In Vivo Animal Models of Heart Disease

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Why Animal Models of Disease?

• Heart Failure (HF)
  – Leading cause of morbidity and mortality in the US
  – Prevalence of associated risk factors such as diabetes, hypertension, obesity, high cholesterol, inactivity and aging is increasing
  – Current treatments only slow the progression of the syndrome
  – Thus, there is a great need to develop novel preventative and reparative therapies
  – Therefore, appropriate animals are necessary.
What is Heart Failure?

• Clinical syndrome
  – Dyspnea
  – Fatigue
  – Exercise intolerance
  – Retention of fluid in the lungs and/or peripheral tissues.

• Fundamental Defect
  – Impaired filling and/or ejection of blood from the heart

Common Causes of HF

• Valvular lesions
• Dilated cardiomyopathies
• Hypertensive heart disease
• Restrictive cardiomyopathies
What makes a good animal model?

• Should mimic critical features of human HF
  – Considerations
    • Mimic the course of HF for the duration
    • Mimic for a single discrete time point
  – Limitations
    • Lack of diversity
    • Lack of co-morbidities
    • Differences between species in cell and molecular make-up

Aortic stenosis (AS)

• Atherosclerotic disease w/wo calcification
• Calcification independent of atherosclerosis
• Valve malformations
• Bottom line
  – increased valve stiffness and reduced ejection orifice area leads to increased LV afterload
Critical Features of AS

- LV-AO pressure gradient
- LVH with increased myocyte x-sectional area preserved systolic function
- Fibrosis
- Increased filling pressure
- Diastolic dysfunction
- Reduced systolic function
- Increased fibroblasts
  - Increased extracellular matrix
  - Collagen accumulation

Animal Models of AS

- **Mouse TAC**
  - Abrupt increase
    - activation of growth regulatory pathways are different
    - contractile protein are different in mice
    - extracellular remodeling is different in mice
  - Test specific molecules

- **Rat TAC**
  - Start with young rats to avoid the abrupt increase in pressure

- **Dog/Pig**
  - Ameroid constriction of aorta
Mitral Regurgitation

Chronic and progressively increasing EDV and LA

Eccentric LVH

Disruption/loss of myocardial matrix

Severity = (regurgitant volume/total stroke volume)100

Animal Models of MR

• Rodents
  – Aortocaval fistula
  – AO insufficiency
    • both lead to volume overload
    • Both feature LV eccentric hypertrophy

• Large animal
  – Severing chordae tendineae
    • Leads to volume overload
    • LV eccentric hypertrophy
    • Myocardial matrix disruption
Unresolved Issues in Valvular Lesions

• How to enhance cardiac repair after correction of the valve defects?
  – Models that correct the defect
    • Unbanding?
    • Repair of mitral valve (large animals)?
    • Repair of aortic valve (large animals)?
    • Fistula correction?

Dilated Cardiomyopathy

• Ventricular dilation, systolic dysfunction, abnormalities of diastolic filling, normal or reduced wall thickness (eccentric hypertrophy), increased diastolic and systolic wall stress, biventricular and bialtrial enlargement, AV valve regurgitation, elevation of left and right sided filling pressures, increases in organ and chamber weight, myocyte hypertrophy, activation of neurohormonal systems
Causes

• Genetic mutations (cytoskeletal, sarcolemmal, scarcomeric, nuclear envelope proteins), MI, longstanding hypertension, CAD, myocarditis, some chemotherapeutic drugs, autoimmune disorders, excessive tachycardia, endocrine disorders, excessive alcohol consumption, nutritional deficiencies, neuromuscular disorders

Critical Similarities

• Hemodynamics
  – decreased systolic and diastolic function
  – diminished contractile reserve (catecholamine stress)
  – increase wall stress
  – depressed isovolumic ejection phase
  – slowed relaxation rate
  – depression of stretch induced force response
  – blunting of force frequency response
  – altered Ca uptake storage and release
  – altered beta adrenergic receptor function
Similarities

• Molecular hallmarks
  – activation of fetal/hypertrophic gene program
  – local and systemic inflammation
  – oxidative stress
  – upregulation of atrial natriuretic factor
  – Down regulation
    • SR calcium ATPase
    • alpha myosin heavy chain
    • Beta 1 adrenergic receptors

