Current Trends in Cartilage Science: An Impression from the ICRS World Conference 2012

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From May 12 to 15, 2012, the 10th world congress of the International Cartilage Repair Society (ICRS) was held in Montreal, Canada. This year, there were 850 participants with backgrounds in both basic scientific as well as clinical research directed at cartilage repair and regeneration. It encompassed more than 135 oral and 270 poster contributions; 7 plenary sessions and 20 special sessions were organized (three in parallel), each of which included two to three invited lectures by international leaders in the field, covering recent developments and opinions both in clinical as well as more basic scientific research on cartilage repair.

This year's program also included two honorary lectures. Prof. Joseph Buckwalter, who was also awarded the Genzyme lifetime achievement award, gave the first lecture elucidating underlying causes of posttraumatic osteoarthritis (OA) including measurement of articular surface impact energy (which correlates to the occurrence of posttraumatic OA), incongruity of the joint,[1] and instability of the joint. Research includes restoring congruity using computed tomography scans and developing a model to see how pieces fit together and evaluation of how reactive oxygen species (ROS), produced by the mitochondria and expressed after impaction, cause damage in the first 48 hours (and matrix components over a longer period of time) and could be addressed with a number of promising therapies (biologicals, distraction, etc.). Prof. Daniel Grande gave the second honorary lecture, in honor of his seminal work in the field of cartilage repair. He spoke about the history and genealogy of the autologous chondrocyte implantation technology, as well as the evolution of histology grading scales and cartilage imaging. He concluded that the field is going toward *in situ* tissue engineering, and he acknowledged the impact of the ICRS on improved standardized protocols for research and clinical application. Both Prof. Buckwalter and Prof. Grande have provided review papers based on their talks that are published separately in this issue.

Animal Models

Multiple presentations at the meeting echoed the awareness of the limitations of the different models used, in particular the small animal models. Moreover, there is further appreciation of the equine model as one of the leading preclinical models, best mimicking the human situation.[2] Mark Hurtig provided a review of basic science mechanisms in animal models of articular cartilage injury in the ICRS-FIFA Plenary Session on Sport Injury. As pointed out in the ICRS consensus report on animal models,[3] delayed repair of chronic chondral defects is a logical target, but Institutional Animal Care and Use Committee realities force treatment of an acute chondral injury in instances where the treatment involves surgery. It was pointed out that cartilage resurfacing might need to be combined with therapies that address the dysregulation of matrix metabolism because many of our patients have long-standing synovitis, cartilage thinning, and other evidence of a catabolic synovial environment. Significant regulatory barriers exist for combination therapies that might include some combination of drugs, biologics, scaffolds, or cells, so a strong case for efficacy and safety needs to be constructed. Naturally occurring models such as canine hip dysplasia could provide added information on controversial clinical practices such as the diagnosis and management of femoroacetabular impingement.

In a special session on Animal Models and Cartilage Tissue Regeneration, Prof. Hurtig also presented the ICRS consensus on animal models, summarizing agreement about the relative strengths and weaknesses of the model systems.[3] His hope was that logical recommendations might stop the wastefulness that have been pervasive in the cartilage R&D culture. A good example of this is the still-widespread use of immature cartilage rabbits for cartilage repair experiments, which due to their brisk intrinsic repair response have little predictive value for patients. A fully mature "cartilage organ" should contain zonal distribution

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of chondrocytes, a continuous tidemark with a calcified cartilage layer, and a regional specialization of biochemical and biomechanical properties. Also because of the concern about durability and maturation of the repair tissue or neocartilage, long-term studies of a year or more in two species are recommended. The cost of such studies is substantial and time consuming, but statistical power is critical. The consensus document reviewed the evidence-based medicine around use of the larger species, because the mini-pig, sheep, goat, and horse have all contributed to new product registration. The thick cartilage for the horse,[4] suitability for arthroscopic procedures including interim biopsies, growing availability of biomarkers and reagents, and similarity of the cartilage volume of the human knee have made this an attractive model for those who have specialized facilities. Despite the relatively thin cartilage of the sheep and goat, which makes fixation of scaffolds and retention of implanted cells or tissue more difficult, experience and expertise can overcome these obstacles. It was pointed out that none of the larger species can simulate the extremely thick subchondral bone plate and low trabecular volume of the metaphysis in the human knee.

Prof. Caroline Hoemann reviewed rabbit cartilage repair models and pointed out both the promising and limiting features as an orthopaedic model for cartilage repair. The conclusion was that rabbit models are useful for proof of concept data and as a stepping-stone to large animal pivotal studies. The overlying recurrent outcome of rabbit studies is the heterogeneous repair response, and there is still no clear explanation for animal-to-animal variation. The high rate of spontaneous repair in adolescent animals is not reproduced in mature or geriatric rabbits. Therefore, skeletally mature animals (i.e., greater than or equal to 7 months old) should be used to screen the efficacy of formulations as indication for uses in adult human patients.

