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Estimating the Time of Death in Domestic Canines

ABSTRACT: Because 36.1% of U.S. households have dogs, the time of death (TOD) of dogs at crime scenes can be useful to forensic investigators. However, there are few published studies based on postmortem changes in dogs. This study, conducted indoors in still air at approximately room temperature, monitored the postmortem reduction in rectal, liver, brain, and aural temperatures in 16 dogs for 32 h after death. Graphs of temperature reduction were prepared to estimate the TOD of dogs within the first 32 h postmortem. Sex, body mass, and hair coat density did not affect the rate of body temperature reduction, but increased body weight and volume slowed it. Rectal temperature was the most convenient, reasonable site for measuring body temperature. Vitreous humor potassium ion concentration [K^+] was measured in both eyes at *c.* 1.5 and 7 h after death. Both eyes had the same [K^+] when measured simultaneously, and [K^+] increased after death.

KEYWORDS: forensic science, postmortem interval, canidae, canines, dogs, time of death estimation, body temperature, vitreous humor, potassium concentration

Estimating time of death aids law enforcement officers in forensic investigations. Estimating the time of death of human victims comes immediately to mind, but, given that in 2001, 36.1% of human households in the United States owned a dog (1), dogs and other animals found dead at crime scenes can also be useful to investigators. This usefulness usually extends to cases where a human and animal death are coincident, but can also include cases involving the violation of animal abuse and cruelty laws, the death of an animal when killing the animal is itself a crime, and the investigation of crimes against protected wildlife. When dead animals and humans are both found together, their deaths are, of course, not necessarily coincident.

Research in determining time of death has naturally focused on humans, but some has been done on animals. Cox et al. (2) used thigh and nostril temperature measurements and multiple linear regression modeling to estimate the time of death of white-tailed deer (*Odocoileus virginianus*). Hadley et al. (3) used carcass temperature, pupillary diameter, rigor mortis, and multiple linear regression modeling to estimate the time of death in white-tailed deer considering the variables body size, ambient temperature, and carcass handling methods. Johnson et al. (4) showed that potassium ion concentrations increased after death in the vitreous humor of mule deer (*Odocoileus hemionus*). Woolf and Gremillion-Smith (5) used potassium ion concentrations in the vitreous humor of white-tailed deer to estimate time of death.

Hood et al. (6) used core body temperatures and the vitreous humor concentration of several chemicals to predict the time of death in harbor porpoises (*Phocoena phocoena*). BG McLaughlin and PS McLaughlin (7,8) studied the changes in several chemical components of vitreous humor after death in horses (*Equus caballus*) (7), cattle (*Bos taurus*) (8), and pigs (*Sus scrofa domestica*) (8). Finally, Crowell and Duncan (9) studied postmortem changes

in potassium ion concentration of vitreous humor in domestic dogs (*Canis familiaris*).

This report describes temperature reduction in the rectum, liver, brains, and ears of dogs after euthanasia as well as the change in potassium ion concentration in the vitreous humor of their eyes.

Methods

Sixteen adult dogs were euthanized at an animal shelter. After death, dogs were weighed and rectal, liver, brain, and aural temperatures were recorded for *c.* 32 h indoors in still air. Temperatures were recorded using four VIP-T series temperature probes connected to a model OM-CP-QUADTEMP 4-channel continuous temperature data logger. (The probes and data logger were obtained from Omega Engineering Inc., Stamford, CT.) Relative humidity was recorded for the first 7 h of the study using a combined temperature-hygrometer device (catalog number 11-661-13; Fisher Scientific, Suwanee, GA). No continuous relative humidity recording device was available for the entire 32 h.

The rectal probe was inserted *c.* 5 cm into the dog's rectum. The liver probe was passed between the dog's ninth and tenth ribs halfway between the spinal column and costal arch and inserted *c.* 4 cm into the liver. The brain probe was inserted to a depth of *c.* 4 cm through a trephined hole in the cranium midway on a line running from the medial point of the left eye to the medial aspect of the right ear canal. The aural probe was inserted *c.* 4 cm into the external ear canal and secured to the ear using masking tape.

