Gentamicin elution profile of SmartSet® GHV Gentamicin Bone Cement

Introduction

There is no doubt that, since the invention of artificial joint replacement, many thousands of people have enjoyed freedom from disabling pain and immobility. However, this remarkable success story is marred by the incidence of local sepsis following surgery, which can lead to prosthesis loosening and the need for revision surgery. In fact, deep wound infections are the most serious adverse event associated with primary cemented joint arthroplasty and have the most profound consequences for both patient and healthcare resources alike. The osseous and vascular disruption associated with joint replacement surgery, combined with the substantial heat generated by the in vivo polymerisation of bone cement, create an immunoincompetent zone, where a smaller than normal amount of bacteria are required to establish an infection. These factors also make it difficult to eliminate microbes from the site, and to provide a relatively safe environment from circulating immunodefences and systemic antibiotics. Improved hygiene techniques, systemic antibiotic administration and the introduction of antibiotic-loaded bone cement as a prophylactic measure during joint replacement surgery, have gone a long way to address this problem. The use of bone cement as a depot for antibiotics was first reported in 1970 by Buchholz and Engelbrecht, who simply mixed Biomet Palacos® R40 Bone Cement with gentamicin. Since that time, despite reports of gentamicin resistance in nosocomial infections, gentamicin has become commonly used in bone cements due to its wide spectrum antimicrobial activity, stability under high temperatures and sterilisation conditions, water solubility and relatively low incidence of allergic response. However, the release mechanism of gentamicin from bone cement was largely unknown when Buchholz and Engelbrecht were first investigating it, and is still debated to this day. There has been much research into the elution of gentamicin from bone cement. Many studies indicate that, as bone cements are hydrophobic, most of the elution occurs on the surface of the cement in the space between cement and bone. Bone cement samples less than 100 μm thick have been shown to release all of the antibiotics incorporated, while release from thicker samples occurs independent of volume. Studies using methylene blue and gentamicin diffusion through, or into, acrylic bone cement discs also show that the bulk of the cement is essentially impermeable. Many of the in vitro studies indicate that only 5-8% of the antibiotic incorporated in an acrylic matrix is eventually released, although the duration of testing, differences in refreshing intervals, sample geometry, temperature, pH and detection assay strongly affect the amount released. In vivo studies also confirm that only a minor portion (5-18%) of gentamicin incorporated in bone cements is eluted. Only recently has one study reported on the measurement of antibiotic release from bone cement in a narrow space, simulating a prosthesis-related gap. This study demonstrated that concentrations up to 1000-fold the minimal inhibitory concentration (MIC) levels for most infecting strains can be achieved in a realistic in vitro model, although the in vivo correlation is difficult to draw from these experiments alone.

The clinical efficacy of the use of antibiotic-loaded bone cements for primary implant fixation is difficult to assess in clinical studies, because many numbers of patients are needed and no double-blind, randomised studies are available for drawing statistically reliable conclusions. Nevertheless, it is clear that:

- it is essential that antibiotic-loaded bone cements provide a therapeutic, yet non-toxic level of antibiotic release in the immediate period after skin closure following artificial joint replacement
- in the absence of a definitive clinical study to establish a correlation between the in vitro elution of antibiotics from loaded bone cement and clinical success, we draw together the existing in vitro data and 30 years of clinical experience.

DePuy CMW has been investigating the clinical performance and elution properties of antibiotic-loaded cements for many years, having launched their first commercial antibiotic-loaded bone cement in 1991.
SmartSet® GHV Gentamicin Bone Cement (SmartSet GHV) is one of the latest antibiotic-loaded bone cements in the DePuy CMW portfolio. It is chemically equivalent in composition to other bone cements on the market and the powder component is sterilised by ethylene oxide to prevent the degradation of polymer molecular weight caused by gamma irradiation sterilisation. Furthermore, the particular formulation of polymers used in its manufacture reduces susceptibility to temperature variations during the mixing and setting phases.

This paper reports the findings of a recent DePuy CMW study to investigate the in vivo elution profile of gentamicin from SmartSet GHV as well as its in vivo safety and efficacy when used in primary total hip and total knee arthroplasty. Furthermore, it compares the antibiotic elution rates of Refobacin® Palacos® R40 (Biomet Merck) and Palacos R40G (Schering Plough) with those of Palacos R+G (Heraeus Kulzer) and Refobacin Bone Cement (Biomet).