Similarities

• Histopathologic
  – Hypertrophy
    • myocyte length and width
    • interstitial and replacement fibrosis
    • alteration of extracellular matrix
    • progressive cardiomyocyte death (apoptosis, necrosis and autophagy)
    • relative capillary reduction.
Similarities

• Neurohormonal
  – increased adrenergic tone
  – increased renin-angiotensin–aldosterone systems
  – increased endothelin
  – increased vasopressin

Animal Models of DCM

• Rodents
  – MI in rodents
    • MI size is hard to control
  – Genetic models in mice
  – doxorubicin or isoproterenol (dilated phenotype and myocardial injury and cell death (apoptosis and oxidant stress)
  – Salt sensitive hypertensive rats
    • often little change in heart structure/function
Animal Models of DCM

- **Large animals**
  - MI (pigs)
  - coronary microembolization
    - contractile dysfunction
    - localized inflammatory responses
    - TNF expression
    - progressive LV dilation
    - Long time to produce, hard to titrate
  
  - pacing induced tachycardia
    - reliable and reproducible model
    - partially reversible over time

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<thead>
<tr>
<th></th>
<th>Advantages</th>
<th>Limitations</th>
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<tbody>
<tr>
<td><strong>Mouse</strong></td>
<td>inexpensive</td>
<td>partial resemblance to humans</td>
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<td></td>
<td>genome manipulation</td>
<td>more atherosclerotic</td>
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<td></td>
<td>atherosclerotic plaques</td>
<td>alpha myosin heavy chain</td>
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<td>similar developmental cardiac anatomy</td>
<td>variable coronary circulation</td>
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<td></td>
<td>few collaterals</td>
<td>fast HR</td>
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<td>resistant to arrhythmias</td>
<td>no plateau phase in AP</td>
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<td>rapid inflammation course</td>
<td>Na/Ca less relevant in rodents</td>
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<td>transient macrophage infiltration</td>
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<td>tolerate large MI</td>
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<td><strong>Rat</strong></td>
<td>inexpensive</td>
<td>No athermo</td>
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<td>useful for restenosis</td>
<td>alpha myosin heavy chain</td>
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<td>few collaterals</td>
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<td>tolerate large MI</td>
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<td>Lewis rats (consistent coronary arteries)</td>
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<td><strong>Rabbit</strong></td>
<td>medium size</td>
<td>need high blood cholesterol levels</td>
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<td>no plaque rupture model</td>
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<td>complex plaques</td>
<td>more neointima formation than atherosclerosis</td>
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<td>restenosis</td>
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<td>beta myosin heavy chain</td>
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<td>similar Ca uptake</td>
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<td>Pig</td>
<td>lesions more similar to human valid for restenosis beta-myosin heavy-chain</td>
<td>expensive difficult handling some genomic tools susceptible to arrhythmias</td>
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<td>some co-morbidities useful for device testing few collaterals more</td>
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<td>consistent MI sizes angioplasty induced injury very similar to humans</td>
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<td>with high fat diet coronary vascular very similar</td>
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<td>Dog</td>
<td>b-myosin heavy-chain</td>
<td>expensive many collaterals variable infarct size</td>
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Large animal models

- Preclinical validation studies
- Testing of devices
- Similar end-point measurements
  - Structural
  - Hemodynamic
  - Physiological assessments
  - Histopathologic, biochemical, cellular, molecular studies should be used to document DCM and improvements
General Considerations

- Cost
- Housing/maintenance
- How to measure end-points?
  - Imaging (echo, MRI, CT, PET, x-ray, molecular)
    - Volumes, function
  - Invasive and implantable physiology
    - PV loops, ECG, pressures, pacing wires, flows,
- Surgical methods (open chest, closed chest)
- Cell delivery methods
- Multiple animal models
- Co-morbidities
- Age
- Male vs. Female

Conclusions

- Randomization
- Blinding
- No one size all fits models
- Careful experimental design
- Choose your models carefully
- Multiple animal models, use Co-morbidities, ages
Conclusions

• Beta-adrenergic receptor antagonists are a great success story that have been translated from animal models of heart failure to humans

• Cell therapies will follow this same path