Abnormal Resting Biomarkers in Sickle Cell Trait

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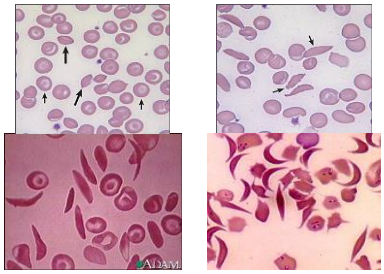
Sickle Cell Disease

- Hemoglobin S
  - History
    - First identified in 1910 in Chicago (Dr. James B. Herrick) on a black Caribbean Dental student
    - Characterized from the 1940s to the 1970s
  - Definition
    - A genetic mutation of one NT in beta globin gene, changing 1 codon, that results in an amino acid substitution yielding a hemoglobin that polymerizes under low oxygen tensions changing the shape of the RBCs causing obstruction, hypoxia, and hemolysis.
  - Inheritance
    - Homozygous (SCD) and Heterozygous (SCT)
    - Compound heterozygotes (HbSC and HbS/β-thal)
  - Etiology
    - The substitution of a valine (+0) for a glutamic acid (-1) at the 6th position of the beta chain (Change in charge of +1)
      - $(\alpha_2\beta_2^{6GLU>>VAL})$
  - Pathophysiology
    - Low O_2 (<85%) > DeoxyHb > Polymerization > sickle cell formation > vaso-occlusion > anemia

Sickle Cell Disease



Health Issues with Sickle Cell Trait

- Mortality
  - 1933: 1st report of SCT exhibiting shortened lifespan
  - 3 early studies between 1940s-1958 suggested shortened lifespan (flawed)
  - 9 subsequent studies between 1966-1975 showed no reduction in lifespan
- Hospitalizations
  - 2 studies in 1952 & 1975 showed no increase in hospitalization for SCT
- Growth & Development
  - 4 studies (1974-1980) showed no different in height, weight, or classroom achievement for SCT children but delays in skeletal development (1975 x 2)
- Multiple Health Issues Reported
  - Splenic infarcts (altitude or anesthesia), pregnancy, newborns, infections, renal dysfunction, hematologic abnormalities, neurological, ophthalmological, pulmonary, bone, leg ulcers
- Exertional Events
  - 1967-1994: 39 total exertional events in the military in soldiers with SCT; most died
  - 1974-1993 10 civilian deaths; 1996-2008: 16 non-athlete civilians had adverse events
    - 15x greater risk of exercise-related death for athletes & 37x greater risk in football

Military and NCAA responses

- Military Regulations
  - 1973 policy pertained to all branches evenly with restricted duties
  - 1981 restrictions were lifted if HbS ≤41%
  - 1985 all restrictions were lifted and no screening was required
  - 1996 Modifications specific to military branch
    - Army ceased to screen for HbS: reevaluated after 8 deaths in 1999/2000
    - Marines screened but maintained identical protocols for SCT
    - Air Force screened and offered option to decline military service (HbS>45% must leave)
    - Navy screened and tagged SCT recruits with a red tag and belt (HbS>45% must leave)
    - NIH recommended the military stop screening for HbS and develop universal standards
- NCAA Policies
  - 1975 NCAA issued comments to alert SCT athletes to risks
  - 2010 All Division I NCAA athletes must provide proof of SCT status or sign waiver
    - 2012 for Division II
  - ASH released a statement recommending the NCAA to reverse the policy
    - Change training regimens universally to reduce all exercise related events

Other side of the debate

- Exercise-related events also happen in non-SCT
- SCT athletes and warfighters have been successful
  - Many athletes and warfighters with SCT are event-free
  - % with SCT running the Abidjan semi-marathon is same as population (Ivory coast)
  - Same for Mt. Cameroon Ascent race (WC Africa)
  - Some US Olympic sprinters had SCT
  - More Ivory Coast Track & Field Champions with SCT than in population
    - Women's high jump (90.9% SCT) and men's shot put (87.5% SCT)
    - 32/33 records for sprint races had SCT
    - SCT have better performance during jump and reach tests
    - SCT have higher type IIx muscle fibers (brief/explosive movements)
  - No direct evidence that death is caused by sickle cell obstruction
    - These deaths do not involve infarction of spleen, kidneys, lung, bone, retina, brain
  - Few reports in SCD of exertional mortality, MI, angina, rhabdomyolysis
    - SCD group rarely exerts, avoids the military, athletics, and altitudes

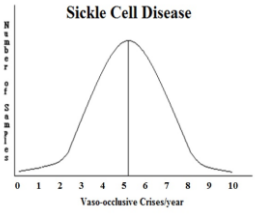
Mechanisms and biomarkers during SCT events