The last paper in this special session was given by Dr. David Frisbie as a review of equine models. Most of the equine cartilage resurfacing models have focused on the equine knee, or "stifle" as it is commonly called. Comparative studies have demonstrated that the horse has articular cartilage of similar thickness to that in the human knee and closer than other species commonly used in preclinical trials.[2,4] Furthermore, this joint in the horse is commonly clinically affected with cartilage lesions, allowing equine clinicians experience with not only research but clinical outcomes as well. A minimum 9-mm diameter defect has been determined to be a critical size, and many studies have used a 15-mm defect. The ability to differentially determine the calcified cartilage and subchondral bone plate have allowed further refinement of defect creation both through open and arthroscopic procedures, and the size of the equine joint allows multiple defects to be placed in the same joint. Disadvantage of the horse include the inability to provide compression bandaging of the stifle area (as is done in human patients) and the lack of a non-weight-bearing period postoperatively (this issue is somewhat overcome based on defect location). The number of horses can be kept to a minimum in many cases by using the horse as its own control, as well as evaluating the outcomes at various time points (second-look arthroscopies and biopsies). Outcome assessments include clinical examination for lameness and synovial effusion, as well as response to flexion; pretreatment and posttreatment radiographs; magnetic resonance imaging; synovial fluid and serum biomarkers; routine synovial fluid analysis; sequential arthroscopies; optical coherence tomography; gross postmortem examination; histopathological, histochemical, and immunological analysis; biochemical analysis for collagen type II/collagen type 1 ratio, as well as aggrecan and glycosaminoglycan content; and real-time quantitative polymerase chain reaction evaluation for mRNA expression of the tissue and biomechanical evaluation. Several studies in which biopsies were taken of repair tissue at 4 to 6 months have indicated no detrimental long-term implications for the repair tissue from biopsies. The paper also presented belief that there had been a positive evolution of model selection from it based on cost and convenience to more critically evaluating how well an animal model simulates the human situation.

Biomarkers

Prof. Stephan Lohmander addressed the question of how can biomarkers be used as outcome measures in cartilage repair and OA? He discussed how we should consider patient-reported outcomes on symptoms, function, and quality of life as the gold standard, the clinical endpoint. Other outcomes may include functional tests, imaging techniques to monitor structure and quality of joint tissues, and molecular biomarkers to reflect the turnover, structure, and state of joint tissues. Studies on cartilage repair and OA with currently available outcome measures require long observation times and trials and, therefore, a great need for new measures that can predict the long-term clinical outcome after a shorter observation time. The presenter predicted that biomarkers developed for OA will likely find use also in studies of cartilage repair and regeneration. For OA biomarkers, a terminology named BIPEDS was proposed, which classifies these biomarkers into five categories corresponding to their proposed use: burden of disease, investigational, prognostic, efficacy of intervention, diagnostic, and safety.[5] Biomarkers that are likely to have the earliest beneficial impact on clinical trials fall into two categories: (a) markers that would allow us to select for trial subjects that are most likely to respond or progress (prognostic markers) within a reasonable time for a clinical study (1-2 years for an OA study) and (b) those that provide early feedback for preclinical decision making and for trial

organizers that an intervention has the desired effect on the primary molecular target (efficacy markers). Both types of biomarkers are highly desirable in chronic conditions where conventional clinical outcomes may take years to present. Validation of biomarkers against a gold standard endpoint depends critically on the performance and specificity of that gold standard endpoint. A second useful classification system divides biomarkers into four categories according to their current level of qualification: (a) exploration-level biomarkers—used as research and development tools with *in vitro* and/or preclinical evidence but without consistent information linking the biomarker to clinical outcomes in humans; (b) demonstration-level biomarkers—associated with clinical outcomes in humans but have not been reproducibly demonstrated in clinical studies; (c) characterization-level biomarkers—which are reproducibly linked to clinical outcomes in more than one prospective clinical study in humans; and (d) surrogacy-level biomarkers—which can substitute for a clinical endpoint, corresponding to "surrogate endpoint" as mentioned above and require agreement with regulatory authorities as an FDA-registered endpoint.

Prof. Robin Poole addressed emerging molecular biomarker technologies and the way forward. His presentation focused on turnover of type II collagen, the dominant component of the extracellular matrix of cartilage without which cartilage could not exist. Important requirements for the development of successful skeletal biomarker technology are the ability to accurately identify the source of the biomarkers, the molecular event(s) that generates it, and what the biomarker assay measures in terms of the molecular fragment and where best to measure this biomarker in synovial fluid, serum, or urine because different results can be obtained with each of the body fluids. The presentation looked at the development of technology to detect the synthesis and degradation of type II collagen cartilage. Tests for cartilage degeneration include the C2,C and C,2C competitive ELISA inhibition assay to detect cleavage of type II collagen by collagenases and a competitive ELISA immunoassay (CPII) to detect collagen II syntheses.[6] These assays used in combination can detect differences between individuals with early, pre-, and radiographic knee OA and those without knee OA,[7] reveal differences between those with knee OA who exhibit progression and those who do not,[8] and indicate early responses to disease-modifying therapy in patients with rheumatic arthritis.[9]

