



GR
BOI³

**27th Interdisciplinary
Research Conference
on Injectable Biomaterials
and Interventional Procedures**

Athens
18-21 May 2017



www.griboi2017.com
email: info@griboi2017.com
www.griboi.org

ABSTRACTS

Congress Secretariat:

 **Theotokis Travel
Congresses**
 **Lufthansa City Center**

Phone: +30 210 6852892
email: info@theotokistravel.gr
www.theotokistravel.gr

Backpropagation neural network-based fast CT imaging screening recognized and predicted trabecular bone microdamage in osteoporotic lumbar vertebrae: a pilot study

Chentian Li¹, Lei Xu¹, [Wing Yuk Ip¹](#), [William W. Lu¹](#)

¹Department of Orthopedics & Traumatology, The University of Hong Kong, Hong Kong, China

INTRODUCTION: Mechanical stress concentration in trabecular bone could cause a potential risk of microfracture[1], and large fracture further, in patient's vertebrae. Establishing a neural network model for prediction of stress concentration based on CT images would be helpful to analyze bone weakness in the individual vertebra, and predict bone fracture risks as a fast screening tool.

METHODS: 15 clinical CT image datasets of lumbar vertebrae from 3 patients with various severity of osteoporosis were drawn. Four sub-regions of interest were randomly selected in each lumbar vertebral trabecular bone. Uniaxial compression was simulated by finite element analysis. A modified texture based analysis was used to recognize the trabecular morphology. Eigenvalues of texture characterization, trabecular morphology, and anisotropy degree were inputted into a back-propagation neural network model, while the stress-concentration factor from the finite-element analysis was set as output data. The backpropagation neural network model was trained based on Levenberg–Marquardt algorithm to predict vertebral trabecular bone stress-concentration, and compared with finite element analysis outcomes.

RESULTS: The average osteoporotic vertebral trabecular bone texture anisotropy degree was 0.0041 ± 0.1561 . The average apparent trabecular bone stiffness was 4.8348 ± 2.4962 GPa. The average stress concentration factor of trabecular bone was 1.0726 ± 0.3636 . A high correlation between stress-concentration and trabecular bone morphology and anisotropy was found ($R=0.89223$). A 4-dimension tensor-based back-propagation neural network model was established and precisely predicted stress-concentration factor based on texture properties of trabecular bone ($R=0.89286$).

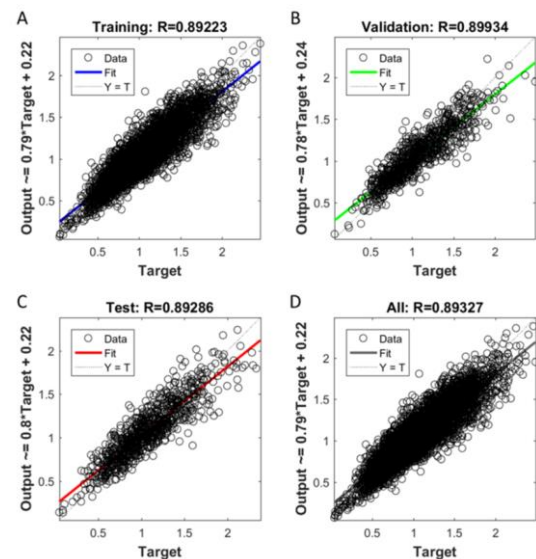


Fig.1: The fitting regression. (The correlation between texture analysis results and stress concentration in A. Training datasets; B. Validation datasets; C. Test datasets; and D. Overall datasets.)

DISCUSSION & CONCLUSIONS: This study established a neural network model based on image texture analysis, which could quickly screen clinical CT imaging data and precisely predict the trabecular bone stress concentration. This technique could be treated as an alternative to complicated finite-element analysis for bone fracture analysis, especially for fast screening in a large population. Further study would be focused on the relationship between the neural network-based CT imaging screening outcomes and the vertebral bone fracture risk in the large-sample clinical trial.

REFERENCES: ¹ Nagaraja, Srinidhi, Tracey L. Couse, and Robert E. Guldborg. *Journal of biomechanics* 38.4 (2005): 707-716.

ACKNOWLEDGEMENTS: This work was supported by grants from the GRF of Research Grant Council of Hong Kong.

Modifying ready-to-use injectable calcium phosphate bone cements for in-situ pore formation and protein delivery applications

M Schumacher¹, S Rößler², T Hanke², A Lode¹, M Gelinsky¹

¹*Centre for Translational Bone-, Joint- and Soft Tissue Research, Technische Universität Dresden, Dresden, Germany* ²*Max Bergmann Center of Biomaterials and Institute for Materials Science, Technische Universität Dresden, Dresden, Germany*

INTRODUCTION: Pasty calcium phosphate cements (pCPC) based on a non-aqueous carrier liquid have been developed to allow micro-invasive application of the material into specific bone defects. Such ready-to-use pastes avoid the limitations of conventional, water-based cements, amongst others their limited application time. Setting of pCPC is delayed until water from the immersion medium or surrounding tissue replaces the oil-based carrier liquid [1]. As conventional cements, pCPC are well suited as carriers for therapeutic substances to be delivered into a specific bone defect. A major drawback, however, is the absence of macro-pores in the set cement, limiting tissue penetration into the material and thus delaying bone remodelling.

By integration of highly soluble secondary phases (bioactive glass particles as well as silica-based xerogels) into the cement we have evaluated time-dependent pore formation and mechanical properties of the set material, protein release kinetics and *in vitro* cytocompatibility.

METHODS: An α -tricalcium phosphate-based pCPC (InnoTERE GmbH, Germany) was modified with up to 10 wt-% of either mesoporous bioactive glass (MBG) particles, derived via a template-assisted sol-gel process [2] or collagen-silicate xerogel (XER) granules [3]. The resulting composites were allowed to set in silicone moulds at 100% humidity and 37°C for 3 days. During subsequent immersion in water or buffer solution (modified simulated body fluid (mSBF)) development of porosity was investigated. Mechanical properties of set cements were characterised at different time points. MBG-containing composites were further loaded with the vascular endothelial cell growth factor (VEGF) and the release was studied in cell culture medium using ELISA. A cell-based bioactivity assay [2] of released VEGF was performed. Cytocompatibility of both composites was studied using primary human mesenchymal stem cells.

RESULTS: Open porosity of both composites was shown to increase during ageing over 21 days. A maximum increase of ~50% was found for pCPC with 10 wt-% XER content (Fig. 1A). However, increase of porosity also resulted in a decrease in mechanical strength, which was less pronounced in case of MBG-containing composites (Fig. 1B), suggesting a contribution of MBG degradation products to the cement setting reaction.

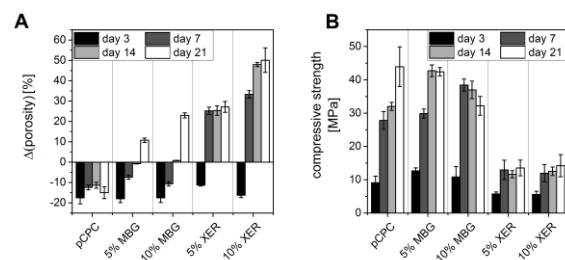


Fig. 1: Relative change of open porosity (A) and compressive strength (B) of pCPC composites containing either MBG or XER over 21 days.

VEGF release from MBG-containing composites was controlled by the presence of MBG particles, and biological activity of the growth factor was maintained over the duration of the experiment. Both composites were found to be cytocompatible.

DISCUSSION & CONCLUSIONS: Composites from pCPC and either MBG or XER were found to exhibit increasing porosity over time. MBG-containing composites were also shown to be suitable for the defect-specific delivery of biologically active growth factors. Both composites had mechanical and biocompatibility characteristics suggesting them for the reconstruction of small, non-load-bearing bone defects.

REFERENCES: ¹S. Heinemann et al. (2013) *Acta Biomater* **9**:6199-207. ²M. Schumacher et al. (2017) *Biomater Sci* DOI: 10.1039/C6BM00903D ³S. Heinemann et al. (2013) *Acta Biomater* **5**:1979-90

ACKNOWLEDGEMENTS: This work was funded by the DFG within SFB Transregio 79.

Biomechanical analysis by finite element method of cement effects for stabilisation of tibial plateau split depression fracture

D Belaid^{1,2}, T Vendevre^{2,3,5}, A. Germaneau², A. Bouchoucha¹,
F. Brémand², C. Brèque^{2,4}, P. Rigoard^{2,5}

¹ *Université des Frères Mentouri, Constantine, Algery.* ² *Institut Pprime UPR 3346, CNRS – Université de Poitiers – ISAE-ENSMA, France.* ³ *Department of Orthopaedic Surgery and Traumatology, CHU, Poitiers France.* ⁴ *Laboratory of Anatomy, Université de Poitiers, France.* ⁵ *Department of Neurosurgery, Prismatic Lab, CHU, Poitiers, France.*

INTRODUCTION: Tibial plateau fracture treatment is carried out in most cases by stable fixation in order to allow early mobilization. Minimally invasive technologies such as tibioplasty or stabilization by locking plate and cement augmentation have recently been used to treat this type of fracture [1]. This work deals with a numerical approach with a personalized finite element (FE) analysis, to determine the mechanical behaviour of the fractured tibial plateau stabilisation. A 3D patient-specific model of the proximal tibia was developed, simulating the fracture and the treatment with locking plate and cement augmentation. The stress distribution was analysed in order to study the effect of cement in the postoperative stabilization of proximal tibial fracture, compared with an intact tibial plateau.

METHODS: A clinical case was analysed and modelled, corresponding to a female patient of 24 years presenting a tibial plateau fracture with a lateral split depression. This fracture was treated with minimally invasive surgery: a balloon was used for bone augmentation and reduction of the depression, and a plate was fixed by locked screws to stabilize the split. PMMA cement was injected in the cavity provided by the balloon. The computed tomography data of the post-operative knees was used to create the personalized FE simulation. The orthotropic elastic material properties of trabecular bone have been incorporated into the FE models using parameters derived from the literature [2]. The applied loading was chosen to simulate a single leg stance during gait, which corresponds to a joint contact force equal to three times the body weight (7).

RESULTS: The maximum values of the von Mises equivalent stress obtained for the fractured tibia treated with and without cement were 134.9MPa and 289.9MPa, respectively. The higher stress value was located in the locking plate and the lower stress of 13.8MPa (with cement augmentation), 23.5MPa (without cement

augmentation) were located in the trabecular bone. Figure 1 shows the distribution of equivalent stress in the trabecular bone. Without cement augmentation, high values tended to be concentrated around the upper screws near the fracture site and the cavity, whereas stress concentrations were reduced with cement.

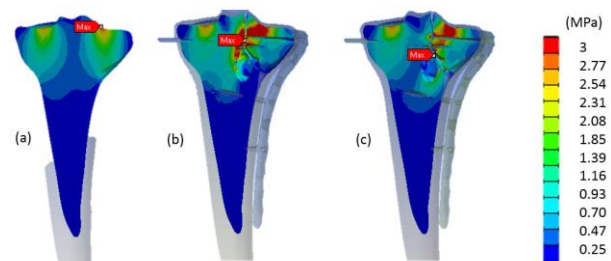


Fig. 1: Distribution of equivalent Von Misès stress in trabecular bone for (a) intact model, fractured and stabilised tibial plateau (b) without cement or (c) with cement.