**Study objectives and methodology**

**In vitro assessment**

The objective of the in vitro investigation was to compare the elution of gentamicin from SmartSet GHV (DePuy International Ltd), Refobacin Bone Cement (Biomet) and Palacos R+G (Heraeus Kulzer) Bone Cements. Elution profiles of the two latter cements were compared to elution profiles of two other Palacos brands, Refobacin Palacos R40 (Biomet Merck) and Palacos R40G (Schering Plough). The analysis was carried out using a high performance liquid chromatography (HPLC) method. The cement samples were prepared by mixing to the dough phase and then moulding into discs. Once cured, each disc was placed into a separate container of phosphate-buffered saline solution, and at predetermined time points the supernatant liquid was analysed for gentamicin content, the disc transferred into a fresh container, and the analysis repeated at the next time point.27,28

**In vivo assessment and clinical evaluation**

A clinical study investigating the in vivo gentamicin elution profile and long-term performance and safety of SmartSet GHV when used in total knee and hip arthroplasty is on-going at Wrightington Hospital in the UK. The study protocol29 required each subject to be monitored with clinical and radiographic assessments at 5 months, 1, 2, 5 and 10 years post-operatively. Subjects are also required to return for clinical and radiographic assessments at 5 months, 1, 2, 5 and 10 years post-operatively. Adverse events and concomitant medications are recorded throughout the study period.

**Results**

**Release of gentamicin from bone cements: in vitro assessment**

SmartSet GHV displayed the greatest elution rate at 15 minutes (0.099 mg/cm²/hr), compared with Palacos R+G (0.031 mg/cm²/hr) and Refobacin Bone Cement (0.028 mg/cm²/hr) [Figure 1]. This initial burst coincides with the immediate post-op erative period where infection risk is greatest.20 The initial elution peak of gentamicin was followed by a prolonged release over 24 hours; however, SmartSet GHV bone cement displayed a greater total amount of gentamicin being released over the first 0.75 hours.

The comparison between Refobacin Palacos R40, Palacos R40G, Palacos R+G and Refobacin Bone Cement products (Figure 2) shows a clear difference in the antibiotic elution profiles between products. The latter two products exhibit an initial burst that is almost half the concentration of Refobacin Palacos R40 and Palacos R40G. However, all products show a prolonged release of gentamicin after the initial burst, with the exception of the Palacos R40G where the gentamicin release appeared to essentially cease after the first 0.75 hours.

**Release of gentamicin from bone cement: clinical evaluation**

A total of 11 subjects recruited into the study undergoing total hip arthroplasty, and 8 subjects for total knee arthroplasty, received SmartSet GHV as part of this study. To date, one subject from the hip cohort has been withdrawn from the study.

A total of 12 adverse events have been reported by nine patients at the time of writing this paper (Table 1), although none of these complications are considered to be related to SmartSet GHV. One further event was reported of a knee subject requiring a prophylactic dose of gentamicin prior to a catheter removal. The data from this subject following the dose has therefore been excluded from this analysis. All remaining subjects have completed 2-year follow-up assessments to date, and will continue to be monitored with clinical and radiographic assessments to determine the long-term performance of SmartSet GHV according to the study protocol.29

The following three response criteria for the elution of gentamicin in wound fluid, serum and urine were achieved on post operative assessment:

- maximum wound fluid concentration of gentamicin >0.06 μg/mL (to ensure effective concentrations were achieved)
- maximum serum concentration of gentamicin <10 μg/mL (for safety)
- reducing urinary concentrations of gentamicin (to indicate clearance from the subject).

Figure 1. Elution rate of gentamicin for SmartSet GHV, Palacos R+G and Refobacin Bone Cement measured by HPLC.27

Figure 2. Elution rate of gentamicin for Palacos R+G, Palacos R40G, Refobacin Palacos R40 and Refobacin Bone Cement measured by HPLC28.
Maximum wound fluid concentrations of gentamicin were >10 μg/mL for all subjects with the exception of one, whose maximum level was four times greater than the prespecified level for effective concentrations (Figure 3a). Maximum wound fluid concentrations were achieved 1 hour post-operatively in all but one subject, who achieved maximum wound fluid concentrations after 2 hours. The maximum serum concentration of gentamicin for any subject was at least 0.5 μg/mL, which is well below the 10 μg/mL that would give rise to toxicity threshold of 10 μg/mL in all subjects, one hip subject. The maximum serum concentration of gentamicin was below the toxicological level in the immediate post-operative period and demonstrated a prolonged elution rate up to 48 hours.

From the comparison between Refobacin Palacos R40, Palacos R40G, Palacos R+G, and Refobacin Bone Cement products at 15 minutes. This is when the rapid release of antibiotics at a therapeutically effective level is vital for the treatment of post-operative infection.26 The initial burst of gentamicin release at 15 minutes was followed by a prolongation of release for up to 48 hours at which point the elution levels out; this can be observed with all products studied. The in vitro results infer that SmartSet GHV can perform effectively in the immediate post-operative period and demonstrates a prolonged elution rate up to 48 hours.

