S eptic arthritis occurs when microorganisms enter the synovial space of a joint, colonize synovial membranes, and multiply, causing severe inflammation. The infection may be of hematogenous origin; may be caused by direct contamination of the joint from a wound, surgery, or intra-articular injection; or may be caused by extension of infection from adjacent structures such as a septic physis. Successful treatment of septic arthritis in horses is often difficult and expensive, and septic arthritis can end a horse's athletic career or life. Rapid elimination of the infection is necessary to avoid loss of normal joint function, secondary osteoarthritis, and euthanasia. Treatment of septic arthritis often requires administration of anti-inflammatory drugs, systemically and locally administered antimicrobial drugs, and removal of bacteria, inflammatory products, devitalized tissue, and other debris. Successful treatment depends on many factors, including virulence of the causative agent, competence of host defense mechanisms, efficacy of chosen antimicrobial drugs, and effective antimicrobial tissue concentrations following antimicrobial administration.

The septic environment decreases both antimicrobial delivery and efficacy. Decreased blood flow, arthritis, chronic lameness, or euthanasia.
increased joint pressures, and capillary obstruction by fibrin and debris in infected joints may impair diffusion of antimicrobials into the joint. Low tissue pH may also inactivate some antimicrobials such as gentamicin sulfate and amikacin sulfate. Aminoglycosides are less likely than gentamicin sulfate. The dose of amikacin sulfate has good activity against many equine orthopedic pathogens, and resistance to amikacin sulfate is less likely than to gentamicin sulfate. The dose of amikacin sulfate routinely administered via IVRLP to horses in clinical practice is 500 mg to 2 g diluted with physiologic saline (0.9% NaCl) solution to a volume of 20 to 60 mL. Regional treatment decreases the risk of nephrotoxicosis associated with systemic use of gentamicin sulfate and amikacin sulfate because smaller doses, compared with systemic administration, can be used.

A tourniquet is a tight circumferential band that is applied around an extremity to compress blood vessels to stop blood flow and control hemorrhage. Tourniquets are often used in horses for orthopedic surgery distal to the carpus and tarsus or to perform regional limb perfusion with either an anesthetic agent to desensitize the limb for surgery or with an antimicrobial drug for treatment of sepsis. The pneumatic tourniquet is applied by wrapping the pneumatic cuff around the limb at the desired location and attaching the cuff to the inflation device, which is then set to pressurize the cuff. Pressure can be adjusted from zero to 500 mm Hg in many pneumatic tourniquets. The Esmarch tourniquet is an elastic rubber bandage that is 10 to 12 cm wide. Blood flow is stopped by tightly wrapping the bandage around the limb. Pressure under the bandage cannot be adjusted but has been recorded in dogs as attaining 1,000 mm Hg. Use of tourniquets has been associated with numerous complications, and it has been assumed that pneumatic tourniquets are safer than Esmarch tourniquets. Regional limb perfusion has been performed with the Esmarch and pneumatic tourniquets. Pneumatic tourniquets are often set at a pressure of 420 mm Hg. However, 1 study that used a pneumatic tourniquet set at 300 mm Hg detected losses of antimicrobial drugs into the systemic circulation. An Esmarch tourniquet prevented leakage of the perfusate into the systemic circulation in another study. The pressure under the tourniquet was unmeasured.

Physical removal of inflammatory products, microorganisms, fibrin, and debris is often accomplished by use of joint lavage and drainage. Common methods for irrigation include tidal irrigation in which fluids are injected and aspirated through the same needle, through-and-through lavage with injection and retrieval of lavage fluids through separate needles, and arthroscopic lavage. In our practice and others, horses with septic arthritis are commonly treated with IVRLP performed simultaneously with joint lavage. To perform IVRLP, a tourniquet is placed around the limb proximal to the infected joint, and the limb is perfused for 30 minutes. During IVRLP, the septic joint is lavaged. Simultaneous IVRLP and joint lavage decrease the time required for treatment on standing sedated horses and the length of general anesthesia in anesthetized horses. We have questioned how much antimicrobial drug is lost in egress lavage fluids. To our knowledge, there are no reports on the amount of antimicrobial drug that is lost in lavage fluids from joints of limbs that are simultaneously receiving IVRLP.

