Objectives & key points

- Identify contractile and regulatory proteins
- Describe excitation-contraction coupling
- Recognize the role and source of Ca in muscle contraction
- Identify energy sources for muscle contraction
- Describe mechanisms that regulate muscle contraction & relaxation
- Contrast structural, energetic & regulatory aspects of 3 muscle types

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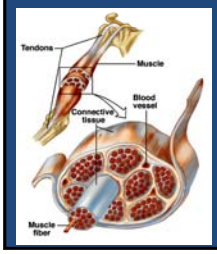
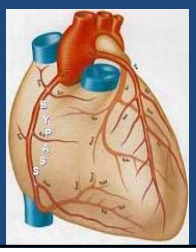
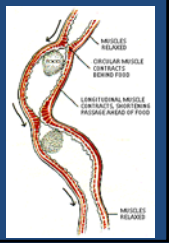
Outline of muscle lectures

- Structure
- Excitation-contraction coupling
- Energetics & mechanics of contraction



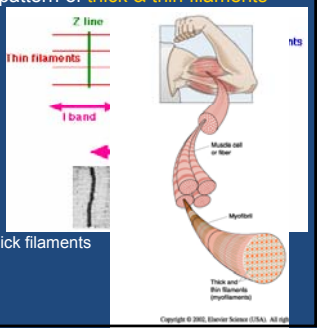




Review of structure

Skeletal muscle cells (fibers) made of myofibrils

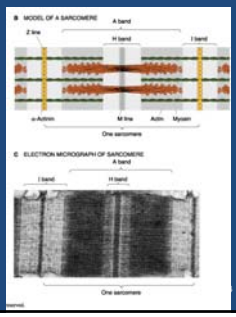
- Myofibrils: Unit of repeating pattern of thick & thin filaments
- Thick filaments
  - Myosin
  - Orientated opposite directions
  - Middle of sarcomere
  - Overlap with thin filaments
- Thin filaments
  - Actin, tropomyosin, troponin
  - 2 sets per sarcomere
  - One end connected to Z line
    - Interconnecting proteins
  - One end overlaps portion of thick filaments
    - A band



Review of structure

Sarcomere: smallest functional unit of muscle

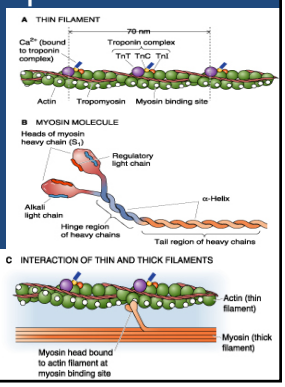
- Area between 2 adjacent Z lines
- Striated pattern
  - dark **A bands**
    - Bisected X **H band**
  - light **I bands**
    - Bisected X **Z line**



Skeletal muscle structure

Contractile proteins

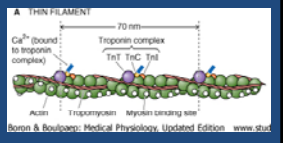
- Actin (2 helical chains)
  - Globular protein
  - Has binding site for myosin
- Myosin (6 subunits)
  - 2 Heavy chains
    - Elongated alpha-helical rod
    - Head binds ATP & actin
      - ATPase activity
  - 4 Light chains
    - Regulatory
      - ATPase activity
      - Motor function



Skeletal muscle structure

Regulatory Proteins

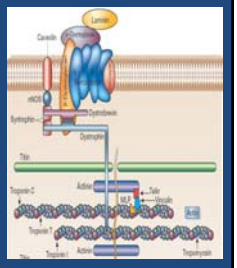
- Tropomyosin
  - 2 alpha helices coiled around
  - Sit in groove of actin chains
  - Cover myosin binding sites
- Troponin
  - Heterotrimer
  - Small globular proteins at intervals on tropomyosin
  - T: binds Tropomyosin
  - I: binds actin & Inhibits actomyosin ATPase
  - C: binds Ca^{++}

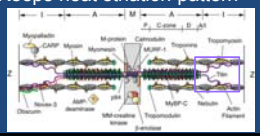


Skeletal muscle structure

Structural proteins

- Dystrophin-glycoprotein complex
  - Links actin to extracellular matrix
  - Structural support & strength to muscle fibril
    - Duchene muscular dystrophy
- Titin
  - Anchors myosin to actin network
  - Keeps neat striation pattern





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Skeletal muscle structure

Sarcotubular System: T tubules & sarcoplasmic reticulum

- In-foldings of plasma membrane
  - Penetrate muscle fiber at A-I junction
  - Lumen continuous with ECF
  - Helps the spread of action potential
- Associated with cisternae
  - Specialized region of SR
    - Ca^{++} stores

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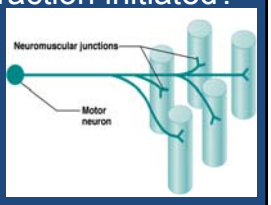
Excitation-contraction coupling skeletal

How is muscle contraction initiated?

