Effectiveness and adverse effects of the use of apomorphine and 3% hydrogen peroxide solution to induce emesis in dogs

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Objective—To determine the effectiveness and adverse effects of apomorphine and 3% hydrogen peroxide solution used for emesis in dogs.

Design—Prospective observational study.

Animals—147 dogs that received apomorphine (IV or placed in the conjunctival sac) or 3% hydrogen peroxide solution (PO) to induce emesis after exposure to toxic agents.

Procedures—Data regarding signalment; agent information; type, dose, route, and number of emetic administrations; whether emesis was successful; number of times emesis occurred; percentage of ingested agent recovered; and adverse effects were collected via telephone during American Society for the Prevention of Cruelty to Animals Animal Poison Control Center operations and stored in a database for analysis. Mann-Whitney and Fisher exact tests were used to evaluate emetic success rates.

Results—Apomorphine and 3% hydrogen peroxide solution successfully induced emesis in 59 of 63 (94%) and 76 of 84 (90%) of dogs, respectively. Mean time to onset of emesis after the first dose of emetic was 14.5 and 18.6 minutes when hydrogen peroxide (n = 37) and apomorphine (31) were used, respectively, with mean durations of 42 and 27 minutes, respectively. Mean estimates for recovery of ingested agents were 48% for hydrogen peroxide and 52% for apomorphine. Adverse effects were reported in 16 of 112 (14%) dogs for which information was available.

Conclusions and Clinical Relevance—3% hydrogen peroxide solution and apomorphine effectively induced emesis in dogs when used as directed. Emesis occurred within minutes after administration and helped recover substantial amounts of ingested agents. Adverse effects of both emetics were considered mild and self-limiting. (J Am Vet Med Assoc 2012;241:1179–1184)
the authors’ knowledge, there are no data describing the effectiveness and adverse effects of these treatments in dogs.

**Materials and Methods**

Data collection—Clinical data were collected and stored in the APCC electronic medical records database and later retrieved for analysis. The APCC is a 24-hour service that provides information to animal owners and veterinarians via telephone in animal poisoning cases throughout the United States and Canada. Upon receiving a call, if no contraindication exists and the patient was exposed to an agent with a potential risk of causing severe clinical signs at the estimated dose, APCC veterinary staff members recommend emesis. Instructions to induce emesis were given via telephone, and data were collected at the time of the initial call or during follow-up. Information was collected retrospectively if the attending veterinarian or dog owner administered the emetics and contacted APCC veterinary staff afterward. Information on the dose of emetic administered and compliance by owners or attending veterinarians was recorded during follow-up.

Study inclusion criteria were that the APCC was contacted for assistance in the case between January 11, 2008, and April 19, 2009; the case involved acute ingestion of a potentially toxic agent by a dog; apomorphine or 3% hydrogen peroxide solution was administered to induce emesis; and the type of emetic used and success of emetic induction (whether the dog vomited at least once after emetic administration) were known. Only cases involving 1 exposure were included in the present study.

Additional data collected included the following when available: signalment (species, breed, age, sex, and body weight of the dog); information regarding the toxic agent (name or type, time of ingestion, approximate amount ingested, and whether exposure was observed or other evidence indicative of exposure was found); details of emesis (dose and route of emetic administration, number of times the emetic was administered [once, twice, or 3 times], interval between exposure and emetic administration [minutes], interval between the first emetic administration and onset of emesis [minutes], approximate number of times the dog vomited, approximate duration of emesis [minutes]); whether emesis was recommended by the APCC staff member or attending veterinarian or was attempted by the owner prior to consultation; whether any adverse effects were observed; and the estimated percentage of the ingested agent that was recovered if induction of emesis was successful. A percentage was assigned to the amount of agent recovered in the emesis if a measurable amount was detected on visual inspection and a known amount was ingested. Reporting of adverse effects was solicited with an open-ended question.