Vitreous humor was sampled using a 1 mL syringe with 18 gauge, 1 inch needle to puncture the sclera and withdraw *c.* 0.5 mL of vitreous humor from the posterior chamber of each eye. Each eye of each dog was sampled separately, all samples were refrigerated, and then were later analyzed, i.e., there was no "pooling" of vitreous humor samples. Vitreous humor was sampled twice, first *c.* 1.5 hours after death, and second *c.* 7 h after death.

Hair was clipped shortly after death from each dog in the upper right thoracic region between the third and sixth vertebrae. The hair was placed in a zip-lock bag and the area from which the hair had

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been clipped was recorded. Hair coat density was calculated by dividing hair weight by the area from which the hair had been clipped.

After the temperature recording was completed, the dog's body was immersed in a container completely filled with water. Displaced water was collected in a containment pool under the container and measured to determine body volume. Body mass was calculated by dividing body weight by body volume.

Data analyses were performed using JMP 6.0.0 statistical software (SAS Institute Inc., Cary, NC) at the 5% significance level. Statistics were generated using the "Distribution" analysis command that provides univariate analysis. The significance level of the effects of specific variables on mean temperature decrease after death at each measurement site was generated using the "Fit Y by X" analysis command that provides bivariate analysis or simple linear regression when one variable is compared to another. Analysis of vitreous humor potassium ion concentrations used the "Matched Pairs" analysis command that compares two means from the same dog. All graphs were produced by the "Overlay Plot" command.

Results

Descriptive Statistics

Of the 16 dogs, eight were male and eight female with average body weight 16.3 kg (median 14.8 kg, range 8.5–26.2 kg, standard deviation [SD] 6.1 kg). Mean body volume was 19.2 L (median 16.9 L, range 9.3–38.6 L, SD 8.5 L). Mean body mass was 0.88 kg/L (median 0.90 kg/L, range 0.66–1.02 kg/L, SD 0.12 kg/L). Mean hair coat density was 0.03 g/cm² (median 0.03 g/cm², range 0.01–0.06 g/cm², SD 0.1 g/cm²).

Body Temperature Data

Ambient temperature in the room where the study was conducted over the 32-h observation period was 21.5°C (range 20.7–22.7°C, SD 0.55°C). Mean ambient relative humidity for the first 7 h of the study was 41.5% (range 27.8–58.7%, SD 10.9%). Each dog was monitored on a separate weekend, so temperature and humidity data reflect a 32-hour period on each of 16 separate weekends.

Rectal, liver, brain, and aural temperatures were graphed (Figs. 1–4, respectively) over the 32-hour time period. Temperature recording began for all dogs at 65 min after death, the longest initial set-up time after death for any of the 16 dogs. In other words, 65 min was the slowest set-up time of all the experimental equipment for any dog. Figure 5 graphs the same data as Figs. 1–4 plus ambient temperature.

The temperatures (°C) (mean, range, SD) at the beginning of the study were rectal (37.8, 35.5–39.2, 1.0); liver (37.7, 33.7–39.5, 1.5); brain (30.8, 26.3–36.1, 3.4); and aural (33.7, 32.3–35.7, 1.1). The temperatures (°C) (mean, range, SD) at the end of the study were rectal (23.2, 22.2–24.3, 0.7); liver (23.7, 22.3–25.1, 0.8); brain (22.1, 21.2–23.0, 0.5); and aural (22.6, 21.9–23.9, 0.5). The rate of temperature (°C) reduction per hour was rectal (0.5); liver (0.4); brain (0.3); and aural (0.3).

Comparison of Mean Temperatures and Descriptive Variables of Dogs

Sex was not significantly statistically associated with temperature decrease after death at any site (rectum, liver, brain, or ear). Weight was associated with temperature decrease after death at all sites except the rectum (Table 1). Body volume and temperature were associated at all sites, and body mass was not associated with temperature except at the aural site (Table 1). Hair coat density was not associated with the temperature change postmortem at any site (Table 1). One could conclude with the exceptions noted, that sex, body mass, and hair coat density do not affect postmortem temperature change, while body weight and body volume do.