DISCUSSION & CONCLUSIONS: This biomechanical study investigated the effect of additional cement augmentation after balloon reduction of tibial plateau impression fractures using a computed method. The analysis of stress distributions highlighted that the cement filling of the tibial depression fracture may increase implant stability and decrease the loss of depression reduction. FE approach showed that the injected cement could reduce the loss of fracture reduction after balloon augmentation and could change stress distribution leading to a decrease of the stress shielding. This also ensures the congruence of the fragments by reducing inter-fragment displacements.

REFERENCES: ¹ T. Vendevre et al (2013) *Orthop Traumatol Surg Res*, **99**:267-272. ² J.Y. Rho (1996), *Ultrasonics*, **34**(8):777-783. ³ W. Taylor et al (2004), *J Orthop Res*, **22**(3):625-632

Kyphoplasty vs Stentoplasty: influence on the fracture reduction and on the risk of cement leakage

T Vendevre^{1,2}, P. Brossard¹, JB Pic¹, A Germaneau²,
LE Gayet¹, P Pries¹, P Rigoard^{2,3}, S Teyssédou¹

¹ Department of Orthopaedic Surgery and Traumatology, CHU, Poitiers France. ² Institut Pprime UPR 3346, CNRS – Université de Poitiers – ISAE-ENSMA, France. ³ Department of Neurosurgery, Prismatic Lab, CHU, Poitiers, France.

INTRODUCTION: Kyphoplasty and stentoplasty are two minimally invasive surgery methods used for the reduction and the stabilisation of spine fractures [1]. The principle of kyphoplasty consists in the restoration of the vertebral body anatomy by balloon expansion after a compression burst fracture (Magerl A) [2]. Reinforcement of the anterior vertebral column is obtained from cement injection. Recently, a stentoplasty technique has been developed to reduce height loss after balloon ablation [3]. The objective of this work was to compare effects linked to kyphoplasty and to stentoplasty from radiological data.

METHODS: A monocentric, prospective and continuous study was performed on 60 patients (mean age: 47.3 years) between October 2011 and November 2014. The main inclusion criterion was a recent non-osteoporotic fracture of the thoracolumbar junction. A first group of patients was treated by kyphoplasty (Kyphon®, Medtronic) and the second one by stentoplasty (VBS®, Synthes). The filling was ensured by high viscosity PMMA cements: Biomet Bone Cement V® (Biomet) and Vertecem V+® (Synthes), respectively. The main validation criteria were the post-operative correction of the vertebral kyphosis and its persistence at 3 months. Secondly, complementary parameters were measured: vertebral heights, Farcy index, Beck index, subsidence percentage, behaviour of adjacent discs, cement leakages and their positioning.

RESULTS: The mean reduction of vertebral kyphosis after 3 months was about $4.73 \pm 4.8^\circ$ after kyphoplasty, and about $4.63 \pm 2.7^\circ$ after stentoplasty ($p=0.9393$). No significant statistical difference was revealed for the following parameters: height restoration of vertebral body, Farcy or Beck indexes and subsidence percentage. Cement leakages were significantly more frequent ($p=0.0023$) for kyphoplasty (41.7%) compared to stentoplasty (4.2%). Radiologic analysis of disks did not reveal any compensation ability (Fig. 1).

Study of disks affected by leakage would tend to a decrease of adaptation ($p=0.0579$).



Fig. 1: Examples of cement leakage.

DISCUSSION & CONCLUSIONS:

There is no significant difference of fracture reduction between kyphoplasty and stentoplasty. A higher rate of cement leakage was observed for specimens stabilized by kyphoplasty with a potential disk impact. The difference in the results obtained by stentoplasty can be explained by an eventual difference in viscosity between both cements and by the contribution of the stent to slow the flow. This tendency could be confirmed by flow simulations (Fig. 2).

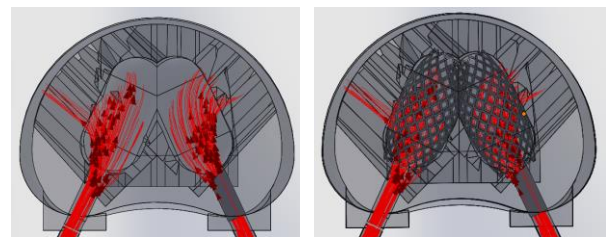


Fig. 2: Kyphoplasty vs Stentoplasty, flow simulation for leakage analysis.

REFERENCES: ¹ M. Freslon, et al (2008) *Rev Chir Orth* **94**:22-35. ² R. Oner, et al (2005) *Injury* **36**:82-89. ³ R. Rotter, et al (2010) *Eur Spine J* **19**(6):916-923.

Title of Abstract:

Minimally Invasive Palliative Procedures in Oncology and Haematology: a retrospective review of 12 months practice in New Zealand

AIMS:

Minimally invasive palliative procedures (MIPP) are sometimes considered Step 4 of the World Health Organization's three-step ladder. Spinal and extra-spinal cementoplasty, thermal ablation of bone and soft tissue metastases including cryoablation and radiofrequency ablation and neurotomies were some of the interventional radiology techniques used to treat patients with bone metastases. We created a multidisciplinary case conference to facilitate access to MIPP for advanced cancer patients, most of them with severe pain not responding to conventional analgesics or cancer modifying therapies such as radiation. The aims of the study are to explore the impact of case conference on clinical decision-making in a real-world setting and assess the tolerability of MIPP in this group of frail and very ill patients.

BACKGROUND:

The MIPP service is a relatively uncommon service available only in several major centres with appropriately trained interventional radiologists. There are relatively limited studies on MIPP. To the best of our knowledge, this is the first MIPP service in New Zealand and Australia. The result of this study in a tertiary referral centre will assist in clarifying the timing and role of MIPP amongst other valuable treatment options by other specialty colleagues in treating complex patients with advanced cancer and severe pain.

METHODS:

A database was prospectively designed and data was prospectively collected including demographic details, cancer diagnosis, recommendations of the committee and rationale, date of procedure recommended, complications and date of death. At 14 months after the initiation of this new MIP service, a retrospective analysis of the database, electronic charts, hard copy clinical correspondences, images on the radiology PACS system of all cancer patients who had MIPP procedures and /or were referred to the MIPP case conference were reviewed. All procedures were performed by the first author of this study.

RESULTS:

Fifty case referrals to the MIPP service were made between 3 August 2015 and 1 October 2016. Twenty-five patients with advanced cancer with pain refractory to conventional analgesics or cancer modifying therapies had MIPP procedures performed over 34 sessions. There were no clinically significant complications or adverse events. All procedures were well tolerated.

CONCLUSION:

A multidisciplinary case conference made a positive impact on the management of complex patients with advanced cancer. Approximately 50% of referrals to the MIPP service resulted in recommendation for MIPP procedure by the multidisciplinary MIPP committee. The remaining 50% of patients gained access to a comprehensive multidisciplinary review that resulted in enhanced care, for example, optimization of opiate analgesia.

The MIPP procedures were safe and well-tolerated, with some patients gaining tremendous pain relief. A small number of patients had modest or no significant improvement. None were reported to be worse than their status prior to having MIPP.

The MIPP service is a valuable service to oncologists and haematologists with patients who have exhausted conventional analgesia or other cancer modifying therapies including radiation therapy.

Authors:

Colin CW Chong¹⁻⁵, **Rachel Wiseman**⁴, **Kate Grundy**⁴, **Brendon Anderson**⁴, **Ruth Spearing**⁴, **Andrew Butler**⁴, **et al.**

Institutions:

1. I-Med Radiology Network, Queensland, Australia.
2. Wesley Hospital, Brisbane, Queensland, Australia.
3. St. Andrew's War Memorial Hospital, Brisbane, Queensland, Australia.
4. Canterbury Regional Cancer and Haematology Service, Canterbury District Health Board, Christchurch, New Zealand.
5. Mercy Hospital, Auckland, New Zealand.

Corresponding Author:

Colin Chong MBBS, M.Med, FRANZCR

Email: drcolincwchong@gmail.com

Tel: +61 411 328 698

Effect of blood proteins on the strength, setting time and relative porosity of novel Portland cement for vertebroplasty

A Moetazedian¹, MP Hofmann¹, RM Shelton¹

¹Biomaterial unit, School of Dentistry, University of Birmingham, United Kingdom

INTRODUCTION: Portland cement (PC) is a hydraulic calcium silicate cement which with addition of a radioopacifier is used as mineral trioxide aggregate (MTA) in Dentistry [1, 2]. Recently, a novel PC-based bone cement was proposed for percutaneous vertebroplasty (PVP); that is stabilisation of fractured vertebral bodies [1]. Adding CaCl₂ to PC acts as both liquefier and setting accelerant, which improves injectability and PC strength to a level comparable with poly methylmethacrylate (PMMA) cement [1]. Exposure of MTA to blood and serum proteins, adversely affected the PC setting time and strength [2]. To date, no study has investigated the effect of serum proteins on this novel PC for PVP at a high powder-to-liquid ratio (PLRs).

MATERIALS & METHODS: Six cement groups (G1-6) were prepared, in which the powder phase consisted of 75:20:5 ratio of PC, Bi₂O₃ and CaCl₂. Single proteins or the complex mixture in foetal bovine serum (FBS) were either added to the liquid phase (intrinsic) (G2-5) or used coating the inner walls of the sample mould (extrinsic) (G6) see Table 1. All slurries were hand-mixed for 2min at a PLR of 4g/ml and set at 37°C for 24h.

Table 1. Protein addition to the cement groups.

| Approach | Groups | protein | wt % |
|-----------|--------|------------|------|
| Control | G 1 | - | - |
| Intrinsic | G 2 | albumin | 5.5 |
| | G 3 | fibrinogen | 0.4 |
| | G 4 | both | 5.9 |
| | G 5 | FBS | 5.9 |
| Extrinsic | G 6 | FBS | |

The initial setting time of cements were measured using the Gilmore needles test. The wet compressive strength (CS) (n>6) was measured using a universal testing machine at a crosshead speed of 1mm/min after 1 and 7 days in distilled water. The relative porosity (RP) of dried fragments were measured using helium pycnometry and weight loss experiments. All data were analysed for a significance (p<0.05).

RESULTS: Incorporation of proteins and FBS significantly reduced CS compared with the control at both incubation times (Table 2.). After 7 days,

all groups except fibrinogen increased the RP. Albumin doubled the setting time, whilst fibrinogen had no effect. FBS incorporation increased the setting time approximately 5 times. In contrast, exposure of FBS had no influence on the setting time (Fig. 1).

| Groups | 1-day CS (MPa) | 7-day CS (MPa) | 1-day RP (%) | 7-day RP (%) |
|--------|----------------|----------------|--------------|--------------|
| G 1 | 40 ± 2 | 55 ± 2 | 21 ± 2 | 17 ± 2 |
| G 2 | 25 ± 2* | 40 ± 3* | 23 ± 1 | *21 ± 1 |
| G 3 | 28 ± 5* | 42 ± 2* | 20 ± 1 | 16 ± 2 |
| G 4 | 32 ± 3* | 40 ± 2* | 22 ± 1 | *21 ± 1 |
| G 5 | 25 ± 2* | 34 ± 3* | 26 ± 1* | *23 ± 1 |
| G 6 | 35 ± 3 | 39 ± 2* | 21 ± 2 | *22 ± 2 |

Table 2. CS and RP of PC groups after two incubation times. * indicates significant difference to control.