**APCC emetic administration recommendations**—Recommendations regarding emetic use were made in accordance with standard APCC procedures. The APCC veterinary staff members generally recommend that clinicians or owners induce emesis if the time after exposure is approximately 2 hours or less and no clinic signs of toxicosis are present. Before recommending emesis, APCC staff follows guidelines to help ensure that emesis is induced with little risk to the patient. The APCC staff members typically recommend that dogs be fed a small meal such as bread or canned dog food unless the dog has eaten during the 2 hours prior to emetic administration. Contraindications to inducing emesis in dogs include clinical signs other than mild lethargy (ataxia, coma, seizures, and hyperactivity); preexisting health conditions that require medication or veterinary intervention; ingestion of corrosive materials, hydrocarbons, or petroleum distillates; the patient having already vomited; conditions in which there is an increased risk of aspiration due to emesis; and exposure to drugs with substantial antiemetic properties, given that antiemetic drugs could potentially alter emesis success.

The recommended dose for induction of emesis with 3% hydrogen peroxide solution was 2.2 mL/kg (1 mL/lb), PO, to a maximum of 45 mL/dog, repeated once after 10 to 15 minutes if vomiting did not occur. The recommended dose of apomorphine hydrochloride was 0.03 mg/kg (0.014 mL/lb), IV, once, or a crushed tablet dissolved in saline (0.9% NaCl) solution, instilled in the conjunctival sac, and rinsed away with water or saline solution after emesis (resulting in a dose to effect).

**Statistical analysis**—Mean, SD, median, and range values were calculated for dogs grouped according to age, body weight, time from administration of the first dose of emetic to onset of emesis, duration of emesis, and number of times emesis occurred. Percentages were calculated for the approximate amount of the potentially toxic agent recovered in the vomitus (determined on the basis of visual inspection), development of adverse effects (determined on the basis of owner or veterinarian reports), and success rate of emetic administration. Mann-Whitney tests were used for comparison of continuous (nonparametric) variables, and Fisher exact tests were used to compare categorical data.

**Results**

The records of 147 dogs treated with apomorphine or 3% hydrogen peroxide solution following exposure to potentially toxic substances were included in the study. Hydrogen peroxide was administered to 84 dogs (37 males, 44 females, and 3 of unknown sex), and apomorphine was administered to 63 (34 males, 28 females, and 1 of unknown sex). There were no significant differences between dogs that received hydrogen peroxide and apomorphine for the following variables: body weight ($P = 0.79$), age ($P = 0.86$), time to onset of emesis ($P = 0.74$), duration of emesis ($P = 0.62$), number of times vomiting occurred ($P = 0.85$), and percentage of agent recovered ($P = 0.81$).

Not all information was available for all dogs. Information regarding emesis recommendations was available for 143 dogs. The APCC veterinary staff recommended emesis for 68 dogs; attending veterinarians made this recommendation for 39 dogs, and owners attempted to induce emesis in 16 dogs prior to consultation. The success of emesis induction was not significantly ($P = 0.06$) affected by this variable. Emesis was
successfully induced with hydrogen peroxide or apomorphine in 135 of 147 (92%) dogs (Table 1).

Of 135 dogs in which emesis was successfully induced, the amount of ingested material recovered was estimated in 116, of which 82 (71%) had successful recovery of some amount of the ingested agent. The type of emetic used was not significantly (P = 0.14) associated with recovery of any ingested agent (yes or no).

Exposure to a toxic agent was observed (ie, someone saw the exposure) in 37 of 143 (26%) dogs, evidenced (ie, exposure was not observed, but evidence indicative of exposure, such as a chewed container, was found) in 92 (64%), and suspected (ie, no conclusive evidence was found, but exposure was suspected) in 14 (10%).

The mean dose of 3% hydrogen peroxide solution was 2.39 ± 1.64 mL/kg (1.09 ± 0.75 mL/lb; median, 1.98 mL/kg [0.9 mL/lb]; range, 0.55 to 7.05 mL/kg [0.25 to 3.2 mL/lb]).

The mean dose of apomorphine administered IV was 0.035 ± 0.01 mg/kg (0.016 ± 0.0045 mL/lb; median, 0.03 mg/kg; range, 0.026 to 0.054 mg/kg [0.012 to 0.025 mg/lb]). Success of emesis induction (yes or no) was not significantly associated with the type of emetic used (P = 0.56) or with the dose of either emetic (P = 0.26).

No significant (P = 0.24) association was detected between success of emesis induction and time between exposure and administration of apomorphine or 3% hydrogen peroxide solution.