- Somatic Nervous System
- Motor neuron
  - Spinal cord or brainstem
  - Myelinated axons
    - AP propagated at high velocity
  - Innervate many muscle fibers
    - Neuromuscular junction
  - Each muscle fiber controlled by branch from 1 motor neuron
- MOTOR UNIT: minimum unit of contraction
  - 1 MN → 2-3 fibers (larynx)
  - 1 MN → 10 fibers (eye)
  - 1 MN → 1000-2000 fibers (gastrocnemius)



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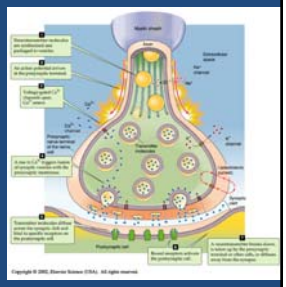
Electrical characteristics

Motor neuron releases Acetylcholine

- AP reaches axon terminal
  - Plasma membrane depolarization
    - Opening of voltage-sensitive Ca^{++} channels
    - Ca^{++} influx to axon terminal
  - ↑ Ach release into cleft
- Ach binds to cholinergic receptor on muscle fiber end plate
  - Nicotinic (N_1) receptor



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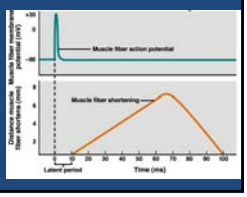
Excitation-contraction Skeletal muscle

Acetylcholine binding triggers skeletal muscle action potential

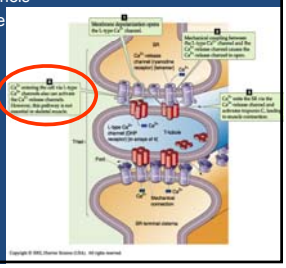
- Ach nicotinic receptor
  - Ligand gated-Na channel
    - ↑ Na+ influx → muscle end plate depolarization
- Depolarize adjacent plasma membrane
  - Open voltage-gated Na+ & K+ channels
  - Spread of AP by T tubules
- Acetylcholinesterase
  - ↓ receptor-bound Ach
  - End-plate Ion channels close
    - Returns to resting potential
  - Ready to respond to Ach again