Although not strictly related to biomarkers, the third excellent paper by Prof. Linda Sandell was on genetic influence on cartilage repair and deserves mention. She investigated cartilage regeneration in genetic murine models using common and bred strains in a set of recombinant inbred lines generated from LG/J (healer) and SM/J (non-healer) inbred strains to investigate cartilage regeneration in acute full-thickness cartilage injury once created in the trochlear groove of 265 mice by the method of Fitzgerald and colleagues.[10] The result showed that both cartilage regeneration and ear wound closure are significantly heritable traits. They concluded that articular cartilage regeneration is heritable, the phenotypic differences between the lines are because of genetic differences, and a strong genetic correlation between the two phenotypes (cartilage regeneration and ear wound healing) exists, indicating that they plausibly share a common genetic basis.[11]

Nerve Dependence on Cartilage Development, Repair, and Joint Pain

Prof. Malcolm Maden introduced a novel topic. His lecture focused on Urodele limb regeneration and how this is relevant to mammalian cartilage regeneration. He investigated the role of nerves in the newly developing Urodele limb. Following amputation and wound healing, the internal tissues—muscle, cartilage/bone, dermis—dedifferentiate and form the blastema, which grows and redifferentiates into the missing structures. The regeneration of these amputated limbs is highly dependent on the nerve fibers remaining at the amputation plane, which is regulated through anterior gradient protein secreted by the Schwann cells of the distal nerve sheath and the gland cells in the wound epidermis covering the amputated limb. Current research in his group now focuses on the regeneration of large punch holes in rabbits and mice[12] that occurs by a process strikingly similar to Urodele limb regeneration.

Dr. David Walsh then addressed neurogenic factors and the eitopathogenesis of OA. He outlined that OA is more than just a disease of the articular cartilage alone, and peripheral sensitization of nerves within the joint contributes to OA. Consequently, the experience of OA will depend on how the signals are processed through the spinal cord in the brain. In the osteoarthritic joint, sensory nerves invade through vascular channels that extend from the subchondral bone into the articular cartilage. This leads to the general activation of the sensory nerve system in the joint. In addition, both blood vessel formation in osteophytes and the meniscus also give rise to further nerve in growth and contribute to pain in OA, even if subjected only to normal mechanical stresses. Nerve growth and angiogenic factors (which overlap in their functions) are each up-regulated in OA, and recent preclinical studies and clinical trials have demonstrated the potential that blocking nerve growth factor or angiogenesis may reduce OA pain.[13]

Prof. Mats Brittberg concluded the session addressing the question of which cartilage lesions are painful and what the cause is of the pain experiences by some patients. Today, it is unclear which defects are causing pain for patients and where that pain specifically originates from. However, the pain sensation may use the same channels as in OA-related pain. Prof. Brittberg described that pain could be a result of the elevated stress, resulting in edema, in the

subchondral bone, although this is difficult to measure. Moreover, like the pain in OA, it could also result from the formation of osteophytes and/or subchondral microfractures. Pain could also originate from the surrounding through the disturbance of joint homeostasis, that is, a focal cartilage lesion can result in the secretion of neuropeptides (e.g., calcitonin-gene-related peptide and substance P) in the subchondral region that may directly interact with the receptors of the chondrocytes.[14] He concluded that in view of improving clinical therapy, a better understanding of the role of the nerves in the subchondral bone and the intra-articular structures is of importance.

Bioprinting and Cartilage Regeneration

The opportunities for bioprinting in the regeneration of cartilage were the basis of a special session on this emerging topic. Prof. Dietmar Hutmacher introduced the session. He presented the basic concepts and potential application of additive tissue manufacturing that allows the generation of living multifaceted structures. Importantly, bioprinting allows the incorporation of patient-specific anatomy to create custom-designed implants potentially in the operation theatre, resulting in the reduction of costs. Prof. Hutmacher provided clinical examples of how with printing technology custom-made scaffolds can be generated that fit exactly in the defect,[15] although directing the embedded cells to generate specific functional tissue still needs further research. Even though we are still far away from implants that can be used clinically with respect to cartilage, building machines have been designed that provide spatial control of placing hydrogels and cells[16] to better reflect the layered architecture of the native cartilage.[17] He concluded that the next challenge in translating this approach to the clinic will be the inclusion of multiple cells, materials, and manufacturing processes in a sterile and controlled environment.

