

Towards Medical Diagnostics Using Hyperspectral X-ray Scatter

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ABSTRACT

This pilot study builds from a recent international, programme that showed, for the first time, the clinical value of new biomarkers to predict fracture risk in osteoporotic patients. The research indicated several potential, high impact routes towards point of care application. Our current studies take the first steps to empirical assessment and practical implementation of the technology. The initial study was performed entirely with interrogating probes applied in transmission mode i.e. bones were illuminated from one side and signals measured from the opposite side. This has some disadvantages that would be overcome were reflection mode (probe and detector on same side) employed. Further, recent advances in detector technology have resulted in the first commercially available hyperspectral X-ray imaging system that has the promise of faster, lower dose imaging. Thus this pilot study involves the creation of unique, high impact data with the potential to form the basis of a new generation of medical diagnostic instrumentation. We present the findings from our initial examination of *ex vivo* tissues. A systematic series of conventional reflection mode experiments were performed in which bone specimens were examined through increasing thicknesses of overlaying muscle/fat/skin. This data provided an indication of parameter precision associated with the biomarkers that were used in subsequent phases. Further, we have applied a new imaging modality (focal construct technology, FCT) to these geometries. FCT is a new approach to data collection using hollow X-ray beams and this was applied in reflection mode to measure bone mineral signatures. This had not previously been attempted and required some initial modelling to ensure correct topologies of the hollow beams. The speed/dose advantage of FCT was evaluated.

Keywords: Focal Construct Technology (FCT), medical diagnostic, biomarkers, osteoporosis

1 INTRODUCTION

Osteoporosis affects approximately 200 million women around the world. In 2014, a report by the National Osteoporosis Foundation (NOF) revealed that this debilitating bone disease affected 10.2 million U.S. adults

aged 50 and over, whilst a further 43.4 million individuals have low bone mass¹. With an ever increasing aging population these numbers will only increase. Consequently, the number of fractures are anticipated to increase proportionally. This will not only have a huge economic burden on the health system but will be detrimental to millions of people's lives. Osteoporotic fractures often occur in the hip, wrist and vertebrae; although studies have shown hip fractures have the greatest detrimental effect on an individual². Hip fractures result in a significant loss of independence, and sufferers are unable to live without support as they cannot walk unaided or perform many of their daily activities. Worryingly, hip fractures are often associated with increased mortality^{3,4}, a statistic which is confounded by the asymptomatic nature of osteoporosis.

Osteoporosis is currently assessed according to an individual's bone mineral density (BMD)⁵. With a decrease in BMD, the risk of fracture is significantly increased⁶. Currently the gold standard for measuring BMD is through the use of dual energy X-ray absorption (DEXA). DEXA however is not without limitations and is arguably a poor predictor of fracture, with a study carried out by Wainwright *et al.* showing that 54% of new hip fractures occurred in women who did not have osteoporosis as determined by their BMD⁷ and data from the National Osteoporosis Risk Assessment, showed that 82% of post-menopausal women with fractures had bone of 'normal' BMD⁸. Arguably, the limits associated with DEXA to predict an individual patient's fracture risk is because BMD does not measure the multiple material factors that contribute to bone strength⁹. If one was to consider the construction of a bridge, the engineer would not only take into account the architecture but also the material from which the construction should be made from, maybe this is the same view point which should be taken for biomaterials such as bone?

In recent years, a number of studies have shown micro-architectural properties of bone could potentially offer a superior way to differentiate between diseased bone (osteoporotic and osteoarthritic) and healthy controls¹⁰⁻¹². Although undoubtedly promising, these studies only account for the microarchitecture of the bone and do not also account for bone chemistry. A much smaller number of studies have concentrated on the bone chemistry of diseased bone, with studies often providing contradictory results and conclusions¹³⁻¹⁹. This is potentially due to small sample

numbers ($n \leq 10$)^{16,20,21}, different bone types (e.g. femur, iliac crest, vertebrae) utilized and the use of ovariectomised animal models^{22,23}. Many of these studies have demonstrated that 'bone quality' (including characteristics such as collagen/mineral ratio, collagen integrity, mineral crystallite size, microstrain) has a marked effect on a bone's mechanical properties and is probably the 'missing' information required to produce diagnostically predictive models of fracture. In 2016, Dicken *et al* highlighted the potential use of X-ray diffraction signatures to differentiate between osteoporotic and 'normal' bone²⁴. This study, for the first time carried out principle component analysis (PCA) on X-ray diffraction signatures from both diseased and 'normal' bone mineral tissue ($n = 108$), in order to develop a classification model to discern each condition. The results suggested a sensitivity and specificity of 93% and 91%, respectively for the model. This study highlighted the potential use of XRD as an *in vivo* diagnostic tool for fracture risk predication.