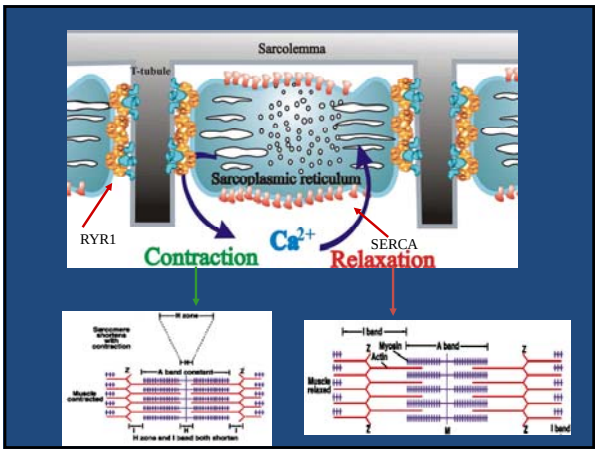


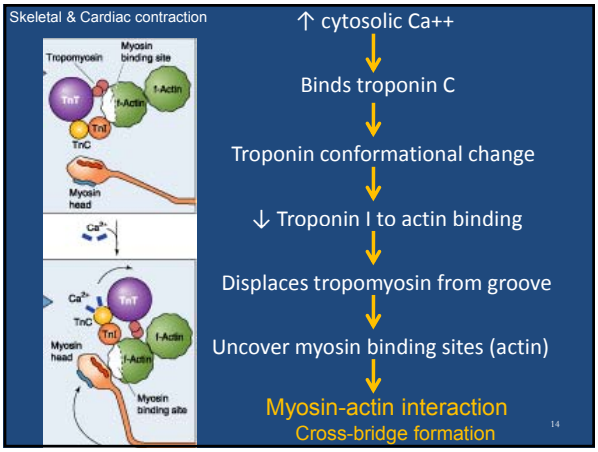
Contractile responses

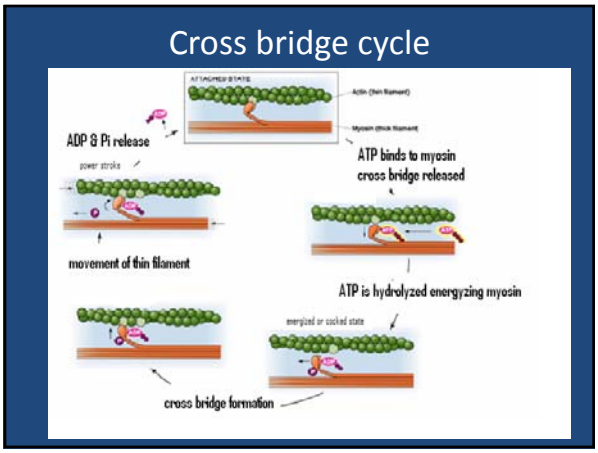
T tubule spreads action potential leading to SR Ca^{++} release

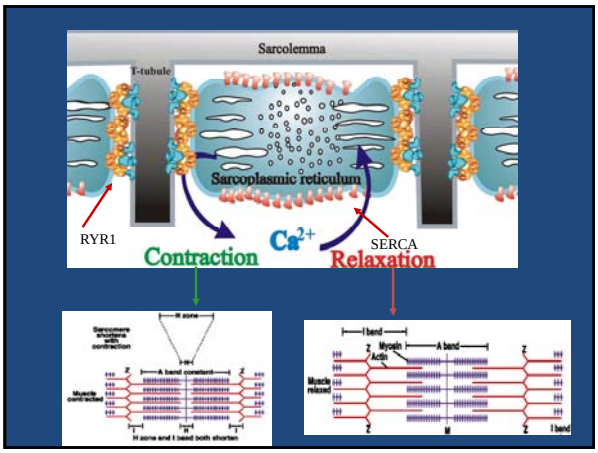
- AP conducted via T tubule
  - Activates voltage sensors: Dihydropyridine receptors
    - L-type voltage-gated Ca^{++} channels
    - Undergo conformational change
  - Opens SR Ca^{++} channel
    - Ryanodine receptor
  - ↑ cytosol [Ca^{++}] 100-fold




Sarcolemma
T-tubule
Sarcoplasmic reticulum
RYR1
Ca^{2+}
SERCA
Contraction
Relaxation


Skeletal & Cardiac contraction
↑ cytosolic Ca++
Binds troponin C
Troponin conformational change
↓ Troponin I to actin binding
Displaces tropomyosin from groove
Uncover myosin binding sites (actin)
Myosin-actin interaction
Cross-bridge formation
Tropomyosin
Myosin binding site
Myosin head
TnT
TnI
TnC
f-Actin

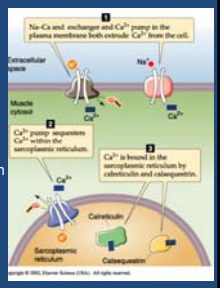

Cross bridge cycle
ADP & Pi release
power stroke
movement of thin filament
ATP binds to myosin
cross bridge released
ATP is hydrolyzed energyzing myosin
cross bridge formation



Contractile responses

Muscle relaxation X ↓ Cytosolic Ca^{++}

- Ca removed
  - Across cell membrane (minor)
    - Na^{+}/Ca^{++} exchanger
  - Reuptake into SR (major)
    - Ca^{++}/Mg^{++} ATPase (active transport)
    - Bound by calsequestrin & calreticulin
      - Ca^{++}- binding proteins in triad junction
- ↓ cytosolic Ca^{++}
  - Ca^{++} removed from troponin
    - Restore tropomyosin blocking action
    - Cover myosin-binding site on actin
    - Actin/myosin interaction stops



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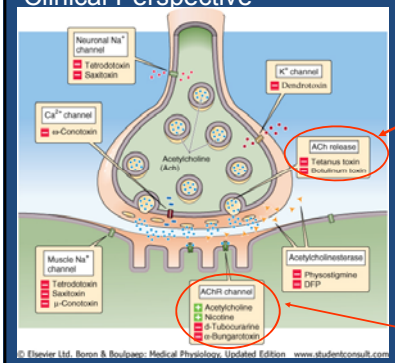
Excitation-contraction coupling: Clinical Perspective