The purpose of the study reported here was to determine whether joint lavage performed simultaneously with IVRLP reduces the effectiveness of IVRLP through loss of antimicrobial drugs in the egress lavage fluid and to compare 2 types of tourniquets used for IVRLP. We hypothesized that substantial losses of antimicrobial drugs from the distal portion of the limb would occur during joint lavage when performed at the same time as IVRLP and that there would be no difference in antimicrobial drug concentrations in the distal portion of the limb of standing horses following IVRLP performed by use of a pneumatic tourniquet set at 420 mm Hg versus use of an Esmarch tourniquet.

Materials and Methods

Animals—Eleven horses, free of lameness and any obvious musculoskeletal abnormalities of the forelimbs on clinical examination, were included in the study. The horses ranged from 14 to 27 years of age (mean, 19.1 years). The protocol was approved by the Purdue University Animal Care and Use Committee.

Experimental protocol—All horses underwent distal IVRLP and simultaneous joint lavage on 1 forelimb. Each procedure was termed a trial. One horse was used once in the pneumatic tourniquet group and once in the Esmarch tourniquet group with a washout period of 68 days between the 2 regional perfusions. The horses were positioned in metal stocks and sedated with detomidine HCl (0.02 mg/kg, IV). The distal portion of the right forelimb (from the coronary band to the proximal aspect of metacarpal bone III) was clipped, aseptically prepared, and regionally anesthetized with 50 mL of 2% mepivacaine hydrochloride by use of a high palmar and palmar metacarpal nerve block. Six horses had a pneumatic tourniquet placed around the metacarpus just distal to the carpus, and the cuff pressure was set at 420 mm Hg. Six horses had an Esmarch tourniquet placed at the same level as the pneumatic tourniquet. The Esmarch tourniquet was applied each time by the same person (first author) and was wrapped around the limb to the proximal aspect of metacarpal bone III was clipped, aseptically prepared, and regionally anesthetized with 50 mL of 2% mepivacaine hydrochloride by use of a high palmar and palmar metacarpal nerve block. Six horses had a pneumatic tourniquet placed around the metacarpus just distal to the carpus, and the cuff pressure was set at 420 mm Hg. Six horses had an Esmarch tourniquet placed at the same level as the pneumatic tourniquet. The Esmarch tourniquet was applied each time by the same person (first author) and was wrapped around the limb 10 or more full circumferential turns, depending on the circumference of the limb, as tightly as possible without tearing the bandage.

A 20-gauge IV catheter was placed into the lateral palmar vein, and a second catheter was placed in the medial palmar vein at the level of the metacarpo-
phalangeal joint just abaxial to the proximal sesamoid bones. The catheter in the lateral palmar vein was used for amikacin sulfate administration, and the catheter in the medial palmar vein was used to collect blood samples from the distal portion of the limb. Intravenous regional limb perfusion was performed by infusion of 500 mg of amikacin sulfate diluted with 60 mL of sterile saline solution into the limb over a period of 1 minute. Time T0 was designated as the time when the IVRLP was started. One 16-gauge, 3.8-cm-long needle was placed dorsomedially, and one 16-gauge, 3.8-cm-long needle was placed palmarolaterally into the metacarpophalangeal joint. Joint lavage with 2 L of lactated Ringer’s solution was started 5 minutes (T5) after instillation of the amikacin sulfate for all horses except the horse used in trial 8. This horse was uncooperative, and only 1 L was flushed into the joint. Lavage in all horses was performed over a 20-minute time period. The fluids flushed through the joint were collected by use of a 76-cm-long sterile tube as a conduit from the needle into a clean container (Figure 1). The tourniquet was removed after 30 minutes (T30).