To further develop XRD as a diagnostic tool, there is a need to translate the laboratory *ex vivo* measurements for *in vivo* diagnostic purposes. Conventional XRD diffraction on bone, as used by Dicken *et al* and other research groups, is performed with interrogating probes applied in transmission mode i.e. bone samples were illuminated from one side and signals measured from the opposite side. This conventional way of collecting X-ray signatures may be problematic for *in vivo* analysis due to the confounding effects of the soft tissue as well as the bone itself, on the resulting diffraction signature. This issue may be overcome if reflection mode (probe and detector on same side) was employed. However, laboratory based XRD in both transmission and reflection mode is conducted with typically relatively soft X-ray energies such as $\text{CuK}\alpha$ (≈ 8 keV), in order to optimize the magnitude and angular distribution of the coherent scatter. With material such as bone, photons at these energies would have limited penetrating capability, with just 40 μm of hydroxyapatite (the mineral component of bone) sufficient to absorb $\approx 95\%$ of these photons. Potentially, pencil beam XRD used in the laboratory may not be suitable as an *in vivo* diagnostic probe due to its low signal strength and consequently high doses. Studies have demonstrated with the use of synchrotron sources that X-ray diffraction signatures from bone can be obtained at significantly greater X-ray energies of ≈ 80.7 keV^{25,26}. However, the use of a synchrotron is impractical when considering widespread implantation of a cost effective diagnostic tool. This has led to a small number of researchers investigating new approaches to obtain coherent scatter data within a laboratory setting, with these researchers developing a new modality termed focal construct technology (FCT). The principles of this alternative XRD technique can be found elsewhere^{27,28}, however primarily this technique employs an annular interrogating beam rather than the conventional pencil beam. When this beam is incident to the sample, a continuum of Debye cones is produced around a circular

footprint at characteristic Bragg angles. As these Debye cones spread away from the sample, they intersect to form relatively high intensity patterns, termed caustics, in the diffracted flux. This results in diffraction intensities which are significantly greater than conventional pencil beam XRD. This modality has previously been used to obtain X-ray diffraction patterns from bone in transmission mode²⁹. However, consideration for confounding factors which will be encountered *in vivo*, such as soft tissue (muscle/ fat/ skin) has not previously been reported.

This study will focus on investigating the X-ray signatures from bone whilst also considering *in vivo* confounders such as soft tissue. Conventional XRD as well as the new modality of FCT will be examined and assessed according to the physicochemical parameters which can be obtained from the data. Further, recent advances in detector technology have resulted in the first commercially available hyperspectral X-ray imaging system (Hexitec) that has the promise of faster, lower dose imaging. The main advantage of this pixelated detector is that it provides both angular and energy dispersive data. As part of this study, the capabilities of Hexitec for conventional XRD and FCT will be assessed, therefore taking the essential steps towards a new, high impact diagnostic tool. Ultimately, this study involves the creation of unique, high impact data with the potential to form the basis of a new generation of medical diagnostic instrumentation.

2 MATERIALS AND METHODS

2.1 Materials

Sintered aluminum oxide (Al_2O_3) (NIST standard SRM 1976) was utilized as a near – ideal polycrystalline sample exhibiting small grain size and low preferred orientation. Al_2O_3 has a well-defined use as a diffraction standard and was used to configure the correct geometry of the equipment prior to analyzing the bone sample. The bone specimen utilized for this study was bovine femur whilst diffraction phantoms to support an increasing thickness of muscle/fat were also utilized.