Dr. Jos Malda then outlined the use of natural and synthetic biomaterials for bioprinting and introduced the concept of osteochondral bioprinting based on the simultaneous printing of various materials and cells. In a layer-by-layer fashion, constructs could be created based on cell-laden hydrogels and thermoplastic polymer fibers to provide additional mechanical properties.[16] Obviously, these two classes of biomaterials have to meet specific and often complimentary requirements: whereas the hydrogels must support cellular survival and differentiation and degrade relatively fast, the thermoplastic polymer should degrade much slower and provide the construct with sufficient strength. In addition, Dr. Malda pointed out the importance of selecting the appropriate biomaterial for the cell-laden phase. He presented his work on the modification of hydrogel systems to improve the physical requirements for the printing, as well as to enhance the cellular differentiation after printing. Although natural materials such as collagen and alginate support cellular behavior, printing with good shape fidelity and high resolution is troublesome, if not impossible. Synthetic materials are, on the other hand, very suitable for bioprinting applications but do not support sufficient chondrogenesis. Hence, the biofunctionalization of synthetic platforms, or rheological adaptation of natural systems by, for example, addition of viscosity enhancers, such as gellan gum, is currently explored.

Dr. Lawrence Bonassar addressed the use of bioprinting for cartilage and osteochondral repair and reviewed the current state-of-the-art of orthopaedic tissue printing. He explained that, at this stage, bioprinting of bone has been explored to a further extent than of cartilage. For the application of bioprinting for the restoration of osteochondral defects, he identified three critical challenges. The first challenge is the generation of implants with a complex shape, that is, a construct that takes in account the curvature or noncircular perimeters of the defect site. He illustrated this with his work on the printing of complex-shaped ear and meniscus cartilage. Using an incorporated laser scanner,[18] his group was able to demonstrate that the printed structures are close to the native tissue with a resolution of 200 to 300 μm. The second challenge Dr. Bonassar identified is the generation of a multitissue implant. A number of groups have achieved printing of multiple domains, using, for example, labeled cell populations, again with a resolution of about 200 μm. Using multiple nozzle strips, heterogeneous tissues, such as vessels-like structures, can be made can be made. The third challenge is the delivery of the implant to the defect site and the potential shift toward *in situ* printing. This was illustrated by recent work demonstrating the possibility of directly filling calverial[19] or osteochondral[20] defects by means of *in situ* bioprinting. In line with the previous two speakers, Dr. Bonassar concluded that, although big steps have been taken, bioprinting is still in its infancy but with significant potential for the field of cartilage repair. Moreover, he stressed that conversation regarding the technological developments is happening largely outside the field of orthopaedics and that to stimulate the advancement it is of importance that the ICRS continues to embrace this topic in the future.

Cell-Free Approaches

The shift toward *in situ* engineering of tissues was addressed in this plenary session by Prof. Jeremy Mao and Dr. Laurie Goodrich. Prof. Mao discussed biological joint replacement. Prof. Mao discussed homing of endogenous stem/progenitor cells in cartilage regeneration. He started his discussion by pointing out the paradigm of biomaterials, cells, and molecules still being the way and there being a need to progress. He presented his work published in the *Lancet*[21]

where the anatomic model of the articular surface of the rabbit glenohumeral joint was generated by three-dimensional printing from 80% poly-ε-caprolactone and 20% hydroxyapatite composite. Although the focus of the planned experiments was on seeding the scaffolds with MSCs to induce bone formation, a cell-free group, which contained a TGF-β-loaded collagen gel was included as well. Against expectations, cartilage regenerated over the surface of the scaffold of the growth factor loaded constructs, resulting in a hyaline-like tissue with appropriate mechanical properties in contrast to their earlier studies in the mandibular joint.[22] Prof. Mao showed that cell homing plays an important role in this response. Using stem cells derived from different tissues (adipose, bone marrow, and synovium), it was demonstrated that by supplementing with factors such as SDF-1 and TGF beta 3, conditions can be created that conditions homed cells and stimulated chondrogenesis.[23] To further elucidate the combined effect of multiple factors, his group is now applying a high-throughput approach using multi-well microfluidics devices.

Dr. Goodrich presented current prospects for gene therapy as a noncellular therapy. The presentation focused on gene therapy as a noncellular therapy and therefore direct *in vivo* injection of gene therapy vectors. Therapeutic complimentary DNA (cDNA) is placed into a vector backbone and the gene therapeutic vector is then injected into the joint. The research presented has focused on adeno-associated viral vectors (AAV) that appear to have overcome the problems of inefficient transduction. The group is focused on the development of AAV vectors to transmit interleukin-1 (IL-1) into equine joints.[24] Previously effective inhibition of OA[25] as well as promotion of cartilage repair[26] with adenoviral vector–mediated IL-1ra therapy have been demonstrated.