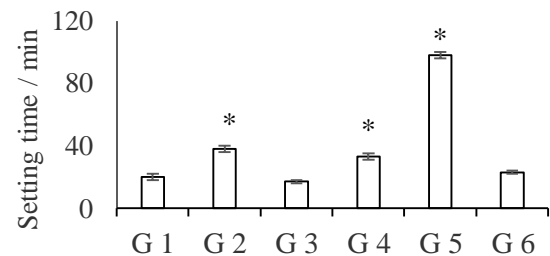


Fig.1 the initial setting time of G 2, 4 and 5 significantly (*) increased compared with the control.

DISCUSSION & CONCLUSIONS: Particular proteins at their highest possible concentration *in vivo* affected the novel PC differently. Albumin's attachment potentially via a calcium-binding site may have disrupted the kinetic solubility of PC, thus doubled the setting time, while fibrinogen's large fibrous-like structure could have helped the hydration [3]. The adverse effect of fibrinogen may have been due to increasing flaws rather than increasing porosity. Incorporation of blood into these PCs should be prevented in application.

REFERENCES: ¹Wynn-Jones G et al. J Biomed Mater Res B Appl Biomater. 2012; 100B(8):2213-2221. ²Nekoofar M et al. International Endodontic Journal. 2010;43(9):782-791. ³Chauhan S et al. Key Engineering Materials. 2006;309-311:841-844.

Smart hydrogels for delivery of drug agents against pancreatic cancer

[D Skoulas](#)¹, [H Iatrou](#)¹

¹ [Laboratory of Industrial Chemistry](#), Chemistry Department, University of Athens, Greece.

INTRODUCTION: While the overall incidence of cancer mortality has steadily decreased over the last several decades, in pancreatic cancer (PC) the mortality is still increasing. Although nanoparticulate drug delivery through intravenous administration transports drugs to most parts of the body, PC presents severe barriers that prevent the nanoparticles from reaching the target. At this tumor it is necessary to apply local administration. The potential benefits of localized chemotherapy are intended to both enhance the efficacy of treatment and reduce patient morbidity. The best systems to achieve local drug release involve hydrogels, but they must present specific characteristics in order to achieve effective and sustained release and should be simply administered to reduce patient morbidity.

METHODS: We have synthesized multifunctional polymers based on polypeptides which are suitable for the selective local delivery of gemcitabine through hydrogel formation to pancreatic cancer rather than healthy tissues. We have shown that they can be implanted very easily by injection through a thin needle and can be immediately reconstructed close to the cancer tissues, due to the combination of their advanced macromolecular architecture and of the unique properties of the hydrophobic part of the polymers whose secondary structure denatures under shear rate and quickly self-heals when the shear rate stops. Once the hydrogels are injected close to cancer tissues, they can selectively deliver gemcitabine to cancer rather than to healthy tissues due to their pH and temperature sensitivity, because of the difference of cancer tissue on these conditions. The targeted delivery of the drugs depends not only on the interactions between the polymers and the drugs but also on the steric hindrance which depends on the structural characteristics of the aggregated polymers within the hydrogel.

RESULTS: We have already examined the rheological properties of the hydrogels under different pH and temperature conditions, as well as the self-healing properties.

The parameters that influence the strength of the hydrogel are the concentration of the polymer, i.e. the polymer to water content ratio, the temperature and the pH. It was found that the concentration of the hydrogel significantly influences the storage modulus G' as well as the loss modulus G'' of the hydrogel, as well as the time required for the hydrogel to self-heal. In addition, we have performed *in vivo* tests of the hydrogels in special mice, showing that the multifunctional hydrogels are able to direct the delivery of the drugs selectively to the cancer tissue and achieve lower tumor growth with smaller amount of anticancer drug gemcitabine.



Fig. 1: Hydrogels at different p

DISCUSSION & CONCLUSIONS: These biomaterials exhibit the simplest, less invasive possible implantation, i.e. not with surgery but via an injectable *in situ*-forming hydrogel that will be placed at the vicinity of the cancer tissue, not inside it. They demonstrate slow release characteristics. They can deliver a mixture of already approved chemotherapeutic drug (hydrophilic gemcitabine and hydrophobic paclitaxel) and genes towards the cancer tissues and diminish cytotoxicity side effects to healthy organs. The material is non-cytotoxic and biodegradable, and its surgical removal after drug delivery is not necessary.

REFERENCES: ¹ L.M. Pakstis, B. Ozbas, T.J. Deming, et al (2004) *Biomacromolecules* **5**:312-318 ² A.P. Nowak, T.J. Deming, et al (2002) *Nature* **417**:424-428 ³V. Breedveld, et al (2004) *Macromolecules* **37**:3943-3953.

The placebo effect of the guided PRP injections in patients with lateral epicondylitis: a double-blinded randomized controlled clinical trial

M-A Malahias^{1,2}, M-K Kasetas¹, E Chronopoulos¹, V S. Nikolaou¹, A P. Efstathiou¹, G C. Babis¹

¹*2nd Orthopaedic Department, National and Kapodistrian University of Athens Medical School, Athens, Greece*

²*International Centre for Hip, Knee and Foot Surgery, Sport Traumatology, ATOS Klinik, Heidelberg, Germany*

INTRODUCTION: According to recent systematic reviews of the literature, there is a paucity of evidence from unbiased trials on which to base treatment recommendations regarding injection therapies for lateral epicondylitis. Despite the trend of using PRP (platelet-rich plasma) nowadays in the treatment of chronic lateral epicondylitis, it remains unclear whether this therapeutic option is advantageous -comparing to placebo injections- or not.

METHODS: We conducted a double-blinded, randomized, prospective, placebo-controlled, clinical trial enrolled 29 patients with clinical and sonographic evidence of chronic (> 3 months) lateral epicondylitis, non-responsive to conservative treatment. Our patients were selected by a nurse picking their name from a sealed closed envelope. The physician who performed the injection was unaware of the injected content (we achieved this through the coverage of the syringe by the same nurse who opened the envelope). Group A (15 patients) underwent three consecutive ultrasound-guided PRP intratendinous infiltrations within a 30-days period. In addition, group B (14 patients) received three normal saline (N/S 0.9%) ultrasound-guided intratendinous injections at the origin of extensor carpi radialis brevis (ECRB). The patients were asked to use the arm minimally for 3 days and then gradually to return to normal use. The follow-up -including Quick DASH questionnaire, visual analogue scale (VAS 0-100mm), and possible complications- was set at 1, 4 and 12 weeks by another physician, blinded to the treatment of the individuals. After 3 months, if the patient was unsatisfied with the result of treatment, he or she could be released from the study to seek other management. We planned to deploy SPSS, t-test, chi square test ($p < 0.05$) to -statistically- analyse our findings.

RESULTS: None of our patients were lost during the follow-up period. The drop-out rate was 53.3%

for group A (8 patients) and 64.2% (9 patients) for group B. When we compared the pre-injected mean Q-DASH score with the post-injected follow-up endpoint Q-DASH score (12 weeks) in group A patients a 46.6% success ratio concerning Q-DASH difference was found (7 out of 15 patients with more than 25% improvement in final Q-DASH score), while group B patients a 35.7% success ratio was found (5 out of 14 patients). Mean VAS scores in group A were 58 mm preoperatively and, 40 mm, 44 mm and 43 mm after 2, 4, and 12 weeks postoperatively, while in group B were 54 preoperatively and, 34, 39 and 46 postoperatively. No adverse events leading to hospitalization, and no reports of infections resulting from the injections, occurred.

CONCLUSIONS: It was illustrated that the ultrasound-guided PRP intratendinous infiltrations yielded no better results than the normal saline injections in the treatment of chronic lateral epicondylitis. The authors identify the placebo effect as an important factor interfering with the PRP-treated patients, which has to be further investigated.

EVOLVE: A prospective and multicenter evaluation of outcomes for quality of life, pain and activities of daily living for balloon kyphoplasty in the treatment of medicare-eligible subjects with vertebral compression fractures

¹DP Beall, ²MR Chambers, ³SM Thomas, ⁴R Easton, ⁵S Talati, ⁶B Goodman, ⁷D Datta, ⁸JR Webb, ⁹D Linville

¹ Clinical Radiology of Oklahoma, Oklahoma city, OK ² University of Alabama at Birmingham, Birmingham, AL, USA., ³ Allegheny University, Meadville, PA, USA. ⁴ Beaumont Health System, Troy, MI, USA. ⁵ Advanced Diagnostic Imaging, PC, Saginaw, MI, USA. ⁶ Alabama Ortho Spine and Sports, Birmingham, AL, USA. ⁷ The Back Center, Melbourne, FL, USA. ⁸ The Osteoporosis Center, Tulsa, OK, USA. ⁹ Scoliosis & Spine Surgery Clinic of Memphis, PLLC, Memphis, TN, USA

INTRODUCTION: Vertebral compression fractures (VCFs) resulting from osteoporosis or cancer are common and painful. Worldwide, osteoporosis causes >8.9 million fractures annually. Quality of life is threatened and mortality and morbidity rates increase following a VCF. Kyphoplasty is a minimally invasive procedure to stabilize a VCF, reduce pain, and improve quality of life and mobility.

METHODS: 354 Medicare patients with painful, acute or subacute, VCFs were prospectively enrolled at 24 sites with 350 undergoing balloon kyphoplasty. Primary objective was to show statistically significant improvement in four co-primary endpoints (SF-36v2 PCS, EQ-5D, NRS back pain and ODI) at 3 months.

RESULTS: Back pain improved from 8.7 (scale 0-10) by 5.2, 5.4, 6.0, 6.2 and 6.3 points, at the 7 day, and the 1, 3, 6 and 12 month time points, respectively ($p < 0.001$ for each). ODI improved from 63.4 (scale 0-100) by 30.5, 35.3, 36.3 and 36.2 points, at the 1, 3, 6 and 12 month time points, respectively ($p < 0.001$ for each). The SF-36 PCS was 24.2 at baseline (scale 0-100) and improved 10.7, 12.4, 13.4 and 13.8 points, at 1, 3, 6 and 12 months ($p < 0.001$ for each). The EQ-5D was 0.383 points (scale 0-1) and improved 0.316, 0.351, 0.356 and 0.358 points, at 1, 3, 6 and 12 months ($p < 0.001$ for each).

There were five AEs considered device- or procedure-related including one asymptomatic balloon rupture, one patient with rib pain and a serious AE of pneumonia aspiration. Another subject had a new VCF 25 days post-procedure considered possibly cement-related; all of these AEs resolved with proper treatment. Asymptomatic cement leakage was reported in 107/499

(21.4%) index levels treated.