2.2. Methods

The X-ray source employed was a standard water cooled sealed tube (40 kV, 30 mA) with a molybdenum (Mo) target of 0.7107 Å (~ 17.5 keV) and a zirconium (Zr) filter. A brass annular collimation was employed during FCT experiments, with a diameter of 5 mm. Two detectors were utilized for this work, PIXIS and HEXITEC. The PIXIS is a Princeton Instrument 13.3. x 13.3 mm area detector with 1024 x 1024 pixels (13 x 13 μm) CCD (2D Gadox – Princeton Instrument PIXIS 1024). The HEXITEC is a Quantum instrument with each 80 x 80 pixel providing a full energy spectrum with an average energy resolution of 800 eV. The pixel size for this detector is 250 x 250 μm).

3 RESULTS AND DISCUSSION

3.1 Transmission Mode

With increasing layers of soft tissue, a decrease in the intensity of the diffraction signatures for both pencil beam and FCT modalities was observed (Figure 1). For conventional XRD (pencil beam), overlaying the bone sample with $\geq 3\text{mm}$ of soft tissue resulted in a dramatic loss in diffraction intensity. Consequently, the biomarkers measured in previous work [REF] could not be quantified from this data with the precision required. For *in vivo* measurements, not only does the confounding effect of the soft tissue need to be taken into account, but the thickness of the bone also needs to be considered. As aforementioned, for soft X-ray energies, just $40\mu\text{m}$ of hydroxyapatite is sufficient to absorb $\approx 95\%$ of the photons. Considering the confounding effects of the soft tissue as well as the bone itself, it will be difficult to obtain diffraction signatures from many of the typical fracture bone sites *in vivo*, therefore it is necessary to consider modalities in reflection mode.

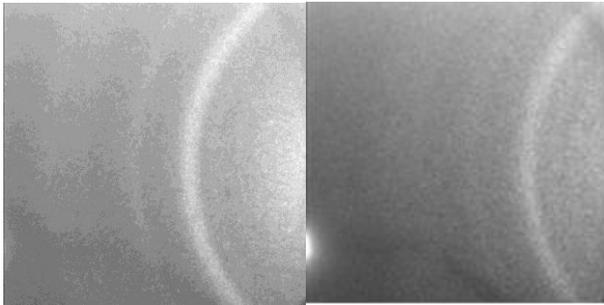


Figure 1: Pencil beam in transmission mode for bone with differing layers of soft tissue, 0 mm (left) and 6 mm (right).

3.2. Reflection Mode

Reflection mode FCT is a more complex geometry than the corresponding transmission mode and thus significant effort was directed towards simulation both for the experimental design and for interpretation of the experimental data (Figure 2). Modelling was undertaken using McXtrace in collaboration with Dr Eric Knudsen (Technical University of Denmark). The modelling environment was modified to accommodate a hollow X-ray beam, diverging source, a polycrystalline planar sample and a pixelated area detector (with variable area). A highlight of this modelling was the realization and demonstration that a specific incident beam footprint could be contrived to form a focal point at a detector wherein all rays possessed an identical scatter vector. This is critical in reflection mode in order to work as efficiently as transmission FCT.

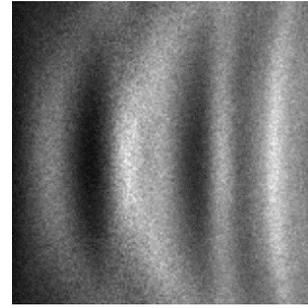


Figure 2: Simulated reflection mode FCT data for Al_2O_3

Due to the complexity of the geometry required for reflection FCT, Al_2O_3 was utilized to ensure the correct geometries of the detector and sample. Reflection FCT empirical data for Al_2O_3 (Figure 3) was consistent with the simulated data (Figure 2). This confirmed the empirical data was obtained from reflection signatures as appose to transmission data, validating the correct geometry had been attained. With increasing thicknesses of soft tissue, there is a decrease in the intensity of the diffraction signatures for both pencil beam and FCT modalities, however this is more prominent in the conventional XRD signatures. The intensity from FCT is much greater than the corresponding pencil beam, even with increasing layers of soft tissue suggesting FCT would be a superior modality for *in vivo* diagnostics.