Whether adverse effects developed was known for 70 dogs that received 3% hydrogen peroxide solution and 42 that received apomorphine. Adverse effects attributed to emetic administration were reported in 10 (14%) dogs that received hydrogen peroxide and in 6 (14%) that received apomorphine. Of the 16 patients with adverse effects, most (10) only developed lethargy or persistent signs of nausea. The occurrence of an adverse effect (yes or no) was significantly (P = 0.01) associated with the number of times emetics were administered; 1 dog that received hydrogen peroxide 3 times had an adverse effect (1/1), compared with 5 of 49 (10%) and 0 of 10 when the same emetic was administered once or twice, respectively. Occurrence of an adverse effect was also significantly (P = 0.003) associated with the individual who recommended or deemed emesis necessary. When emesis was recommended by the APCC veterinary staff, adverse effects were reported in 5 of 45 (11%) dogs, compared with 2 of 8 when the dog’s owner induced emesis prior to consultation or 2 of 4 when the attending veterinarian induced emesis prior to consultation.

**Apomorphine**—Emesis was successfully induced with apomorphine in 59 of 63 (94%) dogs. Apomorphine (administered once in all dogs) induced emesis in 20 of 22 (91%) dogs when placed in the conjunctival sac and in 26 of 26 (100%) dogs when administered IV. Route of administration was unknown in 15 dogs.

Successful induction of emesis was not significantly (P = 0.21) associated with the route of administration.

Mean time to onset of emesis was 18.6 minutes, and duration was 26.8 minutes. The mean number of times emesis occurred was 3.0. Some amount of the ingested agent was recovered in 38 of 49 (78%) dogs, and the mean estimated percentage of ingested agent recovered (in 33 dogs for which a value was determined) was 52%. Adverse effects were reported in 6 of 42 (14%) dogs for which this variable was recorded. Reddened eyes were reported in 1 of 17 dogs in which apomorphine was placed in the conjunctival sac. Other adverse effects included lethargy, persistent signs of nausea, and hypersalivation. Occurrence of adverse effects was not significantly (P = 0.40) associated with the route of administration. Apomorphine successfully induced emesis a mean of 108 minutes after exposure to a potentially toxic agent. The time between exposure and emetic administration was not significantly (P = 0.90) associated with recovery of the agent (yes or no) in the emesis.

**Hydrogen peroxide**—Emesis was successfully induced with 3% hydrogen peroxide solution in 76 of 84 (90%) dogs. The mean time to onset of emesis was 14.5 ± 19.6 minutes, and duration was 42 ± 102 minutes. Hydrogen peroxide was administered a mean of 1.2 ± 0.4 times, and most (49/57 [86%]) dogs vomited after receiving 1 dose. The mean number of times emesis occurred was 4.1. Some amount of the ingested agent was recovered in 44 of 67 (66%) dogs, and the mean estimated amount of ingested agent recovered was 48%.

Adverse effects were reported in 10 of 70 (14%) dogs and included diarrhea, lethargy, and protracted signs of nausea and vomiting. Hydrogen peroxide successfully induced emesis a mean of 61 minutes after exposure to a potentially toxic agent. The time between exposure and recovery of any ingested agent (yes or no) in the emesis.

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**Table 1**—Summary data for dogs in which emesis was successfully induced via administration of 3% hydrogen peroxide solution (76/84; 32 males, 41 females, and 3 of unknown sex) or apomorphine (59/63; 31 males, 27 females, and 1 of unknown sex).

<table>
<thead>
<tr>
<th>Variable</th>
<th>3% hydrogen peroxide solution</th>
<th>Apomorphine</th>
</tr>
</thead>
<tbody>
<tr>
<td>No. of dogs</td>
<td>Mean ± SD</td>
<td>No. of dogs</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>76</td>
<td>58</td>
</tr>
<tr>
<td>Age (y)</td>
<td>76</td>
<td>59</td>
</tr>
<tr>
<td>Time to onset of emesis (min)</td>
<td>37</td>
<td>31</td>
</tr>
<tr>
<td>Duration of emesis (min)</td>
<td>28</td>
<td>25</td>
</tr>
<tr>
<td>No. of times emesis occurred</td>
<td>44</td>
<td>35</td>
</tr>
<tr>
<td>Estimated amount of ingested agent recovered (%)</td>
<td>49</td>
<td>33</td>
</tr>
</tbody>
</table>

Information was not available for every variable in all dogs. Among dogs in which emesis was successful, hydrogen peroxide was administered once in 49 dogs, twice in 7 dogs, and 3 times in 1 dog; the number of doses was not reported for 19. Apomorphine was administered once in 59 dogs. Mean (total) doses of hydrogen peroxide and apomorphine were 2.39 mL/kg (1.09 mL/lb; PO) and 0.035 mg/kg (0.016 mg/lb; IV), respectively; the dose of apomorphine administered conjunctivally could not be estimated. The time to onset of emesis was measured from administration of the first dose of emetic.
and emetic administration was not significantly \( (P = 0.60) \) associated with recovery of the agent (yes or no) in the emesis.