Botox

Curare

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Energy & metabolism

Muscle contraction & relaxation require energy (ATP)

- Cross-bridge cycling
  - Power stroke & myosin dissociation from actin
- Relaxation
  - Removal of cytosolic Ca^{++}
  - Ca^{++}/Mg^{++} ATPase pumps Ca^{++} into SR
- No ATP → Thick & thin filaments bound to each other
  - No relaxation → Rigor mortis

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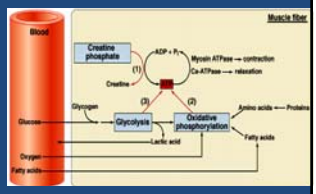
Energy & metabolism

3 sources of ATP

1. ADP phosphorylation
   - Creatine phosphate
     - Creatine PK
     - Immediate & short-lived
2. Glycolysis
   - High rate of ATP supply
   - Low yield/mole glucose
   - Short-lived
3. Oxidative phosphorylation
   - Slowest
   - Most efficient
   - Longer lasting



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Energy metabolism

Biochemical differences in muscle fiber determine preferred energy substrate

- Oxidative (red muscle fibers)
  - High blood vessel & mitochondria content
    - ATP dependent on blood flow; O_2 & fuel
  - ↑ Myoglobin (O_2 binding protein)
- Glycolytic (white muscle fibers)
  - Low blood vessel & mitochondria content
  - ↑ glycolytic enzymes & glycogen stores
  - Larger in size

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Contraction & energy

Skeletal muscle fiber types

- Energy metabolism
  - Major pathway of ATP generation
    - Oxidative or glycolytic
- Rate of force production & shortening velocity
  - Myosin isoform rate of ATPase activity
    - Fast or slow

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Energy metabolism

Myosin ATPase & energetics
3 Types of skeletal muscle fibers

- Type I → Slow-twitch oxidative
  - Low myosin-ATPase activity
  - Dense capillary network & rich myoglobin
  - Rich mitochondrial & oxidative-enzyme content
  - Utilize fats and carbohydrates better because of the increased reliance on oxidative metabolism
  - High oxidative capacity & resistant to fatigue
    - Body posture, skeletal support, endurance activities
- Example: soleus

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Energy metabolism

Myosin ATPase & energetics
3 Types of skeletal muscle fibers

- Type II Fast-twitch
  - High myosin-ATPase activity
    - Short twitch durations & fine skilled movement
      - Greater amounts of force production for shorter periods of time
    - Fast oxidative (IIa)
      - High myoglobin + mitochondria content
    - Fast-twitch glycolytic (IIb)
      - less mitochondria 7 >glycogen
      - fatigue rapidly
  - Example: gastrocnemius and vastus lateralis

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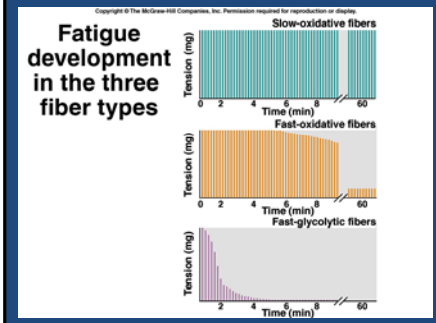
Energy & metabolism

Time to fatigue depending on fiber type



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Energy & metabolism

Fiber type determines resistance to fatigue

- Fatigue
  - ↓ Muscle tension following previous contractile activity
  - Recovery depends on duration & intensity of activity
    - Faster after high intensity low duration (Weight lifting)
    - Slower after low intensity long duration (distance running)
- Slow-oxidative fibers
  - Resistant to fatigue
    - Prolonged & continued (marathon)
- Fast-glycolytic fibers
  - Fatigue rapidly
    - Rapid & powerful (jump, sprint)

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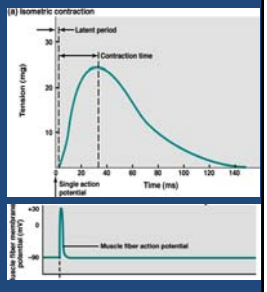
Contractile responses

Relationship of AP to fiber twitch Isometric contraction

- Duration muscle twitch > AP
- Muscle; no refractory period
- Latent period:
  - T from AP to ↑ in tension
- Contraction time:
  - T between initial & peak tension