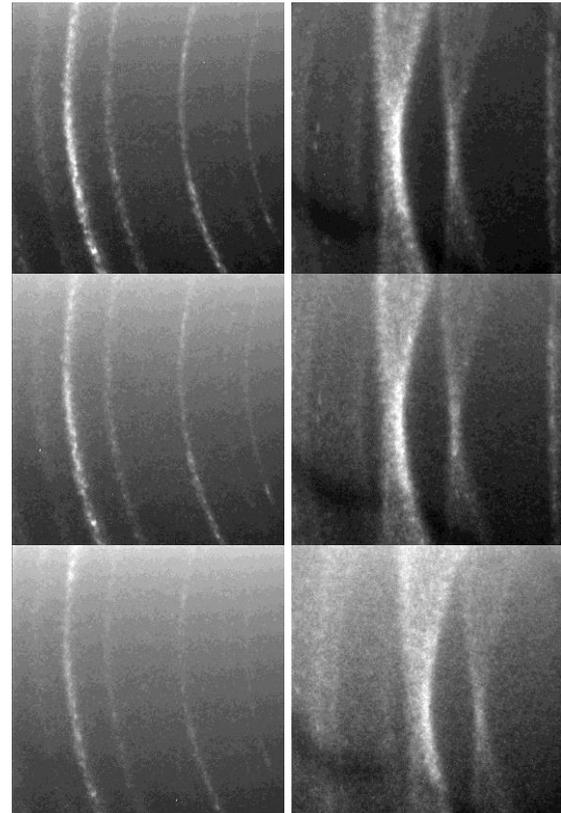


Figure 3: Pencil beam (left) and FCT (right) reflection mode, Al_2O_3 with differing thickness of soft tissue: 0 mm (top), 3mm (middle) and 6mm (bottom).

4 CONCLUSION

Bone is a much poorer scatterer than Al_2O_3 , and therefore obtaining a quantifiable diffraction signal can be much more difficult. For the first time, diffraction signatures from bone were obtained in reflection mode using the FCT modality (Figure 4). The low intensity diffraction signal is to be expected from this material, and when overlaid with increasing soft tissue it is not surprising the diffraction signatures become weaker. However, based on the results from the ideal sample, Al_2O_3 , if *in vivo* diagnostics is required, then FCT in reflection mode appears to showing some potential and perhaps for bone analysis, the detector utilized needs to be considered.



Figure 4: FCT in reflection mode for bone with differing layers of soft tissue, 0 mm (left) and 6 mm (right).

3.2 HEXITEC

The sensitivity in transmission mode of the HEXITEC detector (Figure 5) appeared to be significantly greater than that of the PIXIS detector, especially when the bone sample was overlaid with soft tissue. Further work is currently being carried out to probe the capabilities of the HEXITEC detector using FCT in reflection mode and to investigate the energy dispersive capabilities of this hyperspectral detector.

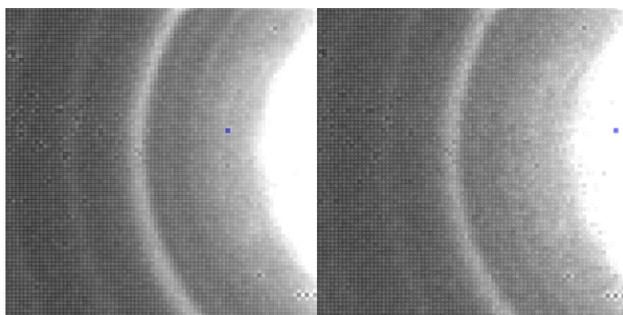


Figure 5: FCT in transmission mode for bone with differing layers of soft tissue, 0 mm (left) and 6 mm (right) collected using HEXITEC detector.

The primary aim of this study was to assess the suitability of both conventional XRD and FCT for *in vivo* fracture diagnostics. This included assessing whether a signature from *ex vivo* bone can be obtained using these modalities, whilst also considering the effect of increasing thicknesses of overlaying muscle/fat/skin. Further, the capabilities of a new hyperspectral detector, Hexitec, was assessed for this work, taking the initial steps towards a new, low cost, high impact diagnostic tool.

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