**Discussion**

The results of the study reported here show that in a clinical or home setting, the mechanistically different emetics apomorphine and 3% hydrogen peroxide were equally effective at inducing emesis in dogs. Emesis was successfully induced in 76 of 84 (90%) dogs with 3% hydrogen peroxide administered orally and in 59 of 63 (94%) dogs with apomorphine administered IV or in the conjunctival sac.

Apopomorphine is a centrally acting nonselective dopamine agonist used as an emetic mainly in dogs. It can be administered orally, IV, SC, or IM or directly applied to conjunctival membranes. Because of its high first-pass hepatic metabolism and slow absorption after oral administration, it is not useful to administer apomorphine orally when ingestion of a toxin is suspected. Vomiting is induced by apomorphine through direct stimulation of the chemoreceptor trigger zone of the brain. Apomorphine also has a depressant effect on the vomiting center in the medulla. Therefore, if apomorphine does not successfully result in emesis after the first administration, it is not likely to have a better result after a second administration. Although the success rate of emesis for apomorphine following IV administration was slightly higher (26/26 [100%]), conjunctival application was also successful (20/22 [91%]), and these rates were not significantly different. Hydrogen peroxide, readily available as a 3% solution from a drug or a grocery store, was administered orally to dogs of the present study. Hydrogen peroxide is not a centrally acting emetic. It is thought to induce emesis through local stimulation of nerve endings in the GI tract transmitted to the vomiting center through vagal nerve. Most dogs of the present study that received hydrogen peroxide (49/57 [86%]) vomited after receiving 1 dose. Mean doses of hydrogen peroxide (2.39 mL/kg [range, 0.53 to 7.05 mL/kg, PO] and apomorphine (0.035 mg/kg [range, 0.026 to 0.054 mg/kg], IV) used in the present study varied from the recommended doses by the APCC (2.2 mL/kg for hydrogen peroxide, not to exceed 45 mL, and 0.03 mg/kg for apomorphine). Despite the minor dose differences, success rates for both emetics were high.

Improper mixing, storage, or stability of apomorphine in solution or availability of apomorphine in different forms (compounded for IV use or as tablets) or from different manufacturers could influence the efficacy of this medication. This type of information was not collected in the present study. Similarly, accurate dosing information for ocular administration is difficult to obtain and could not be evaluated in the present study. Despite these concerns, apomorphine was still considered very effective in inducing emesis after IV administration or placement in the conjunctival sac. We assumed that apomorphine was mixed, stored, and used according to label directions. Similarly, the use of hydrogen peroxide beyond its expiration date could also reduce its ability to induce emesis because it may potentially produce less than the expected degree of GI irritation. The APCC staff routinely asks for expiration information before recommending emesis with 3% hydrogen peroxide, although this information was not recorded in the present study.

Whether emesis was recommended by the APCC veterinary staff member or attending clinician or was attempted by the owner prior to consultation did not significantly \( (P = 0.06) \) influence the success rates of emesis induction. However, the owners’ attempts at inducing emesis in dogs by administration of 3% hydrogen peroxide solution without consulting a veterinarian could influence results in some situations; administration of an incorrect dose, improper number of doses, and extended duration between time of exposure and emetic administration can influence these results, in addition to the previously described contraindications.

To collect data in a consistent manner for the present study, APCC staff members recommended emesis within 2 hours after exposure, although this can vary depending on the dose and type of potentially toxic agent involved. Before emesis is attempted, APCC staff members typically recommend that dogs be fed a small meal such as bread or canned dog food unless the dog has already eaten within the preceding 2 hours. Emesis is likely to be more successful if food is present in the stomach. Moreover, presence of food in the stomach can act as a recovery vehicle for ingested agents. Without sufficient food present in the stomach, only small amounts of the ingested agent will be recovered. It is possible that presence of food in the stomach can also reduce the effectiveness of 3% hydrogen peroxide solution, which produces its effect by irritating the GI tract. Information on whether food was given prior to inducing emesis was not collected in the study reported here. The authors believe the lack of this information was not likely to be important because of the described success rates following emetic administration.

