Trends in Applied Spectroscopy
Vol. 10, 2013

Original Communication

The use of Europiumstearate to trace polyethylene wear debris in joint fluid after prosthetic joint replacement - A feasibility study

J. Kunze[1,2,*], V. Ngai[1], S. Koelling[2], J. J. Jacobs[1] and M. A. Wimmer[1]

[1]Department of Orthopedic Surgery, Rush University Medical Center, 1611West Harrison Street, Chicago, IL 60612, USA. [2]Central Laboratory of Analytical Chemistry, Hamburg University of Technology, Eißendorfer Strasse 38, D-21073 Hamburg, Germany

ABSTRACT

Ultra-high molecular weight polyethylene (UHMWPE) is the most common counterface material against metals or ceramics in artificial hip or knee joints. Wear and the resulting particulate debris, however, limit the life span of the implant. In this study, the general feasibility of using Europium (Eu) as tracer material to quantify UHMWPE wear in joint fluid is investigated. Using Inductively Coupled Mass Spectrometry (ICP-MS), recovery experiments of Eu in artificial joint fluid were performed. In order to dope polyethylene with 50 ppm Eu, nascent UHMWPE powder was mixed with a solution of Eu-stearate. The heterogeneity of the mixture was assessed by determining the coefficient of variation (CV) of the Eu content in various weighted samples. After molding of the UHMWPE powder mixture, cylindrical pins of 10 mm diameter were machined and worn against cobalt-chromium metal disks submersed in artificial joint fluid. The Eu-content of fluid samples taken at certain time intervals was measured and compared with UHMWPE weight loss of the pins. A satisfactory homogenization of Eu in the UHMWPE powder was achieved. Tracer-based and weight-loss determined wear rates were highly correlated (Pearson correlation coefficients > 0.991). Also the detection bias was within acceptable limits. Thus both methods demonstrated good agreement.

KEYWORDS: europium, rare earth stearate, artificial joints, polyethylene wear, tracer

INTRODUCTION

Nowadays, the replacement of joints in the human body with prosthetic devices is a surgical routine. For example, the annual number of total hip and knee replacements in Organisation for Economic Co-Operation and Development (OECD) countries has climbed to 3.3 million in 2009, an incline of 50% since the start of the new millennium [1]. These numbers are expected to keep growing in the next several years [2].

In total hip replacement surgeries, the cup-shaped cartilage articulation of the pelvis is replaced with ultra-high molecular weight polyethylene (UHMWPE). The femoral head is replaced with a metal or ceramic ball. In the case of total knee replacements, the surfaces of the femoral condyles are most often replaced by a metallic component, while the tibial plateau is replaced by UHMWPE. Hence, in both cases the artificial articulation comprises of a hard body (metal or ceramics) that articulates against a soft plastic counterface providing a low friction interface. However, wear of UHMWPE and in particular adverse body reactions to wear debris limit the life span of these devices [3, 4].

*Corresponding author: j.kunze@tuhh.de

Accurate determination of UHMWPE wear *in vivo* would allow early intervention to minimize patient complications [5]. The composition of UHMWPE does not allow a direct chemical determination of wear particles if suspended in protein containing liquids. Hence, a marker that would identify UHMWPE wear appears lucrative. Here, the feasibility of adding Eu-stearate as traceable substance ('tracer') to quantify UHMWPE wear debris in joint fluid is investigated. The Eu-concentration could then be quantified according to Taylor *et al.* [6] and related to polyethylene wear, assuming the concentration of Eu in UHMWPE is known and homogeneous throughout the plastic device.

The specific aims of this paper are :

1. to describe a novel methodology to add a traceable substance to UHMWPE powder,
2. to establish the homogeneity of tracer distribution within the UHMWPE powder, and
3. to validate this novel methodology against established gravimetric techniques of UHMWPE wear determination.

EXPERIMENTAL

Europiumstearate synthesis

Eu-stearate, common for Europium (III) octadecanoate, was prepared using Eu-nitrate as a commercially available product. Europium (III) stearate $(C_{18}H_{36}O_2)_3Eu$ was prepared by adding a solution of $Eu(NO_3)_3\ 6H_2O$ (4.62 mmol, 2.00 g) in water (10 cm^3) to a solution of sodium octadecanoate (13.86 mmol, 3.47 g) in water (100 cm^3). Sodium octadecanoate was obtained by neutralizing octadecanoic acid with 0.5 M NaOH. Immediately after adding the europium (III) solution, a white precipitate is formed. The precipitate was digested for 1 h, filtered and washed thoroughly with water. The compound was dried in an oven for 4 h at 40 °C. Europium (III) octadecanoate was obtained as colorless powder, yield 70% [7].