Platelet-Rich Plasma in Joint Tissue Repair

This topic was the subject of a special session. The first paper was Dr. Lisa Fortier discussing, "Platelet-Rich Plasma: Overview of Current Knowledge: Hope, Hype and Reality." Platelet concentrates such as platelet-rich plasma (PRP) have gained popularity in sports medicine and orthopaedics to promote accelerated physiological healing and return to function. The concept that PRP can improve joint or tendon disease is based on the physiologic role of platelets and their contained growth factors in wound healing. However, PRP is composed of all substances in blood and components and this mixture has bioactive functions that positively and negatively affect musculoskeletal tissue regeneration and healing. Mixed reports of success have been reported after the use of PRP in sports medicine, but with the field in its infancy, there is sufficiently positive outcome data available to continue use and investigation into PRP. Dr. Fortier reviewed the basic science and clinical indications. Originally, PRP was considered as a method to deliver platelets and therefore growth factors that led to the common thought that more platelets is better, leading to a race among manufacturers to develop systems that would increase platelet concentration to a greater level compared with their competitors. However, concerns were raised about the increase in leukocytes in some preparations leading to the concept that PRP is a mixture of all blood components and not simply a means of growth factor delivery. *Ex vivo* studies indicated that concentrations of leukocytes in PRP were directly correlated to loss of normal tendon function and an increase in inflammatory molecules. In comparisons of high platelet count versus low platelet count products, there was a much higher white cell count in the high platelet count product and an associated increase in metalloproteinases. Clinical hype over PRP in North America began in early 2009 when two famous athletes received PRP injections and successfully returned to professional athletics earlier than anticipated. A media blitz began but there are a few level 1 studies and several level 2 or 3 studies that have mixed results regarding the efficacy of PRP for treatment of musculoskeletal ailments including joint pain, patella tendonitis, Achilles tendinosis, and epicondylitis. Clinical observation and opinions suggest that pain relief and restoration of function occur more rapidly than expected for some orthopaedic problems with the use of PRP, and this has led to investigations of antinociceptive and anti-inflammatory properties of PRP in the author's laboratory and others. The data indicate that in patients with OA, PRP decreases the production of pro-inflammatory markers of pain such as tumor necrosis factor, which supports the concept that PRP functions to decrease pain and inflammation. A by-product of decreasing inflammation would be joint preservation, but there are no clinical data indicating that PRP increases production of cartilage extracellular matrix proteins such as aggrecan or type II collagen.

The second paper was from Prof. Elizaveta Kon, who discussed the biological rationale of PRP and its clinical application as a conservative treatment and as a "biological augmentation" during surgical procedures. Good clinical results have been reported in a case report using PRP in conjunction with repair of cartilage avulsion.[27] A further study proving the efficacy of polyglycolic/acid hyaluronan scaffold immersed in PRP for treating full-thickness chondral defects of the knee was discussed.[28] A pilot study in the United States reporting benefit in patients with primary and secondary knee OA[29] and a prospective study by the presenter herself published in 2009[30] where 91 patients (115 knees) treated with three injections of PRP. Patients underwent clinical evaluation at 2, 6, and 12 months of follow-up, and 80% expressed satisfaction for the treatment received. Clinical outcome registered a statistically relevant

improvement of all variables just after 2 months from the end of the treatment at 2 months and 6 months with a tendency of worsening from 6 to 12 months of follow-up. Despite the decrease reported after 1 year, the clinical scores at that time were still higher than the basal level. A later study by the same authors evaluated the patients at 24 months of follow-up, confirming this trend with a further decrease in clinical outcome, thus concluding that intra-articular therapy with platelet-derived growth factors is time dependent with an average age of 9 months and better and long-lasting results in younger patients with lower level of joint degeneration.[31]

Dr. Scott Rodeo then reported on clinical experiences with PRP. He reviewed recent clinical data on the use of PRP for tissues in and around the joint including hyaline cartilage, ligament, tendon, and meniscus. The rationale and attraction of PRP is the ability to deliver numerous cytokines in physiologic-relevant proportions. Dr. Rodeo noted that despite vast basic science and laboratory data demonstrating a positive effect of various PRP formulations on basic cell biology, this has not yet translated into a consistently positive clinical effect. One of the limitations in studying PRP is the fact that there is tremendous variability in various commercially available PRP preparations with regard to platelet content, white cell content, platelet activation, kinetics of cytokine release from PRP/PRFM, ratio between fibrinogen and thrombin concentration, formation of a fibrin matrix, and microstructure of the final fibrin network. It has been noted that there is also variability between individuals/patients with regard to platelet counts, day-to-day variation in platelet count, growth factor content per platelet, and other protein/factors in the plasma. Clinical effects typically wear off after 6 to 12 months, and there is very little (virtually zero) data that have demonstrated a positive structural effect (actual regeneration of cartilage tissue). Kon *et al.* have reported PRP superior to HA.[32] Platelet-rich plasma may inhibit the adverse effects of IL-1β and other negative factors in the inflammatory environment, but much further research is needed. There are little data available on the effect of PRP for patellar tendinopathy, but there are some positive data for lateral epicondylitis, suggesting that PRP may be effective for extra-articular tendons, and one study reported positive results on magnetic resonance imaging appearance of degenerative patella tendon.

