Mouse model for PMM2-CDG

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Mouse as a disease model

Uses
• Determine genetic causation
• Understand pathophysiology
• Use as a preclinical model
Mouse as a disease model

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• Determine genetic causation
• Understand pathophysiology
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Advantages
• Genetic similarities with humans
• Tools for genetic manipulation
• Lifespan/Reproduction rate
• Size/Ease of handling/Cost
Mouse as a disease model

Uses
- Determine genetic causation
- Understand pathophysiology
- Use as a preclinical model

Challenges
- Phenotypes
- Alternative physiology
- Predictive validity
- Inbred strain differences
Types of genetic mouse models

- Transgenic
Types of genetic mouse models

• Transgenic

• Knockin
Types of genetic mouse models

• Transgenic
• Knockin
• Knockout
Types of genetic mouse models

• Transgenic
• Knockin
• Knockout
• Conditional knockout
Current mouse models for PMM2-CDG

Knockout

- *PMM2* gene disruption
- Homozygous lethal around E2.5
- Functional glycosylation machinery is essential in early development

Thiel et al. 2006. Mol Cell Biol. 26:5615-20
Current mouse models for PMM2-CDG

**Knockin #1 and #2**
- **#1 - R141H (R137H in mice)**
  - Most common human mutation
- **#2 - F122L (F118L in mice)**
  - Synthetic predicted mild mutation
- **$Pmm2^{R137H/R137H}$**
  - Embryonic lethal before E5.5
- **$Pmm2^{F118L/F118L}$**
  - Viable, fertile, without major phenotypes
  - PMM2 enzyme activity ~ 40% WT

Current mouse models for PMM2-CDG

Knockin #1/#2
- $Pmm2^{R137H/F118}$
  - Embryonic lethal ~E9.5-E10.5
  - Small
  - Tissue degradation
  - PMM2 enzyme activity = 11% WT
  - Rescue with mannose to mothers

Knockin #3

- #3 - F119L (F115L in mice)
  - 2nd most common human mutation
- \( Pmm2^{F115L/F115L} \)
  - Embryonic lethal in more than \( \frac{1}{2} \)
  - Rescue with mannose to mothers

Current mouse models for PMM2-CDG

Knockin #1/#3

- $Pmm2^{R137H/F115L}$
  - Most common human genotype
    - (R141H/F119L)
  - ½ embryonic lethal
  - No effect of mannose

Knockin #1/#3

- **Pmm2**<sup>R137H/F115L</sup>
  - ½ die by postnatal day 65
  - Small
  - Hypotonia in hind limbs (6%)
  - Curvature of the back (29%)
  - Heart, liver, kidney abnormalities
  - Decreased glycosylated plasma proteins
  - Normal plasma transferrin glycosylation
  - PMM2 enzyme activity = 15-16% WT
  - No histologic brain abnormalities
Current mouse models for PMM2-CDG

Knockin #1/#3

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Conclusions

• Range of phenotypes from early embryonic lethality to normal

• *Pmm2*<sup>R137H/F115L</sup>
  • Most common patient genotype
  • Reproduces several human PMM2-CDG phenotypes
  • Available through Jackson Labs
  • Significant embryonic lethality
  • Significant postnatal lethality
  • No histologic brain phenotype
New mouse model for PMM2-CDG
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- Conditional knockout allele
- Floxed $PMM2$ exon 3
- Remove exon 3 with Cre

$Pmm2^{tm1c(EUCOMM)Wtsi}$
New mouse model for PMM2-CDG

- Conditional knockout allele
- Floxed *PMM2* exon 3
- Remove exon 3 with Cre
- IMPC validated embryonic lethal

*Pmm2*^{tm1c(EUCOMM)Wtsi}
New mouse model for PMM2-CDG

• Generate conditional knockout/knockin mice
• Generate cell type-specific Pmm2 deficiency with Cre
• Avoid embryonic lethality
• Model severe Pmm2 deficiency in isolated cells/organs for pathophysiologic studies
• Neurologic phenotypes
Cre-dependent ZsGreen characterization of Snap25-IRES-Cre
New mouse model for PMM2-CDG

- Currently breeding $Pmm2^{R137H/+}$, $Pmm2^{fl/fl}$ and $Snap25^{Cre/+}$ together
- $Pmm2^{fl/fl}; Snap25^{Cre/+}$ mouse
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