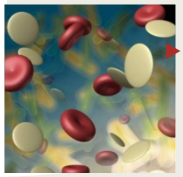
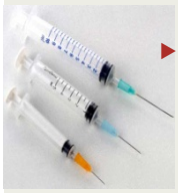
HORMONE PREDICTS RISK OF ACL TEAR. NEW MEDICATIONS ARE BEING DEVELOPED.....COVER



THE DEVELOPMENT OF SECOND GENERATION PLATELET-RICH PLASMA TREATMENT.....3



ARTHRITIS AFTER ACL INJURY- WHY IT HAPPENS AND HOW WE CAN PREVENT IT 4

ISSUE 1 | SUMMER | 2012

Cutting-edge

STANFORD UNIVERSITY TISSUE REGENERATION LABORATORY
DIRECTOR, JASON L. DRAGOO, MD

New strategies in injury prevention and risk assessment may be related to hormonal differences between men and women

A drop of blood to assess ACL tear risk?

Many recent studies have demonstrated that females are anywhere from 2-9 times more likely to tear their anterior cruciate ligament (ACL) compared with their male counterparts. There are a variety of factors that contribute to this gender discrepancy, including biomechanical and biological differences. Relaxin is a hormone produced by females. It was first discovered for its role in child-birth: it dissolves the pubic ligament to facilitate parturition.

Beginning 10 years ago, our laboratory discovered receptors for relaxin on human female ACLs but not in males. We have since found that relaxin increases ligament laxity in animals and that collegiate female athletes who have sustained an ACL tear have higher blood concentrations of relaxin than their uninjured counterparts. In other words, female athletes with high relaxin levels have a greater than 4x increased risk for tearing their ACL.

Consequently, we are developing medications to block relaxin from binding to its receptor on the ACL, and developing a finger-stick blood test to evaluate blood concentrations of relaxin with the hopes of using this information to both prevent and predict future female ACL injury.

Stem cells heal cartilage defects

Articular cartilage is the smooth white tissue that covers the ends of bones where they come together to form joints. Healthy cartilage permits smooth movement of joints by allowing bones to glide easily over one another. However, once damaged, cartilage cannot repair itself, and painful arthritis occurs within the joint. Our laboratory focuses on the use of fat-derived stem cells to stimulate the regrowth of articular cartilage. We have demonstrated the ability of these cells to regrow cartilage in animal models; future endeavors will focus on making this technique feasible in human patients. Our current strategy is to harvest fat tissue from patients and reprogram the stem cells to fill defects in the cartilage surface. *Continued on page 2....*

Stem Cell Therapy

Fat cells transformed into cartilage and bone

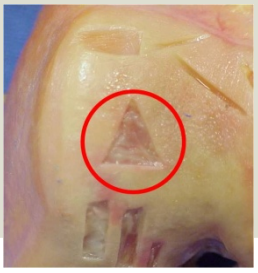
Adult stem cells can be harvested from a variety of human tissues, including bone marrow, neural tissue, muscle, and fat. Cells collected from fat are called adipose-derived stem cells. These cells are of particular interest to orthopaedists because they can differentiate into cartilage, bone, fat, and muscle cells depending on the stimuli delivered to these cells in culture. Furthermore, adipose-derived stem cells are easy to acquire, yield a large number of cells compared with other tissues and do not decline with patient age.

Research in our laboratory has focused on harvesting these cells from the infrapatellar fat pad, which can be conveniently accessed during knee arthroscopy. Through additional work, we have also established that it is possible to heal full-thickness cartilage defects using adipose-derived stem cells in an animal model. After demonstrating the ability to generate cartilage from adipose-derived stem cells and healing cartilage defects using this technology in animals, our next step is to apply this therapy to human patients.

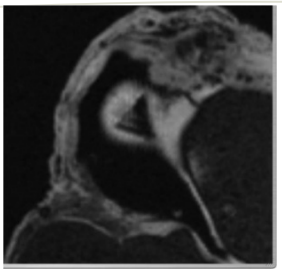
The first step to treating human cartilage defects with stem cells is to identify and assess existing cartilage damage. We have collaborated with musculoskeletal radiologists to develop technology that is highly sensitive to identifying cartilage defects using 3D MRI data. Custom software also developed in our lab is then used to identify the cartilage defect. The software then creates the shape of a custom graft fitted specifically to this defect. The data is then converted to a three-dimensional computer-aided design (CAD) that is processed using rapid-prototyping technologies to create a custom, sterilizable mold. This technology enables us to create unique grafts for each individual patient.

The next step is to fill these custom, patient-specific molds with cartilage cells derived from a patient's own fat cells. By precisely filling the defect and utilizing the patient's own cells, we hope to increase the likelihood of survival and regeneration of healthy articular cartilage.