Collection of samples—The total amount of the egress fluid collected into the clean container was recorded. To determine amikacin sulfate concentrations distal to the metacarpophalangeal joint, samples of synovial fluid were collected from the distal interphalangeal joint of each horse at T5 and T30. At each sample collection, 3 mL of synovial fluid were collected aseptically from the joint via arthrocentesis of the dorsolateral pouch of the distal interphalangeal joint performed with a 20-gauge, 3.8-cm-long needle. Ten milliliters of venous blood from the jugular vein was collected from all horses before T0 and at 5, 10, 20, and 30 minutes (T5, T10, T20, and T30) after T0. Three milliliters of venous blood was obtained from the catheterized medial palmar digital vein at T5, T10, T20, and T30. These samples were collected after the catheter was cleared by drawing out 3 mL of blood, which was discarded. The catheter was flushed after sample collection with 3 mL of heparinized saline solution.

Postprocedure treatment and evaluation—Thirty minutes after termination of the procedures, 250 mg of amikacin sulfate was instilled into the metacarpophalangeal and distal interphalangeal joints, and the distal portion of the limb was placed in a bandage. Phenylbutazone (4.4 mg/kg, IV) was administered for analgesia 60 minutes after T0. The horses were stall confined for 24 hours. After 24 hours, the bandages were removed, the horses were evaluated for lameness, and the metacarpophalangeal and distal interphalangeal joint regions were evaluated for signs of pain, heat, and swelling.

Measurement of amikacin sulfate concentrations—Concentrations of amikacin sulfate were determined in all synovial fluid, venous blood, and lavage fluid samples. Hemorrhagic synovial fluid samples were centrifuged at 1,700 × g for 5 minutes, and the supernatant was collected. Blood samples from the jugular vein and from the digital vein were allowed to clot and were then centrifuged at 800 × g for 5 minutes. The supernatants from the serum, synovial, and lavage fluids were stored in plastic tubes and frozen at –78°C until assayed for amikacin sulfate. Amikacin sulfate concentrations were determined by use of a fluorescence polarization immunoassay. Serum and lavage fluids were analyzed without any additional sample preparation. Calibration curves were established by use of 6 human serum standards, as described. All synovial fluid samples were diluted 1:1 with 10% methanol solution and were placed into a centrifugal concentrator that had a 10,000 molecular weight cutoff filter. The mixture was vortexed briefly and centrifuged at 1,200 × g for 45 minutes. The clear ultrafiltrate was used for determination of amikacin sulfate concentration. This procedure has been validated.

Statistical analysis—The total amount of amikacin sulfate lost in egress lavage fluids was calculated by multiplication of the concentration of amikacin sulfate measured in the fluids times the volume of fluids retrieved from the flush. When concentrations of amikacin sulfate in the egress fluids were less than the LOD, total loss of amikacin sulfate was recorded as less than the product of 0.8 µg/mL times the fluid volume lost in the egress fluids. The lower LOD for the immunoassay was 0.8 µg/mL. Concentrations not detected because they were less than the LOD of the assay were recorded as < 0.8 µg/mL.
Statistical analysis was performed with generalized estimating equations to model time and group differences in amikacin sulfate concentrations with repeated measures. All analyses were performed with a commercially available statistical software program. Significance was set at $P < 0.05$.

**Results**

Amikacin sulfate concentrations in the distal interphalangeal joint—Median and range values of amikacin sulfate concentrations in synovial fluids from the distal interphalangeal joint at times T5 and T30 for the pneumatic tourniquet and Esmarch tourniquet groups were recorded (Table 1). The amikacin sulfate concentrations in the pneumatic tourniquet and Esmarch tourniquet groups were significantly different at T5 ($P = 0.041$) and T30 ($P = 0.002$). Amikacin sulfate concentrations in the distal interphalangeal joint did not exceed 100 µg/mL in any horses in the pneumatic tourniquet group.

Amikacin sulfate concentrations in the digital vein—Serum concentrations in digital vein samples were significantly ($P < 0.001$) greater in the Esmarch tourniquet group than in the pneumatic tourniquet group. There was no significant difference ($P = 0.739$) among time points (Table 1).