Doping UHMWPE powder and testing homogeneity

The Eu-stearate was weighed in a 1 liter round flask, dissolved in ligroin (petroleum ether) by boiling at a reflux equipment and mixed with nascent GUR 1050 UHMWPE powder using an ultrasonic bath. The ligroine was distilled by a rotation evaporator and the powder was dried in an oven for 2 h at 100 °C. 50 mg of Eu-stearate was mixed with 100 g of UHMWPE in order to obtain 50 mg/kg of Eu in the UHMWPE-powder. The homogeneity of the tracer in mixed powder was tested by weighing different amounts of Eu-doped UHMWPE powder together with 0.5 mL water in quartz vessels and applying an established digestion protocol for lubricant samples in the microwave Perkin–Elmer ®(Paar) Multiwave. Digestion mixture and protocol : 0.5 mL lubricant, 1 mL HNO_3, 1 mL H_2O, 800 W 15 min., Temperature during digestion 230 °C.

1, 4, 7, 10, and 100 mg weighted samples of n = 6 each were analyzed. Independent of the weighted sample, the dilution factors were adjusted to yield a final Eu-concentration of 5 ng/mL (ppb). To quantitatively describe tracer homogeneity, the coefficient of variation (CV) was calculated for each of the sample weights. CV is a normalized measure of dispersion and defined as the ratio of standard deviation to sample mean. A low CV was indicative of high homogeneity.

Manufacturing of wear testing samples and wear test

To manufacture components for wear testing, Eu-doped UHMWPE powder was compression molded. Pressure and temperature protocol were similar to those typically applied for implants. Six pins, each 10 mm in diameter and 20 mm in length were then machined from the molded sheet. The as-molded condition of the bottom was used for wear testing.

A six station pin-on-disk wear tester (OrthoPOD™, AMTI, Boston, USA) was employed. Each pin articulated with a multi-directional motion pattern against a polished cobalt-chromium disk. The pin was constantly loaded with 200 N (contact pressure of ~2.5 MPa) and submersed in 15 mL artificial joint fluid at 37 °C. The latter consisted of new born calf serum diluted to a total protein content of 30 g/L which was replaced with fresh fluid after each testing interval which lasted 300,000 cycles or 3.5 days. Then, the components were cleaned as per ASTM F2025-06 and the UHMWPE pins were weighed on a high precision balance (Mettler Toledo AX 205, Switzerland) three times.

The gravimetric readings for each pin at every measurement point were averaged.

Joint fluid sampling and analysis

Three lubricant samples per articulation were taken before the start of the experiment (to confirm that there was no contamination and no tracer was leaking from the pins) and at the end of each interval until 1.2 million cycles were reached. Thus, a total of 90 samples (3 replicates x 6 stations x 5 measurement points) were analyzed for the presence of Eu via ICP-MS. For comparison with gravimetric readings, the three replicate readings were averaged. At the end of each interval, prior to the cleaning of the components, each station was weighed and returned to original weight (weight at the interval start) with high purity water to account for evaporation loss. A thorough mixing and bubbling procedure with nitrogen gas ensured a homogenous solution. 0.5 mL of fluid has been pipetted into microwave containers. The sample weights were controlled by a balance. 1 mL HNO_3 and 1 mL of H_2O were added per sample. The containers were shut and the digestion protocol (800 W, 15 min) was applied. After cooling, 10 µg/L Rh as internal standard was added. A simultaneous measurement of Eu isotopes 151 (isotopic abundance: 47.8%) and 153 (isotopic abundance: 52.2%) was performed, using a quadrupole inductively coupled plasma mass spectrometer (ICP-MS). As both isotopes gave nearly the same readings, Eu 153 was used for calculation.

Analytical instruments

Europium analyses were carried out with a Quadrupole ICP-MS, using the Perkin Elmer Sciex DRCII®, equipped with a Perkin Elmer AS-93plus auto sampler (Perkin Elmer Life Sciences, Shelton, CT, USA).

Sample decomposition was performed using a microwave digestion system (“Multiwave”, Perkin Elmer/Paar, Anton Paar, Graz, Austria).

Reagents

Europium standard stock solutions: 1.000 g/L (B. Kraft GmbH, Duisburg, Germany).
Internal Standard solution: 1 mg/L Rh (Merck, Darmstadt, Germany).
H_2O Milli-Q™ system (Millipore Corporation, Bedford, MA, USA).
HNO_3 (65%) subboiling distillation grade.

Interferences

The determination of Europium can be interfered by polyatomic species from Barium.

The building of $Ba^{136}OH^+$ at 153 m/z depends on the building of oxides in the plasma. Therefore the oxide rate which is measured as ceroxide/cerium (CeO/Ce) ratio is predominant to this interference. A “daily performance” solution containing Ce is analyzed at the beginning of the ICP-MS measurements and the CeO/Ce ratio is determined. The rate is < 3%. To check for the stability of the measurements, two quality check samples with 0.1 and 1.0 µg/L Eu are analyzed every ten samples. The coefficient of variation for both samples was < 5% during one day.