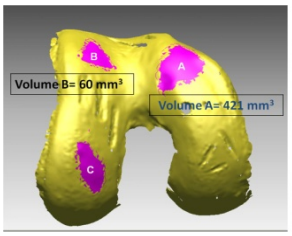
Stay tuned for future clinical trials and updates from our tissue engineering scientists.



Identify defect on surface of articular cartilage.



Use of 3D MRI to visualize cartilage defect.



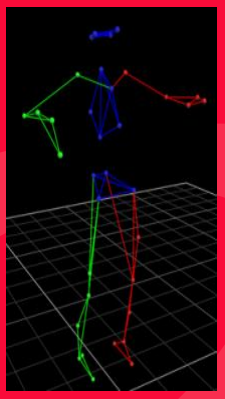
Transform MRI data with custom computer software to create cartilage graft fitting the exact specifications of the original defect.

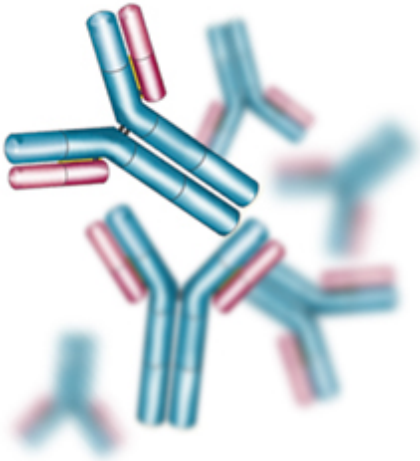
Development of an ACL Injury Prevention Program

Many injuries to the anterior cruciate ligament are due to non-contact mechanisms. These injuries often occur during planting or pivoting movements and can be due to a combination of factors including muscle deficiencies or improper biomechanics. Accordingly, several programs have been proposed to improve athlete strength and flexibility in attempt to minimize these injuries.

Our laboratory is currently collaborating with the Stanford Human Performance Lab to create and implement the Elite Athlete ACL Injury Prevention (EAP) Program. This warm-up and cool-down exercise program designed for athletic teams is aimed at reducing the risk of ACL injury by improving strength, balance, proprioception, and response to unanticipated stimuli.

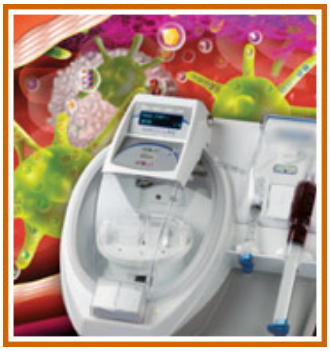
Athletes from five Stanford womens teams are currently enrolled in the program. Using state-of-the-art technology, we direct athletes through a series of tasks and activities to obtain a profile of muscle activation and biomechanical alignment before participants begin the EAP. Following six weeks of participation in the program, athletes return to the lab for post-intervention testing, to evaluate whether their ACL risk factors have decreased.





Perfecting the Next Generation of Platelet-Rich Plasma (PRP)

Developing new technology to selectively remove detrimental growth factors to maximize healing potential of PRP injections



Separate blood components via centrifugation.

Collect specific blood fraction, platelet-rich plasma, for clinical applications. Commercial separation systems employ a variety of centrifugation and separation techniques.

Platelet-rich plasma (PRP) is a fraction of whole blood rich in platelets and growth factors. It is derived from a patient's own blood by acquiring a sample of venous blood, placing it in a centrifuge, and spinning it down for approximately 15 minutes. This separates whole blood into its various components, including red blood cells, platelets, and plasma (the non-cellular fluid in blood). The middle layer constitutes PRP, which contains highly concentrated platelets, the cells that normally promote blood clotting. These cells also contain a number of specialized chemicals called growth factors.

The basic idea behind PRP is to deliver high concentrations of growth factors to areas of injury with the hope of stimulating a healing response and reducing inflammation in the damaged tissue. Injections of PRP are frequently used to treat tendon, ligament, and muscle injuries. However, many recent studies have revealed mixed results regarding the efficacy of these treatments. Some patients experience accelerated healing and report improved outcomes after PRP treatment, while others do not.

One challenge with PRP is that it is composed of many different cellular constituents and it is therefore difficult to pinpoint exactly which components contribute to the improvements in healing seen in certain patient populations. Though largely helpful in the healing and tissue regeneration processes, some growth factors may actually be detrimental. Additionally, the growth factors required for healing may be tissue-specific.

Current research in our laboratory focuses on developing methods to selectively remove certain growth factors from PRP in order to improve the tissue's response to these injections. At present, we use anti-bodies bound to magnetic beads to remove unwanted growth factors and then apply this modified, second-generation PRP to different cell populations and observe the effects of selective administration of growth factors. We are currently collaborating with mechanical engineers at Stanford to develop technology capable of isolating specific growth factors.