Discussion

A key factor in the elution of gentamicin from bone cement is the particle size of the gentamicin. Specifically, larger particles elute in greater quantities than smaller ones, and it is considered that whilst the cement powders for the Refobacin Palacos R40 and Palacos R40G were both supplied by the same manufacturer, it is clear from the differing elution profiles that the respective grades of gentamicin are different. Colour-staining experiments have confirmed this, showing the Biomet Merck gentamicin to be coarser than the Schering Plough (data on file at DePuy). Furthermore, it is possible that coarser particles of gentamicin lead to reduced mechanical performance (the gentamicin acts as a mechanical defect in the cement matrix), hence there is an obvious need to balance particle size against (for example) fatigue performance. SmartSet GHV is formulated with 1 g of a highly controlled particle size of active gentamicin, resulting in a cement with the desired elution profile, and at the same time maintaining its excellent fatigue strength.

In vitro results show that the elution rate of gentamicin from SmartSet GHV is almost three times that of the Palacos R+G and Refobacin Bone Cement products at 15 minutes. This is when the rapid release of antibiotics at a therapeutically effective level is vital for the treatment of post-operative infection.26 The initial burst of gentamicin release at 15 minutes was followed by a prolongation of release for up to 48 hours at which point the elution levels out; this can be observed with all products studied. The in vitro results infer that SmartSet GHV can perform effectively in the immediate post-operative period and demonstrates a prolonged elution rate up to 48 hours.

From the comparison between Refobacin Palacos R40, Palacos R40G, Palacos R+G, and Refobacin Bone Cement, the initial burst of gentamicin release for Palacos R+G and Refobacin Bone Cement products was almost half the concentration of Refobacin Palacos R40 and Palacos R40G products. In addition to this, the Palacos R40G had almost zero gentamicin release after 0.75 hours, whereas the Palacos R+G had a prolonged release of gentamicin up to 48 hours.

The in vitro elution pattern was reflected in the results of the clinical study for SmartSet GHV. Importantly, in all cases the maximum concentration of gentamicin in wound fluid was well above the MIC for staphylococci (4 μg/mL), being the most important species in orthopaedic implant infections.24 Furthermore, maximum wound fluid concentrations were achieved at 1 hour post-operatively in all but one hip subject. The maximum serum concentration of gentamicin was below the toxicity threshold of 10 μg/mL in all subjects, while progressive reductions in urinary concentration indicated clearance from the body.
While it is difficult to extrapolate in vitro data to predict clinical success with absolute certainty, the congruence in gentamicin elution patterns demonstrated in these results, combined with the successful short-term outcome achieved by the patients, clearly supports the theory that a therapeutically effective amount of gentamicin release in the immediate post-operative period, without undue prolongation of antibiotic elution is an important consideration in the management of infection. Furthermore, these results give added weight to the growing body of evidence that supports the use of antibiotic-loaded bone cements as a prophylactic measure during primary and secondary total joint arthroplasty.14,15

Table 1. Adverse events in the subjects receiving SmartSet GHV

<table>
<thead>
<tr>
<th>Hip (n=11)</th>
<th>Knee (n=8)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cardiovascular/circulatory</td>
<td>1</td>
</tr>
<tr>
<td>Gastrointestinal</td>
<td>0</td>
</tr>
<tr>
<td>Genitourinary</td>
<td>0</td>
</tr>
<tr>
<td>Deep vein thrombosis</td>
<td>1†</td>
</tr>
<tr>
<td>Wound problems</td>
<td>1†</td>
</tr>
<tr>
<td>Other*</td>
<td>2†</td>
</tr>
</tbody>
</table>

* Included raised temperature (hip group) and local infection, sacral pressure sore and tight, stiff knee (knee group)
† Three adverse events related to the same patient
†† One patient experienced two ‘other’ adverse events

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Conclusions

- Maximising the efficiency of antibiotic release from bone cement may improve therapeutic efficacy and clinical outcome.
- The in vitro and in vivo gentamicin elution profiles of SmartSet GHV show similar trends which indicate that well-discussed in vitro studies may be a useful predictor of clinical outcome.
- In clinical evaluation, SmartSet GHV is safe and efficacious for the implantation of total hip and knee arthroplasties.
- There are differences in the hourly elution rate of gentamicin between Refobacin Palacos R40 and Palacos R40G, Palacos R+G and Refobacin Bone Cement.

References

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29. Protocol number CMW01-09-2006. Kay P Consultant Orthopaedic Surgeon. A study to evaluate the safety, performance and gentamicin elution profile of a new bone cement Acrylic Gentamicin (SmartSet GHV) in comparison to an existing bone cement CMW01 Gentamicin when used in primary hip and knee arthroplasty.