Antifreeze ingestion by dogs and rats: influence of stimulus concentration

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Abstract — Ingestive responses of 50 rats and 4 gastric-cannulated dogs to ethylene glycol-based antifreeze (AF) were found to be inversely related to concentration. The antifreeze was never preferred to water. Do thirsty animals that encounter weak AF solutions drink them solely for their water content, water-related oral tactile sensations, or both, being inadvertently poisoned in the process?

Résumé — Ingestion d’antigel par des chiens et des rats : influence de la concentration comme stimulus. L’ingestion d’un antigel à base d’éthylène glycol (AG) par 50 rats et 4 chiens pourvus de canules gastriques s’est révélée être inversement proportionnel à la concentration. Jamais l’antigel n’a été préféré à l’eau. Les animaux assoiffés en présence d’un AG à faible teneur le consommaient-ils uniquement pour son contenu en eau et/ou les stimulus sensoriels associés à l’eau n’auraient-ils pas été émoussés par inadvertance dans le processus?

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Ethylene glycol (EG)-based antifreeze (AF), a toxic agent associated with irreversible damage to renal, hepatic, cardiovascular, and central nervous systems, is ingested by thousands of animals each year. Indeed, next to rodenticides, EG counts for more accidental animal poisonings than any other toxin (1). Wildlife, domesticated pets, and working dogs, including ones used in search and rescue operations (cadaver dogs), commonly succumb to such damage (2–5). Toxic exposure to AF is not confined to animals, however. The American Association of Poison Control Centers reported 5816 human exposures to EG in 2003, of which at least 23 were fatal (6).

While it is commonly reported that AF has an attractive — usually termed sweet — taste to animals, experimental evidence for this is lacking. Indeed, the limited empirical data on this point suggest this is not the case. Thus, in 1 study fewer than 10% of a group of 178 adult mixed-breed dogs initiated lick responses in a 5-minute test to a 50% concentration of AF, a concentration most commonly present in automotive cooling systems (7). Furthermore, in this same study, the lick responses of 5 gastric-cannulated dogs to 50% EG, 50% antifreeze, and 50% propylene glycol were markedly lower than those to water or to a 20% sucrose solution, even under conditions of food and water deprivation.

The purpose of the present set of 2 experiments was to determine the influences of AF concentration on the ingestive responses of thirsty dogs and rats. This work was part of a series of studies designed to test and develop additives of potential use in discouraging pets from drinking AF. Specifically, we sought to determine whether, in fact, lower concentrations of AF than those previously tested are attractive tastants. A wide range of concentrations were evaluated in an effort to better understand the functional relationship, if any, between AF concentration and ingestion.

In the 1st experiment, 4 mixed-breed dogs, obtained from a licensed laboratory animal supplier, were tested. They were housed individually in 2.5- × 4.5-m indoor kennels, maintained at ~ 23°C. Before delivery to the laboratory, the dogs had received screening for heartworms, routine treatments for parasites, and standard inoculations. The daily feeding regimen (1 feeding/d at ~ 1800 h) consisted of measured quantities of compressed meat and meat biproduct patties, dry kibbel dog food (Big Red; Agway, Shippensburg, Pennsylvania, USA), and water. During all phases of the study, as well as during the months following the study, the health of the dogs was monitored closely by a licensed veterinarian. No evidence of EG toxicity was observed. All testing was performed within a veterinary hospital and followed a protocol approved by the hospital’s animal care committee. The housing and care that was administered conformed strictly to published laboratory animal guidelines (8).

After the dogs became habituated to the laboratory situation, each was implanted with a stainless steel gastric cannula that allowed for draining of stomach contents during testing, as well as for flushing the stomach contents after a test session. The operation to implant the cannula was performed under sterile conditions by a licensed veterinarian. Several months after the operations, the dogs were placed on a 23-hour water deprivation schedule. They were not allowed to eat solid foods for 12 h before testing to preclude the possibility of undigested stomach contents blocking the cannula’s opening.

The testing apparatus consisted, in part, of a modified metabolic collection cage fitted with a canine restraining sling. A dish connected to a calibrated burette allowed for easy access to and lapping of the stimulus solution, which was constantly replenished. Ingestants from the stomach passed easily through the fistula, which was...
connected by tubing to a stainless steel waste container. Following testing and flushing the stomach with ~3 L of warmed water, the cannula was closed and the subject was allowed ad libitum access to water for an hour before being placed back on the deprivation schedule.

Initially, the dogs were habituated to the test situation and were allowed, under a 23-hour deprivation period, to sample water from the feeding dish. After several days of such habituation, 14-minute test sessions were employed during which lick responses to each of the following concentrations of AF product (Prestone AF; Prestone Products Corporation, Danbury, Connecticut, USA: 95% to 100% ethylene glycol; 0% to 5% diethylene glycol) were determined: 0 (water), 6.25%, 12.5%, 25%, 50%, and 100%. These sessions occurred on different days and were repeated twice, with the results averaged. Test sessions occurred in the morning of each test day. Each dog received a different order of testing across days, in accord with a counterbalancing sequence derived from Latin square sectors. Lick responses were quantified within 30-second time bins by using hand-operated digital counters.