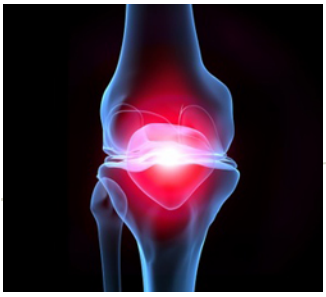
Previous studies in our lab have shown that adipose-derived stem cells treated with 2nd generation PRP produce gene products characteristic of normal chondrocytes. Our next goal is to utilize 2nd generation PRP to help treat osteoarthritis by regenerating damaged articular cartilage. We believe the specific growth factors in 2nd generation PRP combined with the patient's own adipose-derived stem cells will aid in the differentiation and survival of healthy chondrocytes.

Do PRP Injections Have Performance-Enhancing Effects?

Platelet-rich plasma (PRP) was added to the World Anti-Doping Agency Prohibited List in 2010 and removed in 2011 due to concerns that PRP may increase levels of growth factors that could confer an advantage to elite, competitive athletes. Because PRP injections deliver a concentrated dose of a patient's own growth factors to the site injury, it was hypothesized that this local administration might increase the overall amount of performance-enhancing factors like growth hormone, thereby enabling athletes to use these injections to enhance their training efficiency. Our lab is partnering with international anti-doping agencies to assess the concentration of performance-enhancing growth factors in patients who have received PRP injections. We test for levels of growth hormone and five additional banned growth factors by acquiring a small sample of blood from patients at 7 time points after receiving PRP. This will allow anti-doping agencies to test for improper use of PRP by world-class athletes.

OSTEOARTHRITIS AFTER ACL INJURY:

WHY DOES IT HAPPEN AND *HOW* CAN WE PREVENT IT?



Within 20 years of injury, as many as 90% of patients sustaining an anterior cruciate ligament (ACL) injury will develop osteoarthritis (OA), a painful, chronic, and debilitating joint disease. Current orthopaedic practices, including surgical reconstruction, do not prevent the onset of OA.

There are many different opinions regarding the possible mechanisms of OA initiation and progression after ACL injury. One theory focuses on the role of blood in joint disease. When the ACL is injured or torn, a large joint bleed occurs.

Bleeding within the joint likely causes damage that ultimately destroys cartilage and irritates the lining of the joint. The exact components of blood and joint fluid that cause this destruction are unknown.

Our lab is currently implementing a study that evaluates the molecular contents of joint bleeds, joint fluid, and peripheral blood for markers of damage and inflammation. We aim to characterize the precise role of joint bleeding in OA development and improve treatments for OA initiation and progression.

To do this, we recruit patients who report to the Stanford Sports Medicine Clinic within 48 hours of their initial ACL injury. At this time, we remove a substantial amount of joint fluid for therapeutic purposes and retain some of this sample for further analysis. We also obtain a sample of peripheral blood. Then, at the time of ACL reconstruction surgery, we obtain a second round of joint fluid and blood samples while the patient is under anesthesia. We also take several samples of synovium, the tissue lining the joint cavity. We then assay these joint fluid, blood, and tissue samples for markers of oxidative damage. We hope to identify, and in the future block, molecules present in the joint fluid that lead to the onset of osteoarthritis.

A Message from the Director

2011 was an exciting and productive year for our laboratory. Numerous discoveries, such as a new hormone that appears to put females at risk for ACL tears, and establishing the dangers of certain injectable medications that our profession has been using for years, has fundamentally changed the way Orthopaedic Surgery is practiced throughout the world. These advances were largely due to the countless hours of hard work and dedication by our laboratory staff, which includes PhDs, engineers, biomechanics and exercise specialists, Orthopaedic Surgery fellows, residents and medical students.

All of this research would truly not be possible without your continuing support. We cherish our donors, who really keep the laboratory "full-speed ahead" during a time when government funding for science and ortho-paedics is at an all time low. For those of you who wish to contribute, we thank you as well, and will welcome you to our research team for years to come.

Sincerely,

Jason L. Dragoo, MD

Dr. Jason L. Dragoo, MD
Assistant Professor of Orthopaedic Surgery and Sports Medicine
Head Team Physician, Stanford Football Program
Lab Director

STANFORD SPORTS MEDICINE

450 Broadway, Pavilion A
Redwood City, CA 94062

Appointments: 1.800.717.0012

For more information, or to find ways to contribute, please contact:

Jeremy Benjamin
Office of Medical Development
Stanford University
email: Jeremy.benjamin@stanford.edu
phone: 650.234.0622