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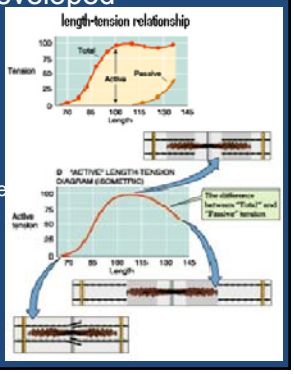
Contractile responses

Relation between muscle length & tension developed

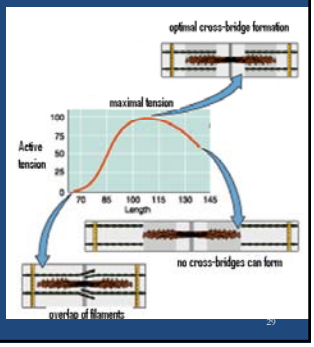
- Tension developed in isometric contraction
  - Varies with length of fiber
  - Is maximal at resting length
- Total tension
  - measured at different muscle lengths
- Passive tension
  - Tension prior to contraction
- Active tension
  - Total - passive tension



Contractile responses

Length - tension relationship

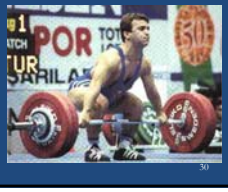
- Maximal tension
  - Max # of cross-bridges
    - thick & thin filament overlap
- Cardiac muscle
  - ↑Tension developed with ↑ fiber length
- Smooth muscle
  - Greater range of lengths over which maximal tension can be developed



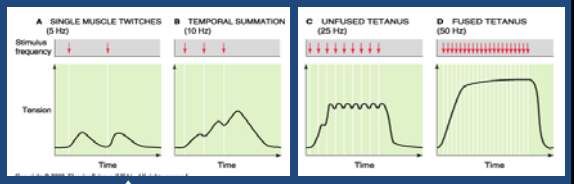
Contractile responses

Load-velocity-length relationship

- Inverse relationship between force & velocity of shortening
  - Slow at heavy load
- Velocity at given load is maximal at resting length
  - Velocity decreases if muscle is shorter or longer



↑ Force of contraction by summation



- Spatial: ↑ # fibers recruited
  - Small to...larger motor units
  - Asynchronous & in tandem
- Temporal: ↑ frequency of stimulation
  - Progressive ↑ frequency → fused
    - No relaxation between stimuli → Tetanus



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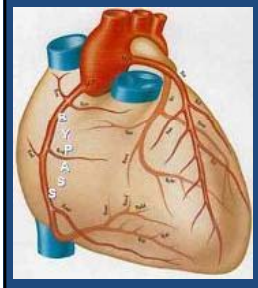
Cardiac muscle



- Structure
- Excitation-contraction coupling
- Energetics & mechanics of contraction

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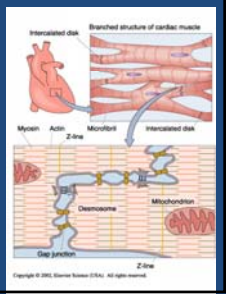
Cardiac muscle structure

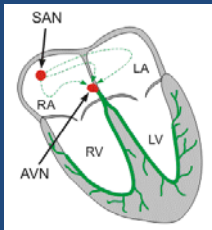
Cardiac muscle vs. skeletal

- Similarity
  - Striations, T tubules, contractile & regulatory proteins, sliding-filament mechanism
- Differences
  - # mitochondria & capillary supply
  - Intercalated disks
    - Between fibers; cell-cell/cohesion
  - Gap junctions (connexons)
    - Connect cytosol of adjacent cells
  - Extra cellular matrix
    - Remodeling and failure
  - Control of contraction



Cardiac muscle has automaticity

- Intrinsic ability to contract spontaneously & rhythmically
- Specialized subset of cardiac muscle cells (**Purkinje cells**)
  - Located in:
    - Sino-atrial (SA) & Atrio-ventricular (AV) nodes
    - Bundle of His, bundle branches, & Purkinje fibers of the ventricles.



SAN, sinoatrial node; AVN, atrio-ventricular node; RA, right atrium; LA, left atrium, RV, right ventricle; LV, left ventricle.