Amikacin sulfate concentrations in the systemic circulation—Serum amikacin sulfate concentrations in jugular vein samples were significantly ($P \leq 0.011$) greater in the pneumatic tourniquet group, compared with the Esmarch tourniquet group (Table 1). Amikacin sulfate could be detected in samples from the jugular vein of all horses at all times when the pneumatic tourniquet was used, and the concentration was often less than the LOD (0.8 µg/mL) when the Esmarch tourniquet was used. Horses in trials 10 and 11 had detectable concentrations of amikacin sulfate in the jugular vein serum of 0.94 and 0.8 µg/mL, respectively, at T0.

**Discussion**

Our hypothesis, that significant loss of amikacin sulfate in the lavage fluid occurs from horses simultaneously undergoing distal limb IVRLP, was not proven. The Esmarch tourniquet significantly reduced amikacin sulfate from entering the systemic circulation, compared with the pneumatic tourniquet. We believe the losses of amikacin sulfate in the egress lavage fluids were greater for the Esmarch tourniquet group than the pneumatic tourniquet group because there was more amikacin sulfate in the distal portion of the limb when the Esmarch tourniquet was used. Use of the Esmarch tourniquet also resulted in significantly greater amikacin sulfate concentrations in the distal interphalangeal joint and digital vein samples than did the pneumatic tourniquet. In all 12 trials in 11 horses, no more than 1.6% of the 500 mg of amikacin sulfate administered via IVRLP was found in the egress joint lavage fluid. In all horses, the total amount of fluids instilled into the joint was not completely recovered, and if the total ingress volume had been collected in the egress fluids, the calculated amount of amikacin sulfate loss could have been greater. However, the amount of amikacin sulfate per milliliter of egress fluids was so small that the increase would not be important.

**Table 1**—Median (range) amikacin sulfate concentrations (µg/mL) in blood samples obtained from the jugular vein and digital vein and in synovial fluid from the distal interphalangeal joint following simultaneous IVRLP and metacarpophalangeal joint lavage in horses ($n = 6$ group) by use of a pneumatic or Esmarch tourniquet.

<table>
<thead>
<tr>
<th>Sampling location</th>
<th>Time</th>
<th>Pneumatic tourniquet</th>
<th>Esmarch tourniquet</th>
</tr>
</thead>
<tbody>
<tr>
<td>Distal interphalangeal joint</td>
<td>T5</td>
<td>62.1 (12.1–33.0)</td>
<td>49.7 (&lt; 0.8–198)</td>
</tr>
<tr>
<td></td>
<td>T30</td>
<td>29.1 (1.2–49.3)</td>
<td>311 (45.1–1960)</td>
</tr>
<tr>
<td></td>
<td>T5</td>
<td>1,002 (9.1–4,570)</td>
<td>2,582 (60.5–12,008)</td>
</tr>
<tr>
<td>Digital vein</td>
<td>T10</td>
<td>927 (12.6–3,054)</td>
<td>4,896 (3,869–6,063)</td>
</tr>
<tr>
<td></td>
<td>T20</td>
<td>547 (14.3–4,500)</td>
<td>5,208 (4,241–12,119)</td>
</tr>
<tr>
<td></td>
<td>T30</td>
<td>391 (5.4–1,115)</td>
<td>4,327*</td>
</tr>
<tr>
<td>Jugular vein</td>
<td>T5</td>
<td>5.3 (1.8–3.9)</td>
<td>&lt; 0.8 (&lt; 0.8–3.9)</td>
</tr>
<tr>
<td></td>
<td>T10</td>
<td>3.3 (1.8–6.7)</td>
<td>&lt; 0.8 (&lt; 0.8–2.0)</td>
</tr>
<tr>
<td></td>
<td>T20</td>
<td>2.9 (1.3–5.0)</td>
<td>&lt; 0.8 (&lt; 0.8–1.6)</td>
</tr>
<tr>
<td></td>
<td>T30</td>
<td>2.1 (1.2–4.5)</td>
<td>&lt; 0.8 (&lt; 0.8–0.9)</td>
</tr>
</tbody>
</table>

*Numbers 5 to 30 indicate interval (minutes) after beginning of IVRLP.
*Only 1 sample was obtainable from the digital vein at T30 in the Esmarch tourniquet group. Limit of detection of the assay was 0.8 µg/mL.