DISCUSSION & CONCLUSIONS: Our large, prospective, multicenter study trial demonstrates that balloon kyphoplasty is a safe, very effective and durable procedure for treating Medicare patients with VCFs due to osteoporosis or cancer

REFERENCES:

1. Lindsay, R., et al., *Risk of new vertebral fracture in the year following a fracture*. *Jama*, 2001. **285**(3): p. 320-3.
2. Dawson-Hughes, B., et al., *Implications of absolute fracture risk assessment for osteoporosis practice guidelines in the USA*. *Osteoporos Int*, 2008. **19**(4): p. 449-58.
3. Baaj, A.A., et al., *Trends in the treatment of lumbar spine fractures in the United States: a socioeconomics perspective: clinical article*. *J Neurosurg Spine*, 2011. **15**(4): p. 367-70.
4. Barr, J.D., et al., *Percutaneous vertebroplasty for pain relief and spinal stabilization*. *Spine (Phila Pa 1976)*, 2000. **25**(8): p. 923-8.

ACKNOWLEDGEMENTS: Medtronic Spine sponsored this study and contributed to study design, data monitoring, statistical analysis, and reporting of results and paid for independent core laboratory services.

Analgesic drug release from CPCs: first in vivo study using CatWalk system

M Dupleichs¹, O Gauthier^{2,3}, M Masson², E Verron², J M Bouler¹, P Janvier¹
¹CEISAM, UMR CNRS 6230, Nantes University, France. ²LIOAD, INSERM U791, Nantes University, France. ³ONIRIS College of Veterinary Medicine, Nantes, France

INTRODUCTION: Postoperative pain following bone reconstruction is a serious complication that could jeopardize the global success of a surgery. The administration of local anaesthetics has proven to be an effective analgesic technique for the treatment of postoperative pain with a significantly reduced drug use. In this clinical context, we proposed to evaluate the benefit in pain relief obtained with the defect filling by an injectable calcium phosphate cement (CPC) loaded or not with local anaesthetics (bupivacaine or ropivacaine). This study follows previous promising experiments related to the association of bupivacaine with calcium deficient apatite (CDA)[1]. To compare postoperative pain after bone filling surgery, a functional evaluation was performed using gait analysis with the Catwalk system.

METHODS: Different formulations of commercial apatitic cements, mainly constituted by α -TCP (78%), were loaded with bupivacaine and ropivacaine. Local anaesthetics were directly introduced into the solid phase at 8% by weight (w/w). The cement paste was prepared by mixing the obtained powder with an aqueous solution of Na_2HPO_4 . The liquid/powder (l/p) ratio was adapted to obtain a cement compatible with a use in bone surgery. The final product after the setting process was in all cases CDA loaded with a local anaesthetic. Eighteen Wistar female rats were unilaterally implanted for eight weeks with 0%, 8% of bupivacaine and 8% of ropivacaine, in critical cylindrical defect in distal femur (right hind limb). The implantation impact on functional recovery and locomotion of the animals was studied using a "Catwalk" gait analysis system (Noldus).

RESULTS: Incorporation of bupivacaine did not modify the chemical reactivity of cement and did not alter the formation of CDA. As for the ropivacaine-cement, an adaptation of the l/p ratio from 0.45 to 0.48 was performed to ensure an injectable ready-to-use forms. The release of the local anaesthetics (60%) is effective in the critical first four postoperative days lowering the risk of

developing chronic pain. The use of Catwalk system provided a functional evaluation of the pain relief related to the implantation of CPC loaded with two different analgesic molecules. A clear analgesic effect in favor of the loaded cements versus cement alone was observed.

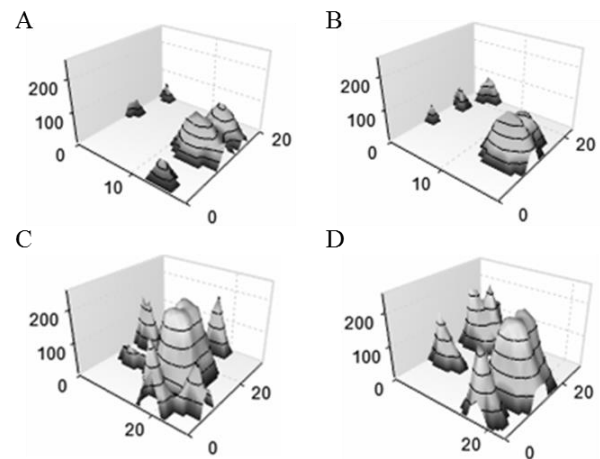


Fig. 1: 3D Footprint Intensities from "Catwalk" of the operated paw (right hindlimb). Unloaded cement (A, B), Bupivacaine-cement (C, D). 24H post-op (A, C), 48H post-op (B, D).

Figure 1 presents the 3D footprint intensities of the right hind paw after the implantation of unloaded and bupivacaine- cements. 24 hours after implantation (A, C), the intensity decreased to 25% and 1%, for unloaded and bupivacaine-loaded CPC respectively compared to a reference value. The difference between these cements was significant. This analgesic effect persists above 48 hours (B, D) and seems to be in accordance with the bupivacaine release profile.

CONCLUSIONS: The CatWalk gait analysis seems to be a relevant tool to evaluate the efficacy in pain relief obtained after defect filling with anaesthetics CPC. Moreover, in view of the analgesic benefit of these biomaterials, it could be therefore possible to include such loaded CPCs into multimodal analgesia associated to autologous iliac bone grafting procedure.

REFERENCES: ¹E.Verron, J.-M. Bouler, O. Gauthier et al (2010) J Biomed Mater Res B Appl Biomater **94**(1):89-96.

Radiofrequency Ablation of Kidney Tumors with High Frequency Jet Ventilation and Real Time US-CT Image Fusion/Virtual Needle-Track Guiding. A Single Center Experience.

Authors: A. Michos¹, V. Grozman¹, T. Josephson²

¹Karolinska University Hospital, Karolinska Institute, Stockholm-Sweden/SE

²Danderyd Hospital, Karolinska Institute, Stockholm-Sweden/SE,

Purpose

In this study we aimed to present the short-term results as well as to assess the efficacy and safety of the Real Time US-CT Image Fusion/Virtual Needle-Track guided (US-CTfusion/VNT) percutaneous Radiofrequency Ablation (RFA) of Kidney Tumors.

Methods and Materials

We have retrospectively reviewed all the patients who were treated with US-CTfusion/VNT guided percutaneous RFA at Danderyd hospital for kidney tumors between September 2014 and November 2015. Our study includes 19 patients (13 males and 6 females). General anesthesia with muscle paralysis and High Frequency Jet Ventilation (HFJV) was used in all cases. In 1 patient with lesions in close proximity to the ureter, an ureteric stent was placed immediately before RFA. In 2 patients was required hydrodissection. A preablation CT scan with intravenous contrast was obtained after the patient had been intubated. Percutaneous biopsy was performed in all the lesions. The Cool-tip RF needle were placed under US-CTfusion/VNT guidance. All patients have been followed at least for 6 months.

Results

In total ablation of 19 lesions was performed, 11/19 (57.9 %) of the tumors were exophytic, 8/19 (42.1 %) of the tumors were non-exophytic. The diameter of the lesions ranged between 0.8 and 4.0 cm. According to percutaneous biopsy, 14/19 (73.7%) of lesions were malignant, 2/19 (10.5 %) were angiomyolipom and 3/19 (15.8%) of biopsies were inconclusive. Per-operative mortality and mortality at 6 months was 0%. In 1 patient (5%) complication were reported occurred in the form of subcapsular bleeding after treatment (no therapy required). Peritoneal seeding at the RFAblation tract developed in one patient within the 3 month follow-up. Of 19 tumors, 2 tumors (10.5 %) required retreatment because of incomplete ablation. All residual tumors were successfully ablated in an additional session of RFA.

Recurrence free survival at 6 months was 94.7%.

Conclusion

Real Time US-CT Image Fusion/Virtual Needle-Track guided percutaneous RF ablation is an effective and safe method for treatment of kidney tumors that can be repeated several times.

References

- (1) Brian C. Allen, Michael Y. Chen, David D. Childs, Ronald J. Zagoria
Imaging-Guided Radiofrequency Ablation of Cystic Renal Neoplasms
- (2) Jung Jae Park, MD, Byung Kwan Park, MD, Sung Yoon Park, MD, and Chan Kyo Kim, MD
Percutaneous Radiofrequency Ablation of Sporadic Bosniak III or IV Lesions: Treatment Techniques and Short-Term Outcomes
- (3) Gideon Lorber, MD,¹ Merce Jorda, MD, PhD,² and Raymond Leveillee, MD¹
Factors Associated with Diagnostic Accuracy When Performing a Preablation Renal Biopsy
- (4) Peter Isfort, MD,* Tobias Penzkofer, MD,* Toshihiro Tanaka, MD,pp Philipp Bruners, MD,* Saskia Westphal, MD,§ Lieven N. Kennes, PhD,|| Thomas Schmitz-Rode, MD, Dipl-Eng,p Christiane K. Kuhl, MD,* and Andreas H. Mahnken, MD*

Efficacy of Antegrade Pyeloperfusion to Protect the Renal Pelvis in Kidney Microwave Ablation Using an In Vivo Swine Model

- (5) Valdair F. Muglia¹, Adilson Prando²
Renal cell carcinoma: histological classification and correlation with imaging findings
- (6) Ely R. Felker¹, Stephanie A. Lee-Felker, Lousine Alpern, David Lu, Steven S. Raman
Efficacy of Imaging-Guided Percutaneous Radiofrequency Ablation for the Treatment of Biopsy-Proven Malignant Cystic Renal Masses

Irreversible Electroporation of Liver Tumors with High Frequency Jet Ventilation and Real Time US-CT Image Fusion/Virtual Needle-Track Guiding. A Single Center Experience.

Authors: A. Michos¹, V. Grozman¹, T. Josephson²;

¹ Karolinska Hospital, Karolinska Institute, Stockholm - Sweden/SE

² Danderyd Hospital, Karolinska Institute, Stockholm - Sweden/SE

Purpose

Irreversible electroporation (IRE) is a non-thermal ablation technique based on high-voltage electrical pulses which can change the membrane properties provoking cell death.

In this study we aimed to present the short-term results as well as to assess the efficacy and safety of the Real Time US-CT Image Fusion/Virtual Needle-Track guided (US-CTfusion/VNT) percutaneous IRE ablation of perivascular malignant liver lesions.

Methods and Materials

We have retrospectively reviewed all the patients who were treated with US-CTfusion/VNT guided IRE at Danderyd hospital for liver tumors between March 2014 and November 2015. Our study includes 12 patients (6 males and 6 females) aged from 51 to 81 years. In total IRE ablation of 12 lesions was performed in patients not eligible for surgery or thermoablative treatment with lesions adjacent large vessels or bile ducts. Diagnosis included primary liver tumor (n= 6) and metastatic disease (n=6). The diameter of the lesions ranged between 1.0 and 3.0 cm.

General anesthesia with deep muscle paralysis and High Frequency Jet Ventilation (HFJV) was used in all cases. A preablation CT scan with intravenous contrast was obtained after the patient had been intubated. The electrode needles were placed under US-CTfusion/VNT guidance. The number of needle electrodes used ranged between 1 and 7. All patients have been followed at least for 6 months.

Results

Per-operative mortality and mortality at 6 months was 0%.

In 3 patients post-ablation complication were reported. In 1 patient complication occurred in the form of intrahepatic/subcapsular bleeding after treatment of a 15 mm HCC in segment 5. In 2 patients complication occurred in the form of post-ablative thrombosis within a portal pedicle.