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Cardiac muscle excitation contraction coupling

Cardiac muscle functions as a syncitium

- Pacemaker potentials originate in SA node
  - Depolarization via gap junctions
  - Activation of ventricular muscle
  - ↑ $[Ca^{++}]_i$ → myocardial contraction
- Control of contraction **Autonomic**
  - Neurotransmitters
    - PSNS: Ach: Cholinergic (Muscarinic) receptor
    - SNS: NE: Adrenergic (ß) receptors

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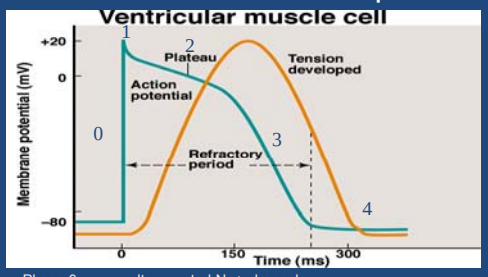
Cardiac muscle excitation contraction coupling

Cardiac muscle action potential



Phase 0: open voltage-gated Na+ channels
Phase 1: close Na+ channels
Phase 2: slow opening of voltage-gated L-type Ca^{2+} channels [DHP]
Phase 3: close Ca^{2+} channels & K^+ efflux
Phase 4: resting membrane potential

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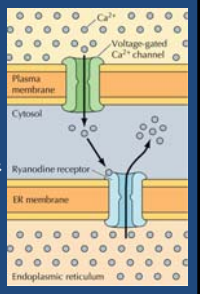
Ca++ influx X L-type Ca++ channels

Required but not sufficient for cardiac muscle contraction

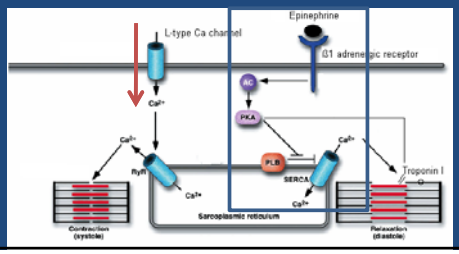
- Ca^{++} binds to SR RyR
  - ↑ Ca^{++} release → ↑ $[Ca^{++}]i$
- Ca^{++} Binds Troponin C
  - – Uncover actin binding sites
  - – Cross-bridge formation, cycling & contraction
- Relaxation x Ca^{++} reuptake
  - – Phospholambdan
    - Inhibits Ca^{++} reuptake into SR
    - ^{+}Pi → removes inhibition



Ca++ regulation of cardiac muscle contraction

2 key points for cardiac muscle contraction!

- Ca^{++} influx X L-type Ca^{++} channels required
- Protein phosphorylation speeds cardiac muscle relaxation



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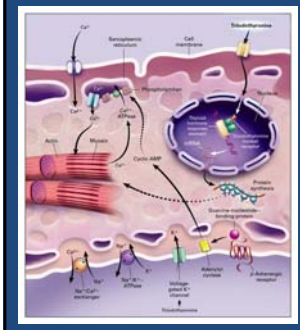
Hormones can modulate cardiac muscle contraction



- Thyroid hormone
  - – Gene transcription
    - Ca^{2+}-ATPase
    - Phospholamban
    - Myosin
    - Adrenergic receptors
    - Adenylyl cyclase
    - Na^{+}/Ca^{2+} exchanger
    - Na^{+}/K^{+}–ATPase
    - Voltage-gated K channels
  - – Nonnuclear actions
    - Na^{+}, K^{+}, Ca^{2+} ion channels

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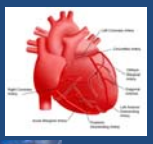
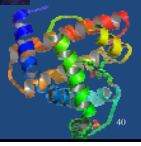
Cardiac muscle energetics





- Abundant blood supply
  - 1 capillary/fiber
- Rich in mitochondria
  - 30-40% muscle mass
- High myoglobin content
  - O_2 stores
- Oxidative & glycolytic
  - Fat 60%
  - CHO 35%
  - Ketones & AA 5%

Cardiac muscle

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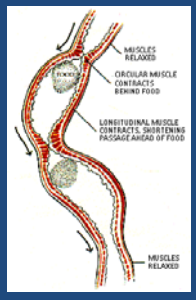
Smooth muscle



- Structure
- Excitation-contraction coupling
- Regulation of contraction & relaxation

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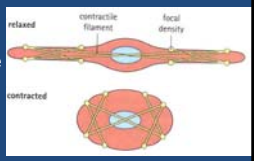
Smooth muscle structure

Smooth vs. skeletal muscle



- Similar contractile proteins
- Different regulatory proteins
  - No troponin
  - Myosin light chain kinase
  - Myosin light chain phosphatase
  - Calponin & caldesmon
    - Inhibit myosin ATPase activity
- Structurally
  - Smaller cells
  - Diagonal filament orientation
    - Anchored to dense bodies ≈ Z lines
  - No T-tubules & undeveloped SR
    - Relies on extra-cellular Ca^{++}
  - Gap junctions