$T =$ Time.
Because of the constant lavage of the metacarpophalangeal joint, amikacin sulfate entering the joint following IVRLP was diluted, and low concentrations were measured in the egress fluids. On the basis of adequate concentrations of amikacin sulfate obtained in the distal interphalangeal joint, we believe it is logical that other tissues around the joint such as the joint capsule, subchondral bone, and collateral ligaments were adequately perfused with amikacin sulfate. Bacteria in septic joints often localize in periarticular structures.24 One advantage of IVRLP is the ability to drive antimicrobial drugs into the periarticular structures. Because of the study design and desire to perform minimally invasive procedures on horses in a survival study, sampling of the periarticular structures around the metacarpophalangeal joint to directly measure amikacin sulfate concentrations was not done. Because we did not know whether the IVRLP was effective when performed simultaneously with joint lavage, 250 mg of amikacin sulfate was injected into the metacarpophalangeal and distal interphalangeal joints at the termination of the study as prophylaxis for postoperative joint infection. In our clinical practice, we routinely administer selected antimicrobial drugs directly into the infected joint space after joint lavage is completed to ensure immediate high intra-articular concentrations of the drug.

All synovial fluid concentrations of amikacin sulfate recorded in the distal interphalangeal joint of the horses in the Esmarch tourniquet group were greater than the reported MIC of 16 μg/mL for the most common equine pathogens susceptible to amikacin sulfate.6,10 The concentrations of amikacin sulfate in the distal interphalangeal joint synovial fluid from the Esmarch tourniquet group were similar to the concentrations obtained in the study performed by Butt et al21 in which IVRLP was performed alone. The maximal concentration of amikacin sulfate in the that study ranged from 322 to 1,791 μg/mL. This further supports the limited loss of the amikacin sulfate in the lavage fluids in the present study. The antimicrobial activity of aminoglycosides is concentration dependent, and a peak concentration of 10 times the MIC is desired to achieve optimal bactericidal activity.21 In the present study, none of the horses in the pneumatic tourniquet group achieved 10 times the MIC for amikacin sulfate in the distal interphalangeal joint at any time. In the Esmarch tourniquet group, all but 1 horse (trial 7) had concentrations of amikacin sulfate > 10 times the MIC. The horse in trial 7 had digital vein serum concentrations of amikacin sulfate > 700 times the MIC. We cannot explain why the high concentration of amikacin sulfate in the vein did not perfuse in higher concentrations into the distal interphalangeal joint. Other authors have indicated that joint lavage and IVRLP should not be performed simultaneously, to avoid antimicrobial loss and the potential for increased toxicity.6,21 On the basis of the results obtained in the present study, we believe this concern is not warranted.

Pneumatic tourniquets are generally set so the pressure is at least 100 mm Hg greater than the systolic blood pressure, to prevent bleeding. Pneumatic tourniquets are usually safe if used for < 120 minutes.27 Pneumatic tourniquets may induce an ischemia-reperfusion injury with potentially harmful local and systemic consequences. The complications of tourniquet application include postoperative swelling, delay of recovery of muscle power, compression neuropraxia, wound hematomata with the potential for infection, vascular injury, tissue necrosis, and compartment syndrome.28 Despite the potential harmful effects, the pneumatic tourniquet has been considered to be safer than an Esmarch tourniquet because of the large cuff that contours to the limb uniformly. No adverse effects are reported in horses with pneumatic tourniquets set to pressure values from 300 to 500 mm Hg.10,20–21 In the present study, the pneumatic tourniquet pressure was set at 420 mm Hg on the basis of personal experience with use of the tourniquet for control of hemorrhage in anesthetized horses and published data indicating that regional IV perfusion in standing horses performed with a pneumatic tourniquet placed proximally to the carpus and a pressure of 300 mm Hg in the cuff resulted in losses of antimicrobial drugs into the systemic circulation.22 We thought that the higher cuff pressure of 420 mm Hg would prevent leakage. During the experimental procedures, motion of the horses was inevitable. The leakage of the perfusate containing amikacin sulfate into the systemic circulation during use of the pneumatic cuff may be attributable to motion in the limb and leakage under the tourniquet as the limb was loaded and unloaded. It was noted that the pressure in the cuff of the pneumatic tourniquet reset frequently to the selected pressure when the horses were moving their limbs. The data are consistent with a human study33 in which a pneumatic tourniquet set at 400 mm Hg on the forearm did not always prevent leakage of contrast media during venography. We did not test the tourniquet in other positions on the limb or at other pressures. It is possible the pneumatic tourniquet would have been more efficacious if placed proximal to the carpus over the forelimb musculature instead of the nonmuscular and relatively unsymmetrical proximal portion of the third metacarpal bone or if the pressure was set to the maximum pressure of 500 mm Hg.