Of 12 tumors, 3 (25%) required retreatment because of incomplete ablation. Recurrence free survival at 6 months was 58,3%.

Conclusion

Irreversible Electroporation of Liver Tumors with High Frequency Jet Ventilation and Real Time US-CT Image Fusion/Virtual Needle-Track Guiding is an effective and safe method for treatment of liver tumors not eligible for surgery or thermoablative treatment.

References

(1) Robert CG Martin, Prejesh Philips, Susan Ellis, David Hayes and Sandeep Bagla
Irreversible electroporation of unresectable soft tissue tumors with vascular invasion: effective palliation

(2) H. J. Scheffer & K. Nielsen & A. A. J. M. van Tilborg & J. M. Vieveen & R. A. Bouwman & G. Kazemier & H. W. M. Niessen & S. Meijer & C. van Kuijk & M. P. van den Tol & M. R. Meijerink
Ablation of colorectal liver metastases by irreversible electroporation: results of the COLDFIRE-I ablate-and-resect study

(3) Marco Dollinger, René Müller-Wille, Florian Zeman, Michael Haimerl, Christoph Niessen, Lukas P. Beyer,
Sven A. Lang, Andreas Teufel, Christian Stroszczyński, Philipp Wiggermann

Irreversible Electroporation of Malignant Hepatic Tumors - Alterations in Venous Structures at Subacute Follow-Up and Evolution at Mid-Term Follow-Up

(4) P. Sánchez-Velázquez¹, Q. Castellví², A. Villanueva³, R. Quesada¹, C. Pañella¹, M. Cáceres¹, D. Dorcaratto¹, A. Andaluz⁴, X. Moll⁴, M. Trujillo⁵, J. M. Burdío⁶, E. Berjano⁵, L. Grande¹, A. Ivorra² & F. Burdío¹

Irreversible electroporation of the liver: is there a safe limit to the ablation volume?

Microwave Ablation of Liver Tumors with Real Time US-CT/MRI Image Fusion/Virtual Needle-Track Guiding. A Single Center Experience.

Authors: A. Michos¹, V. Grozman¹, T. Josephson²;

¹Karolinska Hospital, Karolinska Institute, Stockholm-Sweden/SE

²Danderyd Hospital, Karolinska Institute, Stockholm-Sweden/SE,

Purpose

In this study we aimed to present the short-term results as well as to assess the efficacy and safety of the Real Time US-CT/MRI Image Fusion/Virtual Needle-Track guided (US-CT/MRI fusion/VNT) percutaneous Microwave Ablation (MWA) of Liver Tumors.

Methods and Materials

We have retrospectively reviewed all the patients who were treated with US-CT/MRI fusion/VNT guided percutaneous MWA at Danderyd hospital for liver tumors between March 2014 and April 2015. Our study includes 75 patients (57 males and 18 females) aged from 30 to 90 years, treated for liver tumor between March 2014 and April 2015. After induction of general anesthesia, microwave ablation was performed under US-CT/MRI fusion/VNT guidance using a 2,45 GHz microwave ablation system. All patients have been followed at least for 12 months.

Results

In total ablation of 190 lesions was performed. Number of lesions per patient varied between 1 and 15. The diameter of the lesions ranged between 0.5 and 7.0 cm. Diagnosis included primary liver tumor (n= 38) and metastatic disease (n=37). Per-operative mortality was 0%, mortality at 6 months was 8% (n=6) and mortality at 12 months was 17.3% (n=13) due to generalized spread of cancer. In 1 patient (1.3%) major complication were reported occurred in the form of bleeding after treatment of a 30 mm HCC in segment 6. This complication was successfully treated with embolization. Of 190 tumors, 10 (5,3 %) required retreatment because of incomplete ablation. All residual tumors were successfully ablated in an additional session of MWA.

Recurrence free survival at 3, 6, and 12 months was 86.4%, 76.8%, and 71%.

Conclusion

Real Time US-CT/MRI Image Fusion/Virtual Needle-Track guided percutaneous microwave ablation is an effective and safe method for treatment of liver tumors that can be repeated several times. In selected patients MWA could be considered as a first-choice method for the treatment of hepatic tumors.

References

- (1) Giovanni Mauri • Luca Cova • Stefano De Beni • Tiziana Ierace • Tania Tondolo • Anna Cerri • S. Nahum Goldberg • Luigi Solbiati
Real-Time US-CT/MRI Image Fusion for Guidance of Thermal Ablation of Liver Tumors Undetectable with US: Results in 295 Cases
- (2) Wei Yang, Kun Yan, S Nahum Goldberg, Muneeb Ahmed, Jung-Chieh Lee, Wei Wu, Zhong-Yi Zhang, Song Wang, Min-Hua Chen
Ten-year survival of hepatocellular carcinoma patients undergoing radiofrequency ablation as a first-line treatment
- (3) U. Leung, D. Kuk, M. I. D'Angelica, T. P. Kingham, P. J. Allen, R. P. DeMatteo, W. R. Jarnagin and Y. Fong
Long-term outcomes following microwave ablation for liver malignancies

Efficacy of a vertebral augmentation system used for bone remodelling as treatment of vertebral fractures

S Marcia¹, E Piras¹, A Spinelli¹, S Marini¹, L Saba²

¹ [Department of Radiology](#), PO SS Trinità, Cagliari, IT

² [Department of Radiology](#), University of Cagliari, Cagliari, IT

INTRODUCTION

The aim of this study was to evaluate the effectiveness and safety of a bone remodelling system in the treatment of painful vertebral compressive fractures (VCFs).

METHODS

Thirty consecutive patients with painful vertebral compressive fractures underwent the bone remodelling (Tektona[®], SpineArt, CH) procedure. Patients had been previously evaluated by clinical examination and X-ray, CTms and MRI-T2wSTIR. All the procedures were executed with local anaesthesia and a bilateral approach under digital fluoroscopic guidance. In total, 37 vertebrae were treated. Clinical evaluation and assessment of pain using a 11-point visual analogue scale (VAS, 0–10) were performed at baseline, immediately after the procedure and at 6-month; disability and health status by means of ODI and SF36 were also evaluated. Vertebral height (VH) restoration by vertebral body volume (VBV) calculation with CTms was assessed before (pre-) and immediately after the procedure (post), as well as vertebral heights (anterior, middle and posterior VHs) and the local (LK) and regional (RK) kyphosis angles.

RESULTS

We obtained a progressive reduction of the pain in all patients (VAS pre: 7.6±1.6, post: 2.8±1.1, 6M: 2.1±1.4), improvement of functions (ODI pre: 55.5±18.7, 6M: 22.3±8.4) as well an average improvement of 15% in SF-36 Physical Health score (PHs pre: 40.6±7.9%, 6M: 55.6±6.6%).

We observed a good height restoration (middle VH pre: 13.64±3.3mm, post: 15.25±2.5mm, difference: 1.61 mm), a good increase in the volume of the vertebral bodies (VBV pre: 21.61±2.3cm³, post: 23.08±2.4cm³, difference: 1.47 cm³).

The correction of local and regional kyphosis have been checked immediate postoperatively and the kyphosis correction was confirmed, as follows: an average LK decrease of 2.23° and an average RK decrease of 5.14°. No major complications arose.

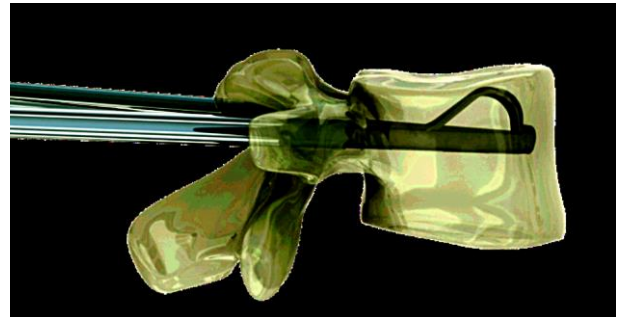


Fig. 1: Tektona[®] system, used for the treatment of vertebral compression fractures (VCFs).



Fig. 2: Device opening, for vertebral height restoration.

DISCUSSION & CONCLUSIONS: From our study, the use of bone remodelling system was found to be safe and effective in the treatment of painful vertebral fractures, providing pain relief and anatomical restoration.

REFERENCES:

- ¹ Muto M, Marcia S, Guarnieri G, Pereira V. (2015) *Assisted techniques for vertebral cementoplasty: why should we do it?* Eur J Radiol. **84**(5):783-8
- ² Schupfner R, Stoevelaar HJ, Blattert T, Fagan D, Fransen P, Marcia S, Schils F, Siddiqi MA, Anselmetti GC. (2016) *Treatment of Osteoporotic Vertebral Compression Fractures: Applicability of Appropriateness Criteria in Clinical Practice.* Pain Physician. **19**(1): E113-20
- ³ Anselmetti GC, Marcia S, Saba L, Muto M, Bonaldi G, Carpeggiani P, Marini S, Manca A, Masala S. (2012) Percutaneous vertebroplasty: multi-centric results from EVEREST experience in large cohort of patients. Eur J Radiol. **81**(12):4083-6. doi: 10.1016/j.ejrad.2012.07.005. Epub 2012 Aug 16

Novel hydrogel is effective in restoring the mechanical property of osteoporotic bone with hole defects

Lei Xu¹, Chen-Tian Li¹, F.K.L. Leung, W.W. Lu¹

¹Department of Orthopaedics and Traumatology, Li Ka Shing Faculty of Medicine, the University of Hong Kong, Hong Kong SAR, China

INTRODUCTION: Although PMMA has been widely used for the management of vertebral osteoporotic fractures in vertebroplasty and kyphoplasty, some elementary complications, including low adhesiveness and leakage after injection, and secondary fracture of adjacent vertebrae, were also reported¹. Therefore, we recently designed a novel hydrogel (unpublished) to replace PMMA and avoid its shortcomings. Before further modification to the material was made, testing of the material was conducted using a novel animal vertebral model imitating osteoporotic bone to investigate the influence of the hydrogel on the mechanical property of vertebrae, and preliminary data was collected for the further improvement of our novel hydrogel.

METHODS: A total of 54 porcine lumbar vertebrae were bought from the market. Vertebrae were isolated separately and the flesh was carefully removed from the bone. A 3-mm drill bit was used to make 48 holes in each vertebral bone either parallel to the longitudinal (Group L) or transverse (Group T) axis. The depth of the hole was equal to approximately half the length of the vertebral bone. Both groups L and T were equally allocated into four subgroups (6 samples each) called LA, LB, LC, LD and TA, TB, TC, TD, and the holes in each subgroup were filled with various novel hydrogel A, B, and C, respectively. The strengths of hydrogel A, B, and C were gradually increased (A<B<C). The holes in the LD and TD subgroups were filled with nothing and served as internal control groups. The remaining 10 samples were left intact and no holes were made, which acted as the external control group. After injection of the different materials, all the samples were mounted in epoxy rubber and fixed on the platform of an MTS machine. The stiffness and failure load of each sample were recorded.