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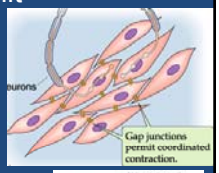
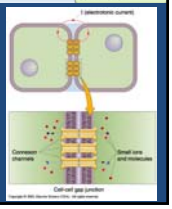
Smooth muscle

Functionally: 2 Types of SM single & multiunit

- Single unit
  - Visceral smooth muscle
    - GI tract, uterus, bladder
  - Poorly innervated
  - Fibers linked by gap junctions
    - Action potential propagated cell-cell
      - Not all cells need to be stimulated
  - Unstable membrane potential
  - Stretch produces contraction

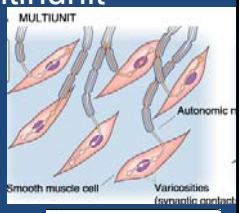


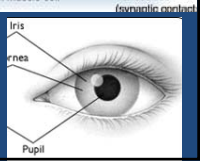


Smooth muscle

Functionally: 2 Types of SM single & multinunit

- Multiunit
  - Richly innervated by ANS
  - Few or no gap junctions
    - Little electrical coupling
    - Fibers respond independently
    - Capable of finer control
      - Large airways, arteries, iris
- Stable membrane potential
- Contractile response depends
  - # of muscle fibers activated
  - Frequency of nerve stimulation

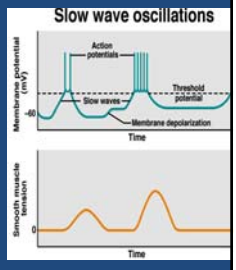




Smooth muscle

Visceral SM electrical activity

- Unstable membrane potential
  - Variable resting potential
  - Slow waves can trigger bursts of action potentials
    - Depolarization-repolarization cycle
    - Rhythmic contractions
- Pacemaker potentials
  - Generated in multiple shifting foci
  - Modulated by ANS



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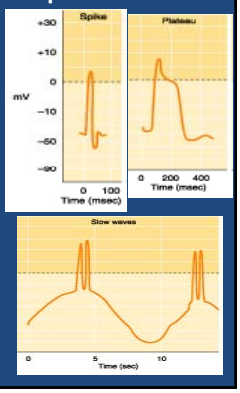
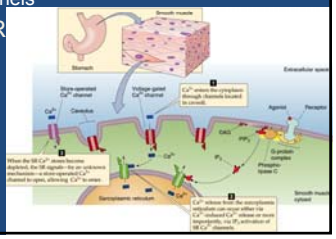
Smooth muscle action potential

- Variable patterns
  - Spike
  - Plateau
    - Delayed repolarization prolonged Ca^{++} entry & contraction
  - Slow waves; oscillations in mV
    - L-type Ca^{++} channels active at resting mV depolarize cell enough to activate more Ca^{++} channels….Ca^{++} influx → activate K^{+} channels….repolarization



Importance of Ca^{++} in SM contraction *Poor SR *

- $[Ca^{++}]_i$ determined by:
  - Ca^{++} entering cells
    - Voltage-gated L-type channels
    - Store operated channels
  - Ca $^{++}$released by SR
    - Ca^{++}, IP_3
  - Removal of Ca^{++}
    - Out of cell
    - Into SR

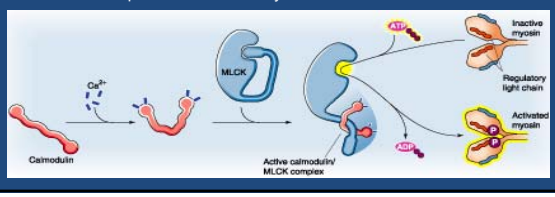


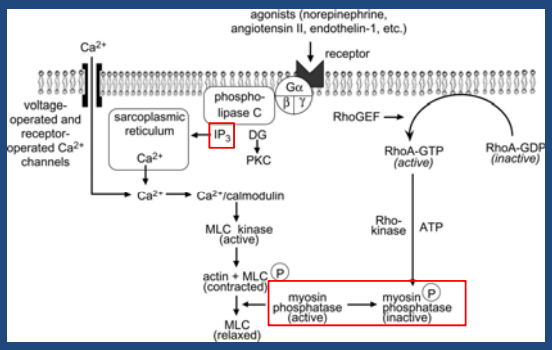
Smooth muscle

Smooth muscle

Smooth muscle: No troponin Myosin: site of Ca^{++} regulation

- Ca binds to Calmodulin → CaCM
  - Activates myosin light chain kinase (MLCK)→ *Pi + MLC
    - ↑ myosin ATPase activity
  - Activates CaCM-dependent kinase → *Pi + calponin
    - ↓ calponin inhibition of myosin ATPase