The mean time to onset of emesis (after administration of the first dose) was similar for both emetics (14.5 minutes for 3% hydrogen peroxide solution and 18.6 minutes for apomorphine) and sometimes occurred ≤ 1 minute after administration. Although mean duration of emesis appeared to be slightly longer following hydrogen peroxide administration (42 minutes) than after apomorphine administration (27 minutes), this difference was not significant \( (P = 0.62) \). Similarly, the mean number of times dogs vomited after administration of hydrogen peroxide appeared to be slightly higher but was not significantly different from that following apomorphine administration. Duration of vomiting or the number of vomiting episodes after hydrogen peroxide administration could potentially be increased in some patients by continuous irritation from oxygen bubbles formed in the GI tract when hydrogen peroxide dissociates into oxygen and water. To reduce the risk of aspiration and to attain maximum benefit, administration of activated charcoal, if needed, must be postponed until vomiting has stopped completely after administration of apomorphine or hydrogen peroxide.

When emesis was successful, the attending veterinarian or owner reported recovery of some amount of the ingested agent (on the basis of visual inspection) for...
82 of 116 (71%) dogs. Mean estimates for the amount of agent recovered (48% for hydrogen peroxide and 52% for apomorphine) were not significantly different between the 2 types of emetic. Similar findings describing recovery of experimentally administered materials have been reported following apomorphine or ipecac-induced emesis in dogs.6,7 These findings are particularly important because they support the idea that the induction of emesis in dogs can help to recover substantial amounts of potentially toxic agents after ingestion. The recovery of ingested agents depends on several factors, such as the interval between potential toxin exposure and administration of the emetic, time to onset of emesis, and amount and formulation of the ingested agent. Although some amount of these products was likely to have been absorbed, the recovery of ingested agents may potentially have been greater than that reported here because all cases of exposures to the agent were not confirmed (thus, some were cases of suspected ingestion). Also, prior to emesis, some of the ingested material may have begun to dissolve and may not have been visible in the vomitus. Due to their colors, sizes, and formulations, some ingested agents may not have been easily identifiable in vomitus. The presence of food may also have made the identification of the ingested agent in the vomitus difficult. A percentage of recovered agent was assigned only when a known amount of the product was ingested and a measureable amount (by visible inspection) of material was detected in the vomitus. Information on the amounts recovered was subjective. As in any other observational study, this was a limiting factor in the present study. However, SE and SD are reduced as sample size increases, and the use of larger sample sizes, as in the study reported here (n = 82), increases the power of the study to detect differences.

Partial recovery of an ingested agent through emesis or other decontamination procedures (gastric lavage or administration of activated charcoal) does not necessarily equate to an improvement in outcome for the patient. Several other important factors, such as the nature and dose of the agent ingested, the animal’s preexisting health conditions, timing of the emesis or gastric lavage, administration of activated charcoal (single or repeated doses, with or without cathartics), IV administration of fluids or other supportive measures, availability and timing of administration of a specific reversal agent if needed (eg, atropine, naloxone, or flumazenil), and financial constraints can influence decision making for treatment and thus alter patient outcome. All patients that undergo such decontamination processes must be treated or monitored further as required for signs of intoxication.8