RESULTS: The MTS tests indicated that the averages of both stiffness and failure load of the samples in the external control group were higher than the samples in the subgroups (A, B, and C),

which were higher than the internal control groups (D). The averages of both stiffness and failure load of each subgroup L were lower than the corresponding samples in subgroup T. However, the average stiffness and failure loads of each subgroup did not correspond to the strength of materials filled in the holes. (Fig. 1)

DISCUSSION & CONCLUSIONS: Osteoporotic bone is characterized by disorganized ultrastructural voids, which tend to decline the mechanical property of bone and therefore induces fracture. Our study revealed that the injection of hydrogel can significantly improve the mechanical property of osteoporotic vertebrae. More importantly, our novel hydrogel can achieve a similar mechanical restoring effect as other materials such as PMMA. Additionally, we demonstrated that longitudinal holes can cause more damage than transverse holes on the strength of bone.

REFERENCES: ¹Lieberman IH, Togawa D, Kayanja MM. Vertebroplasty and kyphoplasty: filler materials. *The Spine Journal*. 2005;5(6): S305-S16.

ACKNOWLEDGMENTS: This work was supported by grants from the GRF of Research Grant Council of Hong Kong.

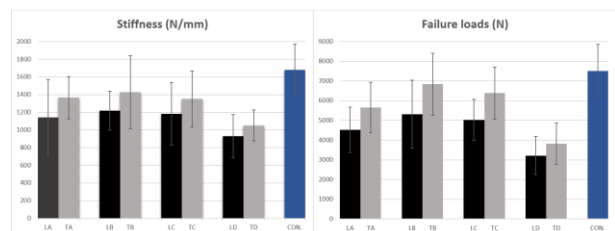


Fig.1: The MTS results measured on the vertebrae in different groups

A single ultrasound-guided PRP (platelet-rich plasma) infiltration in chronic plantar fasciitis: a double-blinded randomized comparative clinical study

M-A Malahias^{1,2}, M-K Kasetta¹, E Chronopoulos¹, V S. Nikolaou¹, A P. Efstathiou¹, G C. Babis¹

¹*2nd Orthopaedic Department, National and Kapodistrian University of Athens Medical School, Athens, Greece*

²*International Centre for Hip, Knee and Foot Surgery, Sport Traumatology, ATOS Klinik, Heidelberg, Germany*

INTRODUCTION: To clinically evaluate the effectiveness of a single ultrasound-guided (U/S) PRP injection in patients with chronic plantar fasciitis. For the evaluation of this technique we conducted a double-blinded randomized clinical comparative study in which a second group of patients was included, who treated with a single ultrasound-guided corticosteroid injection.

METHODS: This double-blinded, randomized, prospective, comparative, single centre, clinical trial enrolled 42 patients with clinical and sonographic evidence of chronic (> 6 months) fasciitis, non-responsive to analgesics, NSAID'S, stretching and ice therapy. Our patients were selected by a nurse picking their name from a sealed closed envelope. The physician who performed the injection was unaware of the injected content (we achieved this through the coverage of the syringe by the same nurse who opened the envelope). Group A (21 patients) underwent a single ultrasound-guided PRP infiltration of the swollen thickened part of the proximal plantar fascia. In addition, group B (21 patients) received a betamethasone-lidocaine ultrasound-guided simple injection at the origin of the plantar fascia. After three days resting, patients of both groups were encouraged to resume to normal activities following a set schedule of stretching exercises. The follow-up -including visual analogue scale (VAS 0-100mm), the time for taking postoperative pain killers, return to normal activities (including work) and possible complications- was set at 2, 4 and 12 weeks by another physician, blinded to the treatment of the individuals. After the initial power analysis, we planned to deploy SPSS, t-test, chi square test ($p < 0.05$) to -statistically- analyse our findings.

RESULTS: Two patients were lost during the follow-up period (one from each group). Mean VAS scores in group A were 61 mm preoperatively and, 38 mm, 50 mm and 51 mm after 2, 4, and 12 weeks postoperatively, while in group B were 63 preoperatively and, 28, 37 and 46 postoperatively.

Mean time in group A patients were 5.4 days for taking pain killers and 4.1 days for returning to normal activities and in group B were 6.9 days for taking pain killers and 1.7 days for returning to normal activities. Two patients of the group A experienced worsening of their clinical symptoms within the first 4 weeks, but at the final follow-up (12 weeks) of our study they were found having the same VAS as their preoperative one. We did not notice any significant postoperative complication in both groups.

CONCLUSIONS: A single ultrasound-guided PRP injection seems to have slightly inferior results (statistically non-significant) in comparison to a single ultrasound-guided corticosteroid injection in patients with chronic plantar fasciitis. Therefore, it qualifies as a refutable technique for the treatment of the afore-mentioned category of patients.

Differential effect of subcytotoxic exposure of folic acid functionalized nanoparticles on migration of normal kidney and cancerous breast cells

E Flampouri¹, D Theodosi-Palimeri¹, S Kintzios¹

¹ *Laboratory of Cell Technology, Department of Biotechnology, School of Food, Biotechnology and Development (TBA), Agricultural University of Athens, Greece.*

INTRODUCTION: Continuous advances in synthesis and functionalization of gold nanoparticles (AuNPs), combined with their biocompatibility, chemo-physical stability and optical tunable characteristics, have led to an immense expansion of their biomedical applications. Folic acid (FA) mediated cancer cell targeting, which is based on the overexpression of FA receptors by certain cancer cells, is one of the most important methods for active cancer targeting. Intravenous injection of nanosystems for cancer theranostics, is gaining increased attention because they bypass the gastrointestinal tract, resulting in increased efficiency. The aim of the present study was to assess the different responses in viability and migration of normal and cancer cells to FA functionalized NPs.

METHODS: AuNPs were synthesized by the NaBH₄ reduction method. NPs were functionalized through GSH capping and DCC/NHS reaction to activate the carboxyl groups of GSH, which then reacted with the free amino groups present in FA, resulting in the formation of FA-conjugated NPs (AuNPs-FA). Viability was assayed through the MTT assay. Cell migration was examined using the *in vitro* scratch assay, analyzed using image analysis software (Image J, 1.29) and extrapolated to % of control.

RESULTS: Both AuNPs and AuNPs-FA exhibited a peak around 526nm (Fig.1A) that corresponds to the characteristic SPR band of AuNPs in the visible region.

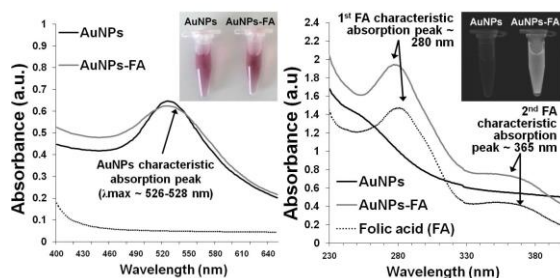


Fig. 1: UV-Vis absorption spectra of AuNPs and FA functionalized AuNPs. Inserts: corresponding optical and UV illuminated images.

In the UV spectra (Fig.1B), the absorption max of AuNPs-FA at 280 nm and 365 nm, confirmed their covalent attachment with FA. Biocompatibility of the prepared NPs was established for both cell types through the MTT assay (Fig.2A). AuNPs-FA 24h exposure affected significantly ($p=0.025$) the motility of MCF-7 cells as indicated by the *in vitro* scratch assay (Fig. 2B). Scratched areas of Vero cells were covered concurrently, while AuNPs-FA delayed the FA receptor-positive MCF-7 migration compared to the control (Fig. 2C, 2D).

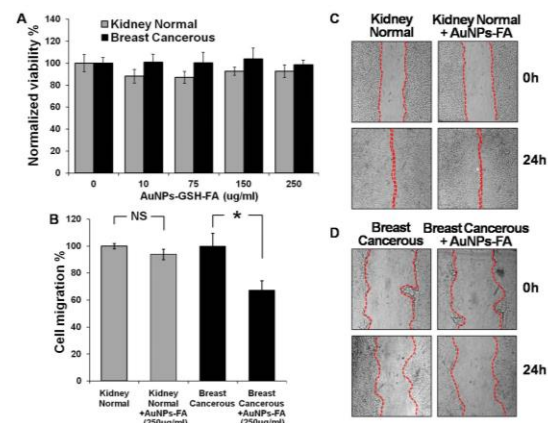


Fig. 2: Effect AuNPs-FA on cell viability (A) by the MTT assay. Involvement of AuNPs-FA on cell migration through an *in vitro* scratch assay (B). Representative images of (C) normal kidney (Vero) and breast cancer (MCF-7) cells (D). Controls are set to 100%. Values are expressed as mean \pm SD, * $p \leq 0.05$ in student's *t*-test.

DISCUSSION & CONCLUSIONS: Our results suggest that sub-cytotoxic exposure of folic acid functionalized nanoparticles exhibited a differential effect on cell motility of normal kidney and cancerous breast cells, suggesting that the FA moiety and AuNPs could be implicated in the migration process.

REFERENCES: ¹ Z. Zhang et al (2010) *Bioorg Med Chem*, 18(15), 5528-5534.

ACKNOWLEDGEMENTS: The authors thank Dr. Bouziotis and Dr. Paravatou from N.C.S.R "Demokritos" for providing the MCF-7 cell line.

Bioactivity and effect of hyperbranched poly(ethyleneimine)-based nanoparticles on plant pathogenic fungi

M Lykogianni¹, EA Papadopoulou¹, Z Sideratou², D Tsiourvas², KA Aliferis¹

¹ *Laboratory of Pesticide Science, Agricultural University of Athens, Greece.*

² *Institute of Nanoscience and Nanotechnology, NCSR Demokritos, Athens, Greece.*

Emails: z.sideratou@inn.demokritos.gr, konstantinos.aliferis@aua.gr

INTRODUCTION: Numerous applications of nanotechnology have been developed in almost any scientific discipline. However, its application in the field of Agricultural and Environmental sciences is still in its infancy. Currently, there is increasing interest of the agrochemical sector in the exploitation of nanomaterials in crop protection. However, although the research related to nanodrugs for human has significantly advanced, that on the development of nanopesticides is still in initial stages. The latest developments in agrochemicals' R&D involves polymer-based formulation and the use of nanometals and nanoemulsions [1]. Such approach could provide solutions to problems and challenges that the crop protection sector is facing *i.e.*, resistance of plant pathogens to pesticides, lack of pesticides for several plant diseases, development of pesticides with improved efficacy and toxicological profiles [2].

Within this context, we have undertaken the task to screen the bioactivity of hyperbranched poly(ethyleneimine) (HPEI) with Mw 25 KDa as well as its functionalized derivatives with guanidinium or quaternary ammonium groups, against various important plant pathogens and mine their effects on their metabolism performing metabolomics.

METHODS: HPEI was functionalized with guanidinium or quaternary ammonium groups, affording GPEI and QPEI, respectively. The resulting polymers were spectroscopically characterized by FTIR, ¹H NMR and inverse gated ¹³C NMR in order to determine the degree of substitution, which was found to be 50% relative to the primary amino groups of HPEI.

The effect of different concentrations of PEI-based nanoparticles on the pathogens' radial growth was estimated and EC₅₀ values were calculated. The effect of nanoparticles on fungal metabolism was studied applying metabolomics. Gas chromatography/ mass spectrometry was employed and the discovery of trends and corresponding metabolite-biomarkers was based on multivariate analysis using the SIMCA-P+ software [3,4].

RESULTS: The PEI-based nanoparticles had a variable effect on the different fungal species being tested (*Fig. 1*). The EC₅₀ values fluctuated from 200 µg mL⁻¹ to more than 500 µg mL⁻¹.

