácná</td>
</tr>
<tr>
<td>2</td>
<td>7p</td>
<td>ITCF1 (HNF1A)</td>
<td>hepatocyte nuclear factor-1</td>
<td>pancreas</td>
<td>vysoká</td>
<td>časté</td>
</tr>
<tr>
<td>3</td>
<td>17q</td>
<td>ITCF2 (HNF4G)</td>
<td>hepatocyte nuclear factor-1</td>
<td>pancreas/ledviny</td>
<td>vysoká</td>
<td>časté</td>
</tr>
<tr>
<td>4</td>
<td>13q</td>
<td>IPF1</td>
<td>insulin promoter factor-1</td>
<td>pancreas</td>
<td>vysoká</td>
<td>časté</td>
</tr>
<tr>
<td>5</td>
<td>15q</td>
<td>NEUROD1</td>
<td>hepatocyte nuclear factor-1</td>
<td>pancreas/ledviny</td>
<td>vysoká</td>
<td>časté</td>
</tr>
<tr>
<td>6</td>
<td>2q32</td>
<td>NEUROD1</td>
<td></td>
<td>pancreas</td>
<td>vysoká</td>
<td>časté</td>
</tr>
</tbody>
</table>

Diabetic “triumvirate” ???
Acute manifestation and long-term consequences (complications) of diabetes

Effect of rising plasma glucose ???

OSMOLARITA = 2 Na⁺ + urea + glukóza
275 - 295 = 2 x 140 + 2.5 + 5
> 300 = 2 x 140 + 2.5 + 35

Clinical presentation of DM

- due to the mild increase of blood osmolarity, osmotic diuresis and dehydration
  - classical
    - polyuria, thirst, polydipsia
    - tiredness
    - temporary impairment of vision
  - others
    - recurrent infections
    - perio-/parodontitis
- extreme hyperglycemia (>40 mmol/l, osmolarity >350 mosmol/l)
  - ketoacidosis/coma
    - ↑ ketone bodies, metabolic acidosis and hyperglycemia
  - non-ketoticidotic hyperglycemic coma
    - hyperglycemia, dehydration and pre-renal uremia
  - lactic acidosis/coma
    - either complication of therapy (biguanides / type of peroral antidiabetics)
    - associated with hypoxic states (sepsis, shock, heart failure, ...)

Diabetic ketoacidosis
Late complications of DM

- microvascular
  - diabetic retinopathy
  - diabetic nephropathy
  - diabetic neuropathy
    - sensoric
    - motoric
    - autonomous
- macrovascular
  - accelerated atherosclerosis (CAD, peripheral and cerebrovascular vascular disease)
- combined
  - diabetic foot (ulcerations, amputations and Charcot’s joint)
- others
  - periodontitis
  - cataract
  - glaucoma

Chronic hyperglycemia

- Advanced glycation end products (AGEs)
  - cross-linking of extracellular proteins
  - modification of intracellular proteins and DNA
    - ubiquitin/proteasome
  - binding to pattern-recognition receptors and activation of signaling pathways