At follow-up, owners or veterinarians were asked whether they noticed any adverse effects after emesis was induced. The proportion of patients reported to have adverse effects after administration of 3% hydrogen peroxide solution (10/70 [14%]) was similar to that reported after apomorphine administration (6/42 [14%]). Lethargy and persistent signs of nausea were the most commonly reported adverse effects. These effects were mild and self-limiting. More serious adverse effects such as bloat, GI ulcers, hematemesis, or hematochezia were not reported. Protracted vomiting (30 times) after hydrogen peroxide administration was reported in 1 dog. Ingestion of 3% hydrogen peroxide solution or more highly concentrated forms ranging from 10% to 40% is known to cause severe pathological changes in the GI tract, gas embolism, and death in humans.9–11 The most commonly reported adverse effects following apomorphine administration in dogs are respiratory and CNS depression.9 Protracted vomiting due to apomorphine, although not reported in the present study, can be reversed with a dopamine antagonist such as metoclopramide (0.1 to 0.4 mg/kg [0.045 to 0.18 mg/lb], SC or IM). Substantial CNS depression from apomorphine, if detected, can be reversed with naloxone (0.01 to 0.02 mg/kg [0.0045 to 0.009 mg/lb], IV). Rinsing of the conjunctival sac with saline solution or water to remove apomorphine residues after successful emesis is routinely performed; however, this information was not collected in the present study. It is possible that some of the adverse effects reported in the present study were unrelated to the use of emetics and were attributable to effects of the ingested agent. The frequency of adverse events is highly dependent on the individual who was induced. The proportion of patients reported to notice any adverse effects after emesis was induced 2 times was significantly (P = 0.01) associated with the number of times the emetic was administered (once, twice, or 3 times). These results should be interpreted cautiously. In the event that emesis with 3% hydrogen peroxide was not successful the first time, APCC staff recommends emesis 1 more time but very rarely a third time (emesis was induced 3 times in only 1 dog of the present study, and this was due to noncompliance). However, in the authors’ experience, an increase in the occurrence of adverse events is possible when emesis is attempted a third time. The occurrence of adverse effects was significantly (P = 0.003) associated with whether emesis was recommended by the APCC veterinary staff member or attending clinician or was attempted by the owner prior to consultation. This dependence of adverse effects on the individual who recommended inducing emesis may have been related to the fact that the APCC veterinary staff follows strict contraindication guidelines based on our experience to help prevent the occurrence of adverse effects. Owners who do not consult with the APCC or their veterinarian before inducing emesis may be more likely to administer incorrect doses, induce emesis too many times after the first or second attempt was unsuccessful, or induce emesis in a patient that ingested an agent that has antiemetic effects, is caustic, is a CNS stimulant, or is a hydrocarbon or petroleum distillate.

Despite some limitations, the results of the present study indicate that the 2 mechanistically different emetics evaluated are highly effective at inducing emesis in dogs rapidly after administration. The use of 3% hydrogen peroxide solution or apomorphine placed in the conjunctival sac or administered IV can be beneficial when used appropriately in a potential poisoning situation because prompt emesis can recover a substantial amount of the ingested agent from the GI tract with typically mild adverse effects. To ensure no contraindications exist and to maximize safety and efficacy, it is advisable that pet owners consult a veterinary professional before the use of 3% hydrogen peroxide solution for emesis in dogs at home.
References


From this month’s AJVR

Evaluation of inflammatory and hemostatic surgical stress responses in male cats after castration under general anesthesia with or without local anesthesia

Elena R. Moldal et al

Objective—To characterize acute inflammatory and hemostatic surgical stress responses following castration in cats and to evaluate whether the addition of local anesthesia to the anesthetic protocol attenuates these responses.

Animals—39 male cats.

Procedures—Cats undergoing castration were randomly assigned to 2 groups: both groups underwent surgery with general anesthesia, and 1 group additionally received a local anesthetic (lidocaine [2.0 mg/kg in total, divided intratesticularly and SC]) prior to incision. Blood samples were collected after anesthetic induction (baseline) and 1, 5, and 24 hours later. Thromboelastography and coagulation variables (activated partial thromboplastin time [aPTT] and prothrombin time [PT]) were analyzed; fibrinolysis was assessed with plasma D-dimer concentrations. The acute-phase response was evaluated via measurement of plasma fibrinogen and serum amyloid A (last time point, 28 hours) concentrations. Hematologic variables were analyzed at baseline and 1, 5, and 24 hours later.

Results—Evidence of hemostatic and inflammatory activation after surgery was detected in both groups. Maximum amplitude and G (global clot strength) were significantly increased at 24 hours, and significant, but not clinically relevant, decreases were detected in aPTT at 5 and 24 hours and in PT at 24 hours, compared with baseline values. Serum amyloid A concentrations were significantly higher at 24 and 28 hours than at baseline, and plasma fibrinogen concentration was significantly increased at 24 hours; WBC and RBC counts and Hct were significantly increased at multiple time points. No differences between groups were detected for any variables.