According to Kent/Ungerer [8] contributions from BaO to Eu are significant for materials with Ba/Eu ratios >> 1000. The Ba concentration in the serum samples was 32 µg/L. The Eu concentration in our samples (diluted for measurement) was in the range of 0.06–0.15 µg/L; thus, the Ba/Eu ratios were 213–533.

Matrix effects

The artificial joint fluid was composed of 30 g/L protein, 9 g/L NaCl and 18 g/L Tris(hydroxymethyl) aminomethan adjusted to a pH of 7.6 with HCl.

0.5 mL of the solution is used for the analysis. Microwave acid digestion reduces the carbon content to a minimum, but the salt matrix changes the viscosity of the solution and after analyzing a couple of samples a deposition of salt can be observed at the sampler and skimmer conus of the instrument which causes poor repeatability. After 30 samples, the sampler cone had to be cleaned with a solution of 10% HNO_3 in a beaker put in an ultrasonic bath for 10 minutes.

Therefore 1:10 dilution of the sample is recommended, as described above, and the sample introduction system should be rinsed with Millipore water for 60 seconds between two samples.

Method validation

Blank artificial joint fluid samples were used for recovery experiments of Eu in matrix.

Table 1. ICP-MS conditions for the determination of Eu.

Isotopes	:	151, 153 m/z
Internal Standard	:	Rh, 103 m/z
ICP RF power	:	1200 W
Ar gas flow	:	15 L/min
Sample Uptake Rate	:	~ 1.0 mL/min
Nebulizer Gas Flow	:	1.0 mL/min
Oxide Rate CeO/Ce	:	< 3%
Nebulizer Type	:	Concentric Glass Nebulizer, High Efficiency, HEN-170 AA.
J.Meinhard		
Associates Inc. California, USA.		
Calibration		
Calibration in digested blank artificial joint fluid samples.		
Internal standard	:	10 µg/L Rh
Standard concentration	:	0/0.1/0.5/1/2/5/10 µg/L Eu
Linearity of the curve Eu 153 m/z	:	0.999973
Limit of detection (LOD) calculated from the calibration curve according to DIN 32645.		
LOD (95%)	:	0.15 µg/L. Equal to 0.012 µg/L in the solution ready for measurement.
Method variation coefficient (V_{xo})	:	0.8%

A spike solution of 10 and 100 µg/L Eu was prepared. Six levels were tested in replicates:

0.5 g serum + 0.2/0.4/1.0/2.5/5.0/10.0 µg/kg Eu. Recovery rates with < 2% error were achieved. Occurring interferences were considered as described above. As shown in Table 1, the limit of detection was determined to 12 ng/L.

RESULTS

Homogeneity of tracer distribution

The coefficient of variation, as a measure of homogeneous tracer distribution, was dependent on weighted mass and declined from 7.8% to 3.6% to 3.4% to 1.5% for samples masses of 1, 4, 7, and 10 mg, respectively. After 10 mg the CV stayed constant at 1.5%. The actual concentration of Eu in the powder was close to target value of 50 mg/kg.

Validation using wear test

There was no Eu detectable in any of the fluid samples at the start of wear testing (blank value = zero). Then, after each testing interval, Eu-concentrations in the artificial joint fluid samples ranged from 25 to 160 ng/L. For each of the six stations, the wear readings based on Eu concentrations and those based on weight loss measurements correlated highly and significantly. Pearson correlation coefficients ranged from 0.991 to 0.999. The slope of the correlation line was not significantly different from 1 ($p = 0.557$; 95% confidence interval: 0.82…1.29), however, as can be seen from the Bland-Altman plot in Figure 1, the mean difference between both measurement techniques was biased towards higher wear readings measured by the chemical technique. This difference of 0.067 mg was largely driven by wear readings measured during the early phase of the experiment (i.e. by small wear readings).