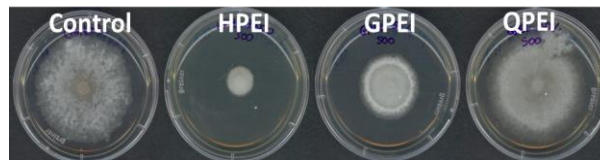


Fig. 1: Radial growth of 14-day-old Phytophthora infestans cultures on Potato Dextrose Media (PDA) amended with 500 µg mL⁻¹ of HPEI, 500 µg mL⁻¹ GPEI, 500 µg mL⁻¹ QPEI, or equal volume of sterilized water (Control).

Accordingly, differences were recorded in the metabolome of the fungi in response to treatments.

DISCUSSION & CONCLUSIONS: Here, we have studied the potential of HPEI, GPEI, and QPEI to be used as pesticides *per se* or as nanocarriers in nanopesticide formulation. The polymers being studied exhibited variable fungitoxicity, which was closely associated to their structure. The changes in the metabolome of fungi confirmed the toxicity at the cellular level with multiple biosynthetic pathways being up- or down-regulated in response to the treatments. Experiments are in progress towards the elucidation of the mode(s)-of-action of the nanoparticles and their further exploitation in the development of next-generation pesticides [3,4].

REFERENCES:

- ¹M. Kah, S.B., K. Tiede & T. Hofmann (2013) Nanopesticides: State of Knowledge, Environmental Fate, and Exposure Modeling in *Critical Reviews in Environmental Science and Technology*, 43: 1823-1867.
- ²R.S. Kookana, et al. (2014) Nanopesticides: Guiding Principles for Regulatory Evaluation of Environmental Risks in *Journal of Agricultural and Food Chemistry*, 62: 4227-4240.
- ³K.A. Aliferis, and M. Chrysayi-Tokousbalides (2011) Metabolomics in pesticide research and development: review and future perspectives in *Metabolomics* 7: 35-53.
- ⁴K.A. Aliferis, and S. Jabaji (2011) Metabolomics—A robust bioanalytical approach for the discovery of the modes-of-action of pesticides: A review in *Pesticide Biochemistry and Physiology*, 100: 105-117.

Morphology and biophysical properties of pH-responsive nanotechnological chimeric/mixed liposomal platforms as innovative drug delivery systems

N Naziris¹, N Pippa^{1,2}, V Chrysostomou², S Pispas², C Demetzos^{1,*}, M Libera³, B Trzebicka³

¹ [Department of Pharmaceutical Technology](#), Faculty of Pharmacy, National and Kapodistrian University of Athens, Greece. ² [Theoretical and Physical Chemistry Institute](#), National Hellenic Research Foundation, Greece. ³ [Centre of Polymer and Carbon Materials](#), Polish Academy of Sciences, Poland

INTRODUCTION: Nanocarriers with stimuli-responsive properties are expected to offer much in the fields of Therapeutics and Theranostics¹. Chimeric/mixed nanosystems of lipids and polymers create new innovative platforms for drug delivery, providing the benefits of both classes². The aim of this work was to design and build chimeric/mixed liposomes, consisting of the phospholipid L- α -phosphatidylcholine, hydrogenated (Soy) (HSPC) and the pH-sensitive amphiphilic diblock copolymer poly(2-(dimethylamino)ethyl methacrylate)-*b*-poly(lauryl methacrylate) (PDMAEMA-*b*-PLMA).

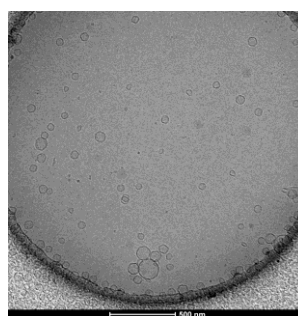
METHODS: Liposomes comprising the phospholipid and the copolymer were constructed, with the thin-film hydration method. Two distinct copolymers PDMAEMA-*b*-PLMA 1 and 2 of different composition were synthesized, through RAFT polymerization and both were incorporated into the liposomal systems in various molar ratios. The size, size distribution, ζ -potential and radius of gyration of the developed structures were evaluated by dynamic, electrophoretic and static light scattering (DLS, ELS and SLS) and their pH-responsive properties we verified through exposure to acidic conditions (pH = 4.5). In addition, the morphology of the systems was studied by atomic force microscopy (AFM) and cryogenic transmission electron microscopy (cryo-TEM) and finally, *in vitro* screening was carried out, to ascertain their toxicity.

RESULTS: Vesicular nanostructures were developed, with chimeric/mixed types having larger diameters than the conventional HSPC one, while their polydispersity depended on the membrane polymer content. A few representative examples are provided in *Table 1*. Acidic protocol led to alteration of the chimeric/mixed liposomes' physicochemical properties, indicating the pH-responsive behavior of these preparations. The imaging techniques revealed the membrane biophysics and morphology of the chimeric/mixed nanosystems, as well as the existence of non-

vesicular structures, whose number was proportional to the concentration and hydrophobicity of the copolymer. In *Fig. 1*, a chimeric/mixed liposomal system is presented. Finally, the *in vitro* toxicity was found higher for systems of high polymer content.

*Table 1. Size and distribution of conventional and HSPC:PDMAEMA-*b*-PLMA 1 and 2 9:0.1 liposomes.*

| System | Hydrodynamic Diameter (nm) | Polydispersity |
|-----------------|----------------------------|----------------|
| lipid | 104.0 | 0.384 |
| lipid:polymer 1 | 134.0 | 0.304 |
| lipid:polymer 2 | 124.6 | 0.384 |



*Fig. 1: Cryo-TEM image of HSPC:PDMAEMA-*b*-PLMA 2 9:0.1 liposomes.*

DISCUSSION & CONCLUSIONS: In a previous study, the pH-responsive drug release from polymer-grafted liposomes was evident³. The physicochemical and morphology studies provide useful information for the biophysical behaviour of pH-responsive chimeric/mixed nanocarriers, indicating that they are promising advanced drug delivery nanosystems (aDnSs).

REFERENCES: ¹ N. Naziris, N. Pippa, S. Pispas, et al (2016) *Curr Nanomed* **6**:166-85. ² M. Schulz and W.H. Binder (2016) *Macromol Rapid Commun* **36**:2031-41. ³ N. Naziris, N. Pippa, A. Meristoudi, et al (2016) *J Liposome Res* [Epub ahead of print].

Randomized evaluation of bone ingrowth after Anterior Lumbar Interbody Fusion (ALIF) with a new osteoinductive bone graft material (NanoBone / putty)

I Papachristos, M Rickert, M Rauschmann

Orthopaedic University Hospital Friedrichsheim in Frankfurt am Main, Germany

INTRODUCTION: In our days spondylodesis is a standard method for the treatment of degenerative diseases of the spine. To achieve a permanent therapeutic success a fusion of the stabilized vertebrae is needed. The autologous bone graft is considered the "gold standard" because it is the only material with all three bone augmentation important characteristics, osteogenesis, osteoinduction and osteoconduction. However, the limited availability and the donor morbidity have led to research for alternative bone grafts und substitutes.

NanoBone, a new bone substitute material with osteoinductive properties, is produced synthetically and is a mixture of nanocrystalline hydroxyapatite, and a silica gel matrix. It has a high porosity and biocompatibility. Within a few days after implantation, the silica gel matrix is replaced by the body's own BMP and formed an organic matrix. Due to the synthetic preparation immunogenic responses, as well as the transmission of pathogens are excluded.

METHODS: The study was prospectively randomized. There were treated 50 segments in 40 patients, 20 patients and 25 segments in each study group (NanoBone-, control group). The mean age was 62.2 years in total. The ratio between men and women was 3:1. The fused segments were L4/5 and L5/S1. Patients completed preoperative questionnaires (VAS, EQ-5D, ODI). The follow-ups were carried out 3, 6 and 12 months postoperatively. The questionnaires were completed again at each follow-up. The radiological follow-up was conducted for 3 months postoperatively by means of CT scan of the region of interest, 6 and 12 months postoperatively using radiographs in 2 planes. The fusion grade was assessed using the Bridwell classification. In case of delayed fusion, lateral radiographs in flexion and extension were performed in addition 12 months postoperatively, to assess the stability. A difference of the segmental motion between flexion and extension more than 5 ° was classified as instability.

RESULTS: The complication rate was 15% (n = 6/40). These were wound healing disorders like

superficial wound infections (n = 1/40), dislocation of implants (n = 2/40), pedicle fracture (n = 1/40) and non-implant-associated stenosis of the intervertebral foramen (n = 2/40). The fusion rate was 3, 6 and 12 months postoperatively in the NanoBone group at 92%, 96% and 96%, in the control group at 80%, 88% and 88%. A statistical significance could not be documented ($p > 0.05$) between the two study groups. Those in the 12-month follow-up evaluated as non-fusion were asymptomatic all the time and did not have to be revised. The clinical parameters (VAS, ODI, EQ-5D) showed a significant improvement in both groups. However, in direct comparison between the groups, there were no statistically significant differences.

DISCUSSION & CONCLUSIONS: NanoBone was showing good results in terms of osseointegration, as well as patient satisfaction. Higher fusion rates could be documented compared to homologous spongiosa. In summary, due to its high availability, high biocompatibility and sterility, NanoBone represents a reliable and promising bone substitute material. However, It should be mentioned that nanobone is only used in ventral fusion procedures and thus is associated with increased access morbidity. Points of criticism of this work are the low number of cases and the different surgical procedures of instrumentation.

REFERENCES: Abshagen K. (2009) *In vivo analysis of biocompatibility and vascularization of the synthetic bone grafting substitute NanoBone.* J Biomed Mater Res A. Nov; 91(2):557-66. Adam (2014) *In vivo and in vitro investigations of a nanostructured coating material – a preclinical study.* Int J Nanomedicine.; 9: 975–98. Götz W. (2010) *A preliminary study in osteoinduction by a nano-crystalline hydroxyapatite in the mini pig.* Folia Histochem Cytobiol.; 48(4): 589-596

ACKNOWLEDGEMENTS: This template was modified with kind permission from eCM Journal.

Temperature-sensitization of HSPC liposomes by use of thermosensitive homopolymer PNIPAM

A Tzani¹, N Naziris¹, A Meristoudi², S Pispas², C Demetzos^{1,*}

¹ [Department of Pharmaceutical Technology](#), Faculty of Pharmacy, National and Kapodistrian University of Athens, Greece. ² [Theoretical and Physical Chemistry Institute](#), National Hellenic Research Foundation, Greece.

INTRODUCTION: Thermoresponsive polymers have the ability to undergo a conformational transition when temperature change is applied¹. The aim of this study was to incorporate the thermoresponsive polymer C₁₂H₂₅-poly(N-isopropylacrylamide)-COOH in L- α -phosphatidylcholine hydrogenated (Soy) (HSPC) liposomes, at six different molar ratios and to characterize the resultant structures in biophysical and thermodynamic manner.