Clinical Studies Using Cartilage Fragments

Drs. Jack Farr and David Caborn presented a plenary session discussing the use of autologous and allogenic cartilage fragments. Dr. Farr presented the two new approaches that use minced/particulated cartilage to treat chondral defects. One technique uses autograft cartilage (cartilage autograft implantation system [CAIS]) (DePuy Mitek; Raynham, MA) and the other uses juvenile allograft cartilage (DeNovo NT; Zimmer, Warsaw, IN).[33] With the CAIS technique, preclinical data[34,35] were compelling enough for the FDA to approve a safety pilot study. The clinical outcomes are now published at 2 years and an extension follow-up study is complete to 4 years postoperation and an extension is just being initiated. A parallel pilot study has been completed in Europe. This was presented in one of the free paper sessions on cartilage/cell transplantation by Prof. Brittberg. In the United States, the FDA has approved a pivotal study of the technique and the plan is to enroll more than 300 patients for a randomized, perspective comparison of CAIS to microfracture. The case study is restricted to ICRS grade 3a-4a chondral lesions of the femoral condyles or trochlea that, after debridement, measure from 1 cm^2 to 10 cm^2. Dr. Caborn presented the use of allogenic cartilage fragments (DeNovo NT) with clinical case data. Though an extended abstract is not available, good clinical results were reported. After the paper, the Co-Chairs Profs. Anthony Hollander and Alan Gross led an active discussion of the current limitation of clinical data and outcome information in this emerging area of cartilage repair.

Stem Cells for Cartilage Repair

Another plenary session on stem cells for cartilage repair moderated by Prof. Anthony Hollander had two speakers, Prof. Frank Barry, addressing stem cell therapy for joint repair, and Prof. C. De Bari, discussing stem cell-based therapeutic approaches to joint surface repair. Prof. Barry discussed both adult mesenchymal stem cells (MSCs) isolated from bone marrow as well as the use of allogeneic stem cells. He noted that there are many aspects of the biology of MSCs that are poorly described, and a more exhaustive characterization is necessary to exploit these cells fully in the context of tissue repair. Adequate translation of MSC therapy will only be successful if the following are addressed: (a) development of new cell-specific markers, (b) deciphering the therapeutic mechanism of action and unraveling the paracrine signals that contribute to tissue repair, (c) understanding clonal heterogeneity in cultured populations, (d) ensuring that batch variability is controlled, and (e) understanding the nature of host immunomodulation by transplanted MSCs and allongeneicity. There is evidence that human MSCs isolated by current methods are not homogeneous and in fact consist of mixtures of progenitors and other cells. The possibility of culture-induced heterogeneity and the lack of highly specific markers raise issues of regulatory compliance that may need clinical testing. Presently, there are several antibodies that are routinely used to characterize MSCs from human bone marrow by flow cytometry and other methods,

and some have been adopted as release tests for clinical grade cells. These include CD105, CD73, CD90, Stro-1, and CD271. None of these represents a canonical marker of MSCs, and therefore, the homogeneity, reproducibility, and consistency of isolated populations are not assured. The author's laboratory has produced an avian immune phage display library to overcome potential immune tolerance and to generate a new antibody discovery platform for human MSCs. It has been well demonstrated that MSC proliferative activity *in vitro* is high and that when exposed to TGF-β they have a chondrogenic capacity that far exceeds that of primary chondrocytes in cultures. It was also proposed that MSCs are fully tolerated in an allogenic setting when delivered to an immunocompetent host, which created new opportunities for the therapeutic use of these cells without the need for a tissue biopsy.

Prof. De Bari discussed how the use of MSCs is being pursued as chondrocyte substitutes in an autologous chondrocyte implantation equivalent procedure because MSCs are easily accessible, easy to isolate and to expand in culture, and the ability to form cartilage and bone. He also proposed that they appear to be immune privileged under specific conditions. At the same time, preclinical and clinical studies are needed to compare MSCs with articular chondrocytes and see if implantation of MSCs will result in a cartilage tissue that is durable and if the use of MSCs would extend the application of cell-based technologies to nonlocalized, chronic lesions in OA patients as has been reported.[36] Prof. De Bari also noted that there is evidence that MSCs can be poorly immunogenic *in vivo* under specific conditions[37]; however, the differentiation into a mature phenotype of the implanted stem cell is likely to result in the loss of the immunological privilege with consequent rejection. He also discussed that another approach to the repair of the joint surface could be the activation of intrinsic regenerative mechanisms by using medications that target the stem cells naturally present in their own environments and related reparative signaling pathways. In this respect, several joint associated tissues such as synovial membrane and fluid, fat pad, periosteum, bone marrow, and even the articular cartilage itself have been reported to contain cells that after isolation and culture expansion display properties of MSCs.