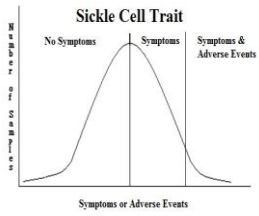
- rhabdomyolysis
  - SCT are 100-200x risk of developing exertional rhabdomyolysis
    - Exertional heat illness > rhabdomyolysis, thrombosis, renal failure, cardiac arrhythmias > adverse event > death
    - Heat illness: ↑ Core temp, ABG (acidosis), glucose (↓), lactate (↑)
    - Rhabdomyolysis: ↑ CK, ALT, AST, LD, urine myoglobin, urinalysis (↑sp. gr. & Blood)
- activation of the coagulation system
  - (2007) SCT have 2x increased risk of VTE, 4x increased risk for PE, 7% have VTEs
  - Increased vWF, D-dimer, TFIII, Fibrinopeptide, Prothrombin F1.2, thrombin/antithrombin complex, decreased ATIII/Protein C/S, Abn PFA-100
    - May explain splenic infarcts, hematuria, isothenuria, & exercise-related adverse events
  - Some SCT patients developed DIC
- Renal disease (renal infarcts or myoglobinemia)
  - electrolytes (anion gap >30 & hyperkalemia), ↑ BUN, creatinine, uric acid

SCT and Exercise

- Can exertional events be initiated & measured in controlled study?
- Controlled studies showed no difference after exercise for SCT & AA
  - Normal ECG, lung/heart function, gas transport, O_2 consumption, metabolic recovery, "getting into shape"
  - Sickle cell numbers ↑ after exercise (1%) and more with higher altitudes
    - 18,000 ft (2-5%), lung scans showed microvascular infarcts in 50% but normal lung function
    - May be connected to splenic infarcts and renal papillary infarcts
  - Unethical to induce events in research subjects
  - Risky to exercise to intensely
  - Difficult to show mortality relationship because deaths occur infrequently
  - Blood biomarkers were not measured

Hypothesis to explain variability





Role of Biomarkers in SCT

- Can we risk stratify athletes & warfighters with SCT for exertional events?
  - Measure biomarkers at baseline
- Can we verify a SCT-related exertional event
  - ID elevated biomarkers during exertional event
- Can we determine safe re-entry into athletics or active duty
  - Rest for an hour, a day, a week, a month, a season
  - Biomarkers normalize?

Pilot Study for Abnormal Biomarkers in SCT at Rest

- 4 AS subjects and 4 AA control subjects
- Collected blood & urine 4 times for each subject several weeks apart
- All subject's genotypes were confirmed
- Measured 2 biomarkers for hypercoagulability
  - D-dimer & Fibrin monomer
- Measured 4 biomarkers for muscle damage
  - CK, Haptoglobin, TP, U/A (blood)
- Measured 3 biomarkers for renal dysfunction
  - U/A (sp.gr., protein), microalbumin
- Study funded by ASCLS E & R Grant

Pilot Study for Abnormal Biomarkers in SCT at Rest

- Results
  - All results were normal in all AA samples (4 subjects; 16 samples)
  - Hypercoagulability in SCT
    - D-dimer was elevated in 3/4 subjects and 9/16 samples (56.25%)
    - Fibrin monomer was elevated in 2/4 subjects and 2/16 samples (12.5%)
  - Muscle damage
    - CK was elevated in one SCT subject (1/4) & one sample (1/16)
    - Mean CK and TP were higher in SCT than in control but not statistically significant
    - Haptoglobin was lower in SCT than in control but not statistically significant
  - Renal Dysfunction
    - No differences in urinalysis and microalbumin results

Pilot Study for Abnormal Biomarkers in SCT at Rest

- Conclusions
  - Hypercoagulability
    - All SCT subjects demonstrated evidence of hypercoagulability
      - 2 SCT subjects demonstrated elevated D-dimer but not fibrin monomer
      - 1 SCT subject had elevated fibrin monomer but not D-dimer
      - 1 subject showed elevation of both D-dimer and fibrin monomer
    - SCT subjects were elevated one day and not the next
      - 1 SCT subject had elevated for D-dimer all four collections
  - Muscle Damage
    - CK and TP were higher in SCT and haptoglobin was lower (not statistically significant)
- Questions
  - Do elevations at rest suggest chronic activation independent of exertion?
  - Can elevations at rest predict risk of exertional events?
  - Will all values elevated at rest increase with exertion?
  - If elevated at rest, will the level increase more dramatically with exercise?

Future Studies

- Study Design
  - Increase the study N (30-50 subjects in each group)
  - Use age, race, and gender matched control group
  - Measure more biomarkers for hypercoagulability, muscle damage, and renal dysfunction
  - Measure them at rest and following treadmill exertion
  - Measure each subject serially, several weeks apart (at least 4 times)

Future Studies

- Questions
  - Will pattern continue with some SCT being positive and some negative at rest?
  - Will exertion elevate these values in all subjects?
    - Will normal increase to slightly elevated?
    - Will slightly elevated increase to moderately elevated?
  - Will elevations post-exercise be more dramatic in those with resting elevations?
  - Can resting elevations predict exertional elevations?

Thank you?

Questions???????