METHODS: At first, we developed conventional and mixed liposomal nanocarriers by thin-film hydration. The resultant multilamellar vesicles were subjected to three, 10min sonication cycles interrupted by a 10-min resting period in order to decrease their size. Then, we quantified the physicochemical characteristics and physical stability of the prepared chimeric systems, with dynamic and electrophoretic light scattering. Finally, we studied their thermotropic behavior, through measurement of thermodynamic parameters, using differential scanning calorimetry (DSC).

RESULTS: Conventional HSPC liposomes exhibited the smaller size and polydispersity than the other prepared systems. After the incorporation of C₁₂H₂₅-PNIPAM-COOH at the lowest molar ratio, a gradual increase of D_h was observed while at higher concentrations of PNIPAM, biomaterials exposed better cooperativity, leading to chimeric liposomes of smaller size. Their PDI followed the same trend with their D_h, as the percentage of C₁₂H₂₅-PNIPAM-COOH was increased. Moreover, an inversion of ζ -potential from positive to negative indicated that the polymer was successfully incorporated into the liposomal membrane. As shown in Fig.1, conventional and mixed formulations, apart from HSPC:PNIPAM-S2 (9:0,02 and 9:0,05 molar ratio), were found to retain their original physicochemical characteristics (size and size distribution) at least for the time period that they were studied (30 days). Thermodynamic findings are in line with

physicochemical results, since small amounts of polymer provoke a major perturbation in lipid bilayers, while higher molar ratios exhibited better cooperativity, altering slightly the thermotropic behavior of pure HSPC lipid bilayers.

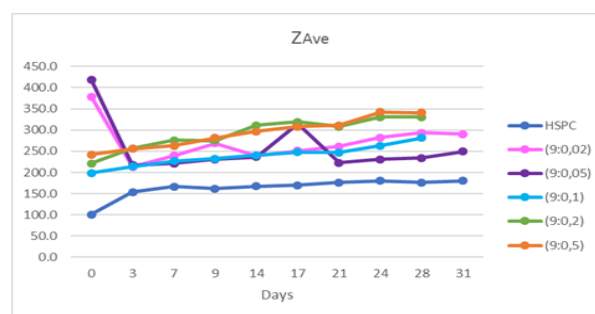


Fig. 1: Vesicle size stability of HSPC:C₁₂H₂₅-PNIPAM-COOH liposomes, during 31 days.

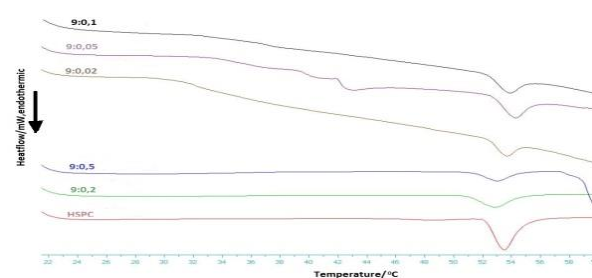


Fig. 2: DSC heating-scans of HSPC:PNIPAM lipid bilayers.

DISCUSSION & CONCLUSIONS: This study showed a physicochemical and thermotropic approach to thermosensitive chimeric liposomes. The presence of the polymeric component plays a key role in the thermal behavior and the structural rearrangement of lipid membranes. Conventional liposomes can be altered after the incorporation of temperature-responsive polymers, such as the homopolymer C₁₂H₂₅-PNIPAM-COOH, and can be utilized as advanced drug delivery nanosystems if we further entrap any pharmaceutical drug.

REFERENCES: ¹ N. Naziris, N. Pippa, S. Pispas, et al (2016) *Curr Nanomed* **6**:166-85.

Surface functionalization of titanium with strontium-substituted hydroxyapatite and miR-21 Nanocapsules to Enhance Bone Regeneration

Zhen Geng, Zhenduo Cui, Zhaoyang Li, Xubo Yuan, Shengli Zhu, Yanqin Liang, Xianjin Yang
School of Materials Science and Engineering, Tianjin University, Tianjin 300072, China.

INTRODUCTION: Titanium (Ti) and its alloys have been widely used for dental and orthopedic implants. However, Ti and its alloys are known to be bio-inert, which results in lack of active induction of bone growth and thus poor osseointegration. Therefore, direct efforts have been made to improve their bioactivity. Strontium (Sr) has been proved to possess both bone formation stimulating and anti-resorptive effect. Thus, Sr-substituted hydroxyapatite (SrHA) has attracted considerable interest. Meanwhile, our previous study showed that miRNA-21 could promote osteogenic differentiation of mesenchymal stem cells. Taken together, designing a SrHA and miRNA-21 composite coating might be an effective way to improve the osseointegration.

METHODS: Firstly, hydrothermal method was used to fabricate SrHA coating. Then, miRNA-21 nanocapsules were synthesized by in situ polymerization. After that, the miRNA-21 nanocapsules were absorbed onto the SrHA surface to fabricate the composite coating. Finally, it was evaluated by a rabbit model. Samples were implanted into the femur's metaphyses and tibial platforms of mature New Zealand White rabbits. The rabbits were anesthetized via intravenous injection of phenobarbital sodium. Four cylindrical holes (4 mm indiameter) were created on femur's metaphyses and tibial platform using surgical drills for each rabbit. Different Ti rods were inserted into the predrilled holes.

RESULTS: The size of microRNA-21 nanocapsule was about 30 nanometer. Meanwhile, the SrHA coating can be synthesized by one-step hydrothermal method and can greatly improve the hydrophilic property. In addition, the microRNA-21 nanocapsules can be loaded onto the SrHA layer to fabricate the composite coating. Histological results showed that microRNA21 could accelerate the bone remodeling process

and reduce the time of mineralization. Meanwhile, the composite coating further improved the bone remodeling and maturation. X-ray and microCT results indicated that microRNA21 and SrHA had a synergistic effect on bone growth. Immunohistochemical examination showed that microRNA21 could not only promote the expression of osteogenesis related proteins, such as RUNX2, osteocalcin(OCN) and osteopontin(OPN), but also promote the expression of osteoclastogenesis related protein, such as RANKL. The composite coating showed an improved expression of osteogenesis related proteins and a decreased expression of osteoclastogenesis related protein. The bone-implant binding force results indicated that both the SrHA and miRNA-21 nanocapsules coating could improve the bone-implant bonding strength. The Raman measurements indicated the new bone formed on the composite coating had the highest degree of mineralization.

DISCUSSION: Both the histological and SEM-EDS results showed that miR-21 accelerated the progress of bone remodeling and maturity, which was further confirmed by the immunohistochemical analysis. It was worth noting that miR-21 also promoted the osteoclastic activity. This meant that miR-21 was a microRNA with multiple and complex functions. On the one hand, miR-21 promoted the blood vessel formation and then further promoted the osteoblast proliferation and differentiation, which accelerated the new bone formation and then resulted a fast bone maturity and an excellent osseointegration. The surface modification of Ti with miRNA-21 could promote the blood vessel formation. SrHA could not only stimulate osteoblast proliferation, but also reduce the osteoclast activity. The release profile and metabolism mechanism of miRNA-21 and SrHA are different. They can accelerated synergistically

the bone remodeling and maturity process, resulting an improved implant-bone binding strength.

Long-term drug delivery depots formed by isostatic pressing

S Grudén¹, H Lennernäs², G Alderborn², N Axén¹

¹ LIDDS AB, Virdings allé 32b, SE-754 50 Uppsala, Sweden. ² Uppsala University, Department of Pharmacy, Box 580, SE-751 23 Uppsala, Sweden

INTRODUCTION: A biomaterial that is injectable, can deliver long-term slow release of drugs and is water soluble and bioresorbable has long been searched by the pharmaceutical industry^{1,2}. This study describes an injectable, two-component formulation that solidifies *in vivo* to a solid local drug depot (NanoZolid®). Due to an optimisation of the microstructure of the biomaterial, the drug release rate is controlled. Injectable depot formulations based on biomaterials for long-term drug release have numerous applications for local and focused treatment of cancers while reducing the systemic side-effects, e.g. for anti-cancer drug substances such as cytostatics, antibodies and hormones.

METHODS: The inorganic and water soluble compound calcium sulfate was used for the encapsulation of 2-hydroxyflutamide (2-HOF) in a structurally combined matrix. An injectable and *in vivo* solidifying formulation was achieved by exploring the ability of calcium sulfate to solidify by hydration, i.e. recrystallization and uptake of crystal water. Hence, a highly dense and slowly dissolving calcium sulfate structure with was achieved by combining hydration and isostatic pressing.

RESULTS: An injectable and *in vivo* solidifying drug product consisting of a combination of a porous (ordinary slow release – weeks) and a dense (long-term slow release – months) microstructure for modified drug release and an aqueous cellulose diluent is described. *In vitro*, the depot releases 2-HOF over about 6 months as the calcium sulfate is dissolved.

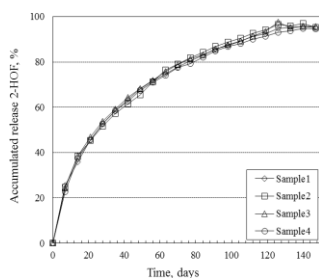


Fig. 1: *In vitro* drug release of 2-HOF over time from Liproca Depot in 0.9 % saline water.

An optimally dense microstructure was achieved from applying 400 MPa isostatic pressure for 60 minutes and wetting with near stoichiometric amounts of water.

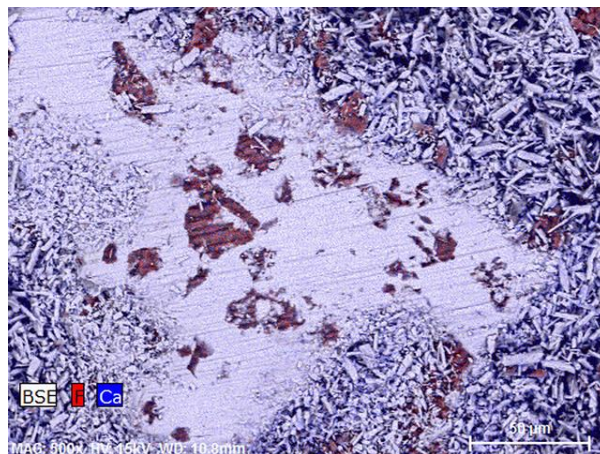


Fig. 2: Scanning electron microscopy (SEM) with energy dispersive X-ray analysis (EDX) mapping image of microstructure of Liproca Depot in solidified form, showing precipitates of 2-HOF (red) in the porous matrix as well as in the densified granules (white-grey).

DISCUSSION & CONCLUSIONS: Isostatic pressing of hydrating biocompatible inorganic materials, such as calcium sulfate, can be used to produce highly dense structures, which when size-reduced are suitable for injection through standard cannulas, and for use in formulations for long-term drug release. The process is a low temperature and chemically gentle process that can be used for a range of active substances.

REFERENCES: ¹A. Moghanjoughi, et al., A concise review on smart polymers for controlled drug release. *Drug Deliv Transl Res* 2016, 6:333-340. ²C.A. Hewes, et al., Use of cisplatin-containing biodegradable beads for treatment of cutaneous neoplasia in equidae. *J Am Vet Med Assoc* 2006, 229:1617-1622. ³E. Sjogren, et al., Pharmacokinetics of an injectable modified-release 2-hydroxy-flutamide formulation in the human prostate gland using a semiphysiologically based biopharmaceutical model. *Mol Pharm* 2014, 11:3097-3111.