Concluding Remarks

The 2012 ICRS meeting brought together clinicians, health care professionals, and basic scientists and provided an overview of the current state-of-the-art in the field of cartilage repair. Based on this, we identified that the field is moving toward induction of endogenous repair, patient profiling, and the use of cartilage fragments and extracellular matrix scaffolds. Although the use of stem cells is promising, additional markers are needed. There is a need for better-controlled clinical trials, particularly for newer biological therapies such as PRP. In addition, preclinical models continue to be needed with better definition of appropriate model selection.

Information will continue to emerge at the next meeting in Izmir, Turkey, in September 2013.

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Ethical Approval

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References

1. Anderson DD, Van Hofwegen C, Marsh JL, Brown TD. Is elevated contact stress predictive of post-traumatic osteoarthritis for imprecisely reduced tibial plafond fractures? J Orthop Res. 2011;29(1):33-9.
2. McIlwraith CW, Fortier LA, Frisbie D, Nixon AJ. Equine models of articular cartilage repair. Cartilage. 2011;2:317-26.
3. Hurtig MB, Buschmann MD, Fortier LA, Hoemann C, Hunziker EB, Jurvelin JS, *et al*. Preclinical studies for cartilage repair: recommendations from the International Cartilage Repair Society. Cartilage. 2011;2(2):137-52.
4. Malda J, Benders KE, Klein TJ, de Grauw JC, Kik MJ, Hutmacher DW, *et al*. Comparative study of depth-dependent characteristics of equine and human osteochondral tissue from the medial and lateral femoral condyles. Osteoarthritis Cartilage. 2012;20(10):1147-51.
5. Bauer DC, Hunter DJ, Abramson SB, Attur M, Corr M, Felson D, *et al*. Classification of osteoarthritis biomarkers: a proposed approach. Osteoarthritis Cartilage. 2006;14(8):723-7.
6. Poole AR, Ionescu M, Fitzcharles MA, Billinghurst RC. The assessment of cartilage degradation in vivo: development of an immunoassay for the measurement in body fluids of type II collagen cleaved by collagenases. J Immunol Methods. 2004;294(1-2):145-53.
7. Cibere J, Zhang H, Garnero P, Poole AR, Lobanok T, Saxne T, *et al*. Association of biomarkers with pre-radiographically defined and radiographically defined knee osteoarthritis in a population-based study. Arthritis Rheum. 2009;60(5):1372-80.
8. Cahue S, Sharma L, Dunlop D, Ionescu M, Song J, Lobanok T, *et al*. The ratio of type II collagen breakdown to synthesis and its relationship with the progression of knee osteoarthritis. Osteoarthritis Cartilage. 2007;15(7):819-23.
9. Mullan RH, Matthews C, Bresnihan B, FitzGerald O, King L, Poole AR, *et al*. Early changes in serum type II collagen biomarkers predict radiographic progression at one year in inflammatory arthritis patients after biologic therapy. Arthritis Rheum. 2007;56(9):2919-28.
10. Fitzgerald J, Rich C, Burkhardt D, Allen J, Herzka AS, Little CB. Evidence for articular cartilage regeneration in MRL/MpJ mice. Osteoarthritis Cartilage. 2008;16(11):1319-26.