The results of this experiment are shown in Table 1. It is apparent that as the AF concentration increased, the number of lick responses generally decreased. In only 1 of 4 cases did the amount of licking an AF solution exceed that of water, and only at the lowest AF concentration presented (6.25%). It should be noted that within this small sample of dogs, considerable variation in licking the stimuli was present. For example, dog #2 never licked any of the AF test solutions, whereas dog #4 licked 3 of the 5 solutions on both the 1st and 2nd tests. Nevertheless, on average, a monotonicly increasing order of preference, with the lower concentrations eliciting higher ingestive preferences, was present.

In the 2nd experiment, 50 adult male Long Evans rats were used. A week after arriving at the laboratory, the rats were placed on a 23-hour water deprivation schedule, with laboratory chow (Purina Laboratory Chow; Purina Mills, Richmond, Indiana, USA) available ad libitum. The number of licks given to the AF solutions, as well as to the water solutions on the 3 d prior to the introduction of the AF test, varied among rats. Because of the wide range of licks given to AF solutions and the fact that the underlying data were not normally distributed, the data were subjected to a log_{10} transformation before parametric analysis. In 3 of 50 cases, where no responding occurred, the mean of the rest of the group was assigned to the missing datum point to allow for calculation of logarithms and to minimize the effects of extreme scores on the data.

As can be seen in Figure 1, an inverse relationship between AF concentration and licking responses was observed during the AF trials. In contrast, no such association was noted for water drinking prior to these trials in the same rats. When these data were subjected to an AF concentration group by solution type (water, AF) analysis of variance (ANOVA), with repeated measures on the 2nd factor, significant main effects of AF concentration group [F(9,40) = 6.89, P < 0.000] and solution type [F(1,40) = 109.82, P < 0.000] were noted, as well as a significant interaction between these 2 factors [F(9,40) = 5.30, P < 0.000]. To further explore this interaction, the AF drinking data were subjected to a one-way analysis of variance (ANOVA), with AF concentration as the independent variable. A strong association between AF concentration and lick responses was observed [F(9,40) = 8.80, P < 0.000]. A similar analysis found no differences in water drinking among the 10 groups of rats [F(9,40) = 0.75, P = 0.662]. A series of repeated measures t-tests performed between the water

Table 1. Number of licks in 14-minute test periods with water and 5 concentrations of antifreeze (AF) for 4 gastric cannulated dogs. See text for details

<table>
<thead>
<tr>
<th>Subject number</th>
<th>Water</th>
<th>6.25% AF</th>
<th>12.5% AF</th>
<th>25% AF</th>
<th>50% AF</th>
<th>100% AF</th>
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<td>85</td>
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<td>415</td>
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<td>191</td>
<td>100</td>
<td>11</td>
<td>8</td>
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</tr>
<tr>
<td>Median</td>
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<td>175</td>
<td>52</td>
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and AF conditions within each group revealed that AF was drunk less than water at AF concentrations 18% and higher. Importantly, in no instance did AF drinking significantly exceed water drinking.

The present series of experiments clearly demonstrate that the ingestion of AF depends upon its concentration in both rats and dogs that are water deprived. While previous work has shown that few dogs exhibit an interest in licking AF at concentrations commonly used in motor vehicle cooling systems (~50%), the data of experiment 1 indicate that this is not the case when weaker antifreeze solutions are available. In experiment 2, the amount of AF ingested by water-deprived rats at concentrations below 18% did not differ from the amount of water ingested by these rats under the same deprivation and testing conditions. Taken together, these observations indicate that while readily ingested, even relatively low concentrations of AF are not preferred to water in thirsty rats and dogs. This finding throws into question the commonly held belief that AF is a highly preferred ingestant to such animals.

That being said, dogs and other mammals are commonly poisoned from drinking solutions of AF. The present data indicate that such drinking depends on the concentration of AF solution available (7). Our previous work suggested that water deprivation facilitates the drinking of AF. Other factors, which are not addressed here, may also influence the propensity of mammals to ingest AF. In the case of dogs, this would include subject variables (age, breed, gender, temperament, sensory sensitivity), environmental factors (ambient temperature, housing conditions), and social factors (presence of other dogs, the nature of the owner/animal interactions) (7). It should be noted that the short-term ingestion tests of this study may not generalize to longer-term situations where dogs are confined to a garage or basement where antifreeze might be spilled. Under these circumstances, there may be an increased propensity to explore or ingest novel substances as a result of boredom or anger.

References