DISCUSSION

As was expected, the recovery of Eu in artificial joint fluid proved to be highly reliable with excellent precision. Also, an acceptable homogeneity of the Eu-stearate within the UHMWPE-powder was achieved. Both are prerequisites for a successful tracer application. Purposefully, an Eu concentration of 50 ppm in polyethylene was targeted, because this is the limit for calcium in medical grade UHMWPE Type 3 containing

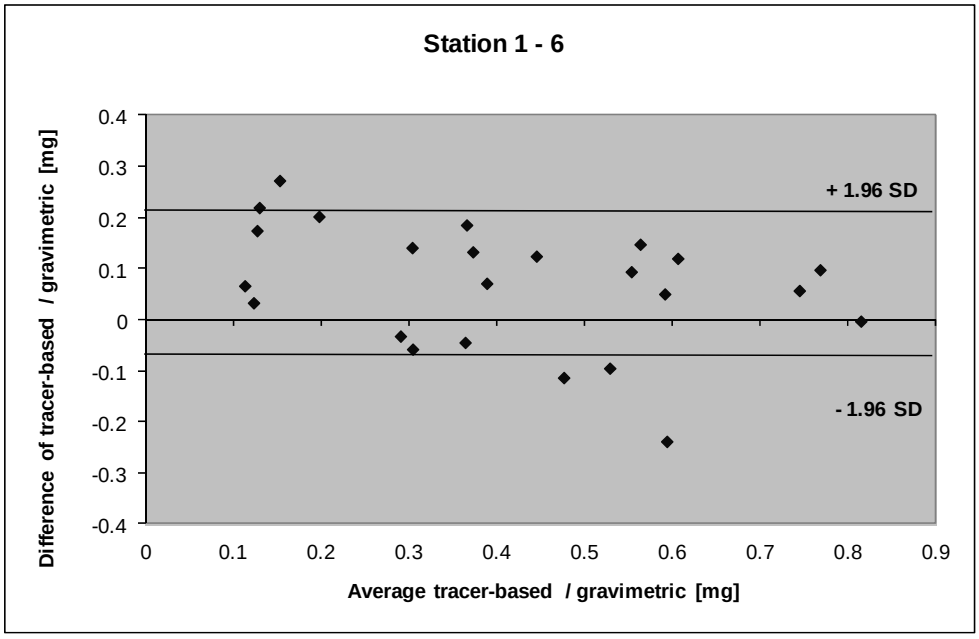

Figure 1. Tracer-based wear readings of 6 Eu-doped UHMWPE pins from six different wear stations compared with "gold standard" gravimetric measurements.

calcium-stearate as per ASTM standard F648 [9]. Further, since the annual material loss of conventional UHMWPE *in vivo* is in the range of 50 mg (and that of cross-linked UHMWPE between 5 and 10 mg), the determined CV provides sufficient accuracy to measure meaningful clinical wear rates in joint fluid, tissue samples, and possibly blood vials. Eu has been chosen as tracer material because of its high sensitivity and little interference with substances in biological media using ICP-MS. In addition, Eu has a track record of biocompatibility and demonstrated anti-oxidative and anti-inflammatory behavior *in vivo*.

The wear experiment was meant to mimic the process of UHMWPE particle release *in vivo*. All obtained Eu-concentrations were well above the detection limit and none of the manufactured pins leaked tracer without wear. Tracer determined measurements were highly correlated with pin weight loss, however, some bias was noticed. It is possible, that the UHMWPE pins soaked fluid during initial rubbing, thus, masking the weight loss. Yet, to this date, gravimetric techniques are considered the gold standard of wear determination for UHMWPE.

CONCLUSION

In conclusion, this study demonstrated the feasibility of tracing UHMWPE wear debris through an Eu-marker. The introduction of a tracer substance may open opportunities to quantify polyethylene debris using tissue and/or liquid samples.

ACKNOWLEDGMENTS

The authors received grants from the Taiho Kogyo Tribology Research Foundation and National Institutes of Health (NIAMS R01AR059843; PI Wimmer). Further, the support of Zimmer Inc. is acknowledged who helped with the manufacturing of UHMWPE pins and CoCr disks.

REFERENCES

1. OECD 2011, OECD Publishing. Accessed 5 June 2013.
2. Kurtz, S., Ong, K., Lau, E., Mowat, F. and Halpern, M. 2007, J. Bone Joint Surg. Am., 89(4), 780-5.
3. Jacobs, J. J., Hallab, N. J., Urban, R. M. and Wimmer, M. A. 2006, J. Bone Joint Surg. Am., 88(Suppl.), 299-102.
4. Catelas, I., Wimmer, M. A. and Utzschneider, S. 2011, Semin Immunopathol., 33(3), 257-71.

5. Beck, R. T., Illingworth, K. D. and Saleh, K. J. 2012, J. Orthop. Res., 30(4), 541-6.
6. Taylor, A., Day, M. P., Hill, S., Marshall, J., Patriarca, M. and White, M. 2013, J. Anal. Atom. Spec., 28(4), 425-459.
7. Binnemans, K. and Leuven, K. U. 1999, Liquid Crystals, 26(11), 1717- 1721.
8. Kent, A. J. R. and Ungerer, C. A. 2005, J. Anal. At. Spectrom, 11, 1256-1262.
9. ASTM F648-10a. ASTM International, 2010, DOI: 10.1520/F0648-10A.