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Multiple mechanisms regulate SM excitation-contraction coupling

- Electrical depolarization
  - Voltage-gated Ca^{++} channels
    - L-type Ca^{++} channels
- Chemical stimuli
  - Hormones, NT, local factors
    - Receptor-mediated
- Mechanical
  - Passive stretching
    - Myogenic response

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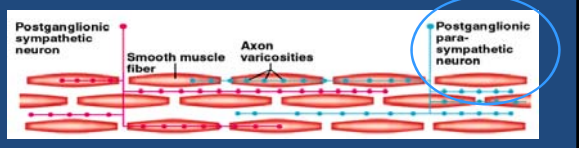
Smooth muscle

Neurotransmitters & hormones



- Do not have specialized end-plate region
  - Varicosities filled with neurotransmitters
  - NT released when AP passes the varicosity
  - Varicosities from one axon may be in more than one fiber
  - Single fiber may be located near SNS & PSNS varicosities

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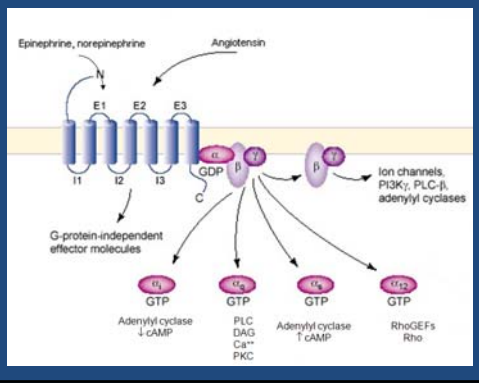
Receptor type determines response



Smooth muscle

Neural stimulation of smooth muscle

- Norepinephrine contracts vascular smooth muscle (α_1 receptors skin)
- Acetylcholine contracts intestinal smooth muscle
  - ↑ Phospholipase C & IP_3 → ↑ intracellular Ca^{++} → Ca•CM →MLCK
- Epinephrine relaxes bronchial, uterine & vascular smooth muscle (skeletal muscle)
  - ↑ cAMP → PKA → inhibits activity of MLCK

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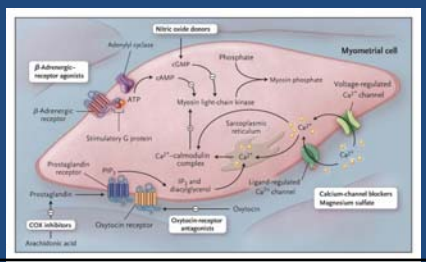
Smooth muscle

Hormones modulate SM function & structure

- High estrogen: SM hypertrophy & ↑ gap junctions
- Oxytocin stimulates uterine contractility



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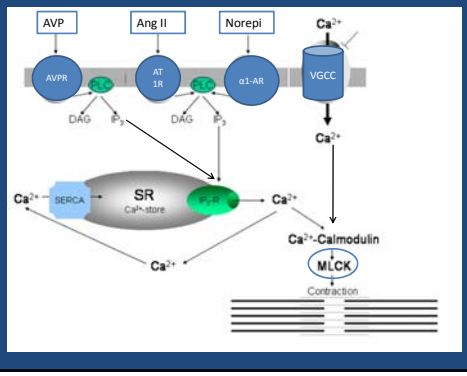
How is blood pressure controlled?



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Key features of smooth muscle

- Slow cross bridge formation and cycling rate
- Maintains tension for prolonged periods
  - Latch or tonic state
  - Minimal ATP needs (oxidative & glycolytic)
- Increased tension developed over greater range of length
  - Contracts in response to stretch
- Does not need action potential to contract

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Review Table

Characteristic	Skeletal	Cardiac	Smooth
Thick & thin filaments	Yes	Yes	Yes
Striated pattern	Yes	Yes	No
T-tubules	Yes	Yes	No
Sarcoplasmic Reticulum	++++	++	+
Gap junctions	no	Yes	Yes
Ca^{++} source	SR	EC & SR	EC & SR
Site of Ca^{++} regulation	Troponin	Troponin	Myosin
Hormone effects	no	yes	yes
Stretch induces contraction	no	no	yes

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