On the basis of comparative data in dogs,18 the pressure obtained under the Esmarch tourniquets in horses may be substantially higher than the pressure under pneumatic tourniquets. The pressure under the Esmarch tourniquet was not measured in the present study. Esmarch tourniquets prevented detectable concentrations of amikacin sulfate from entering the systemic circulation in a previous study.24 In that study, tourniquets were centered on the proximal metacarpal region, gauze sponges were placed over the neurovascular bundles on each side of the limb, and the full length of the tourniquet was used. Results of the present study confirmed that Esmarch tourniquets are a reliable way to minimize the amount of amikacin sulfate entering the systemic circulation in standing horses. However, 2 horses in the study had detectable systemic concentrations of amikacin, indicating that the tourniquet was not completely effective. This may be attributable to the application technique used for the Esmarch tourniquet. The safe duration of application for Esmarch tourniquets has not been established in horses. Clinical experience in horses suggests that Esmarch tourniquets generally do not cause complications if removed...
within 2 hours. Standard recommendations for the duration of regional limb perfusion do not exceed 30 minutes. Esmarch tourniquets are efficacious when placed at the proximal portion of the metacarpon on anesthetized and standing horses, are widely used in clinical practice, and are less expensive than pneumatic tourniquets.

The present study had some limitations. All horses had clinically normal joints that were not inflamed because of sepsis; loss of amikacin sulfate from lavage of inflamed joints may be greater. Because of some subcutaneous fluid leakage of the lavage fluid from around the needles, some of the digital vein serum samples were not obtainable because the swelling collapsed the digital vein. The fluorescence polarization immunoassay used to determine the amikacin sulfate concentration detected a low concentration of amikacin sulfate systematically in 2 horses at T0 before injection of amikacin sulfate into the limb. Horses used in this study had not received amikacin sulfate or other medications within the 12 months preceding their inclusion in this study. The small amount of amikacin sulfate reported in these samples reflected limitations of the fluorescence polarization immunoassay, the high degree of variability in this assay, and evidence of false-positive results. The fluorescence polarization immunoassay is an automated system that requires human serum standards provided by the manufacturer for calibration. Composition of equine serum may alter the specificity of the assay. False-positive results have been reported elsewhere.

On the basis of results of this study, we believe that it is safe and efficacious to perform IVRLP with amikacin sulfate simultaneously with joint lavage in horses, and losses of amikacin sulfate in the egress lavage fluids are minimal. Losses of amikacin sulfate and other antimicrobial drugs in the egress fluids from IVRLP of horses with septic joints are also likely to be negligible. Future studies should be done to confirm the negligible loss of antimicrobial drugs in infected joints. We presently perform IVRLP simultaneously with joint lavage for treatment of septic arthritis in horses. Minimally invasive procedures such as arthroscopy done simultaneously with IVRLP may not decrease effectiveness of IVRLP in achieving high concentrations of antimicrobial drugs in periarticular tissues.

We recommend using the Esmarch tourniquet for IVRLP in standing horses. The Esmarch tourniquet is more effective than a pneumatic tourniquet set at 420 mm Hg in standing horses, is easier to apply, and is less expensive. The Esmarch tourniquet did not cause any complications when applied for 30 minutes.

References