11. Rai MF, Hashimoto S, Johnson EE, Janiszak KL, Fitzgerald J, Heber-Katz E*, et al*. Heritability of articular cartilage regeneration and its association with ear wound healing in mice. Arthritis Rheum. 2012;64(7):2300-10.
12. Seifert AW, Kiama SG, Seifert MG, Goheen JR, Palmer RM, Maden M. Skin shedding and tissue regeneration in African spiny mice (Acomys). Nature. 2012;489(7417):561-5.
13. Mapp PI, Walsh DA. Mechanisms and targets of angiogenesis and nerve growth in osteoarthritis. Nat Rev Rheumatol. 2012;8(7):390-8.
14. Suri S, Gill SE, Massena de Camin S, Wilson D, McWilliams DF, Walsh DA. Neurovascular invasion at the osteochondral junction and in osteophytes in osteoarthritis. Ann Rheum Dis. 2007;66(11):1423-8.
15. Woodruff MA, Hutmacher DW. The return of a forgotten polymer—polycaprolactone in the 21st century. Prog Polym Sci. 2010;35(10):1217-56.
16. Schuurman W, Khristov V, Pot MW, van Weeren PR, Dhert WJ, Malda J. Bioprinting of hybrid tissue constructs with tailorable mechanical properties. Biofabrication. 2011;3(2):021001.
17. Klein TJ, Rizzi SC, Reichert JC, Georgi N, Malda J, Schuurman W*, et al*. Strategies for zonal cartilage repair using hydrogels. Macromol Biosci. 2009;9(11):1049-58.
18. Ballyns JJ, Cohen DL, Malone E, Maher SA, Potter HG, Wright T*, et al*. An optical method for evaluation of geometric fidelity for anatomically shaped tissue-engineered constructs. Tissue Eng Part C Methods. 2010;16(4):693-703.
19. Keriquel V, Guillemot F, Arnault I, Guillotin B, Miraux S, Amedee J*, et al*. In vivo bioprinting for computer- and robotic-assisted medical intervention: preliminary study in mice. Biofabrication. 2010;2(1):014101.
20. Cohen DL, Lipton JI, Bonassar JL, Lipson H. Additive manufacturing for in situ repair of osteochondral defects. Biofabrication. 2010;2(3):035004.
21. Lee CH, Cook JL, Mendelson A, Moioli EK, Yao H, Mao JJ. Regeneration of the articular surface of the rabbit synovial joint by cell homing: a proof of concept study. Lancet. 2010;376(9739):440-8.
22. Alhadlaq A, Mao JJ. Tissue-engineered osteochondral constructs in the shape of an articular condyle. J Bone Joint Surg Am. 2005;87(5):936-44.
23. Mendelson A, Frank E, Allred C, Jones E, Chen M, Zhao W*, et al*. Chondrogenesis by chemotactic homing of synovium, bone marrow, and adipose stem cells in vitro. FASEB J. 2011;25(10):3496-504.
24. Goodrich LR, Choi VW, Carbone BA, McIlwraith CW, Samulski RJ. Ex vivo serotype-specific transduction of equine joint tissue by self-complementary adeno-associated viral vectors. Hum Gene Ther. 2009;20(12):1697-702.
25. Frisbie DD, Ghivizzani SC, Robbins PD, Evans CH, McIlwraith CW. Treatment of experimental equine osteoarthritis by in vivo delivery of the equine interleukin-1 receptor antagonist gene. Gene Ther. 2002;9(1):12-20.
26. Morisset S, Frisbie DD, Robbins PD, Nixon AJ, McIlwraith CW. IL-1ra/IGF-1 gene therapy modulates repair of microfractured chondral defects. Clin Orthop Relat Res. 2007;462:221-8.
27. Sanchez M, Azofra J, Anitua E, Andia I, Padilla S, Santisteban J*, et al*. Plasma rich in growth factors to treat an articular cartilage avulsion: a case report. Med Sci Sports Exerc. 2003;35(10):1648-52.
28. Siclari A, Mascaro G, Gentili C, Cancedda R, Boux E. A cell-free scaffold-based cartilage repair provides improved function hyaline-like repair at one year. Clin Orthop Relat Res. 2012;470(3):910-9.
29. Sampson S, Reed M, Silvers H, Meng M, Mandelbaum B. Injection of platelet-rich plasma in patients with primary and secondary knee osteoarthritis: a pilot study. Am J Phys Med Rehabil. 2010;89(12):961-9.
30. Kon E, Buda R, Filardo G, Di Martino A, Timoncini A, Cenacchi A*, et al*. Platelet-rich plasma: intra-articular knee injections produced favorable results on degenerative cartilage lesions. Knee Surg Sports Traumatol Arthrosc. 2010; 18(4):472-9.
31. Filardo G, Kon E, Pereira Ruiz MT, Vaccaro F, Guitaldi R, Di Martino A*, et al*. Platelet-rich plasma intra-articular injections for cartilage degeneration and osteoarthritis: single-versus double-spinning approach. Knee Surg Sports Traumatol Arthrosc. 2012;20(10):2082-91.
32. Kon E, Mandelbaum B, Buda R, Filardo G, Delcogliano M, Timoncini A*, et al*. Platelet-rich plasma intra-articular injection versus hyaluronic acid viscosupplementation as treatments for cartilage pathology: from early degeneration to osteoarthritis. Arthroscopy. 2011;27(11):1490-501.
33. Cole BJ, Farr J, Winalski CS, Hosea T, Richmond J, Mandelbaum B*, et al*. Outcomes after a single-stage procedure for cell-based cartilage repair: a prospective clinical safety trial with 2-year follow-up. Am J Sports Med. 2011;39(6):1170-9.
34. Frisbie DD, Lu Y, Kawcak CE, DiCarlo EF, Binette F, McIlwraith CW. In vivo evaluation of autologous cartilage fragment-loaded scaffolds implanted into equine articular defects and compared with autologous chondrocyte implantation. Am J Sports Med. 2009;37(Suppl. 1):71S-80S.
35. Lu Y, Dhanaraj S, Wang Z, Bradley DM, Bowman SM, Cole BJ*, et al*. Minced cartilage without cell culture serves as an effective intraoperative cell source for cartilage repair. J Orthop Res. 2006;24(6):1261-70.
36. Wakitani S, Imoto K, Yamamoto T, Saito M, Murata N, Yoneda M. Human autologous culture expanded bone marrow mesenchymal cell transplantation for repair of cartilage defects in osteoarthritic knees. Osteoarthritis Cartilage. 2002; 10(3):199-206.
37. MacDonald GI, Augello A, De Bari C. Role of mesenchymal stem cells in reestablishing immunologic tolerance in autoimmune rheumatic diseases. Arthritis Rheum. 2011;63(9):2547-57.