Vitreous Humor Potassium Ion Concentrations

Table 2 contains the vitreous humor potassium ion concentrations. Two results are clear. First, at both the first and second collections, there was no statistically significant potassium ion concentration difference between the right and left eyes. Second, for both the right and left eyes, the potassium ion concentration increased significantly ($p < 0.001$) between the first and second collection. The mean of all eyes at the first collection was 6.0 mEq/L (range 4.7–7.2 mEq/L, SD 0.61 mEq/L). The mean of

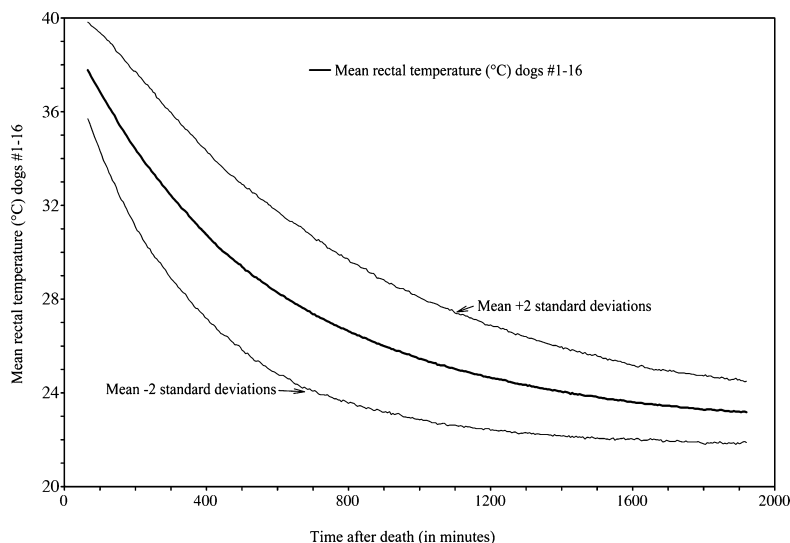


FIG. 1—Data logger mean rectal temperature readings (°C) for all dogs every 5 min ($n = 16$).

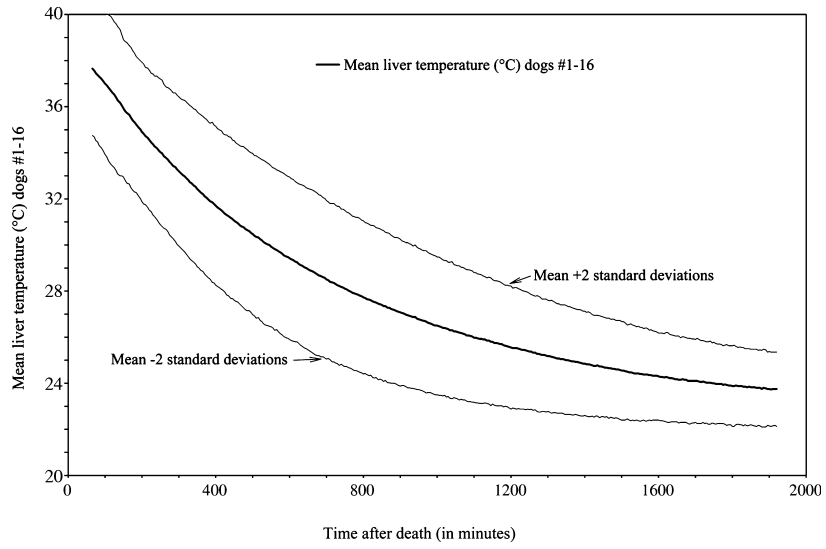


FIG. 2—Data logger mean liver temperature readings (°C) for all dogs every 5 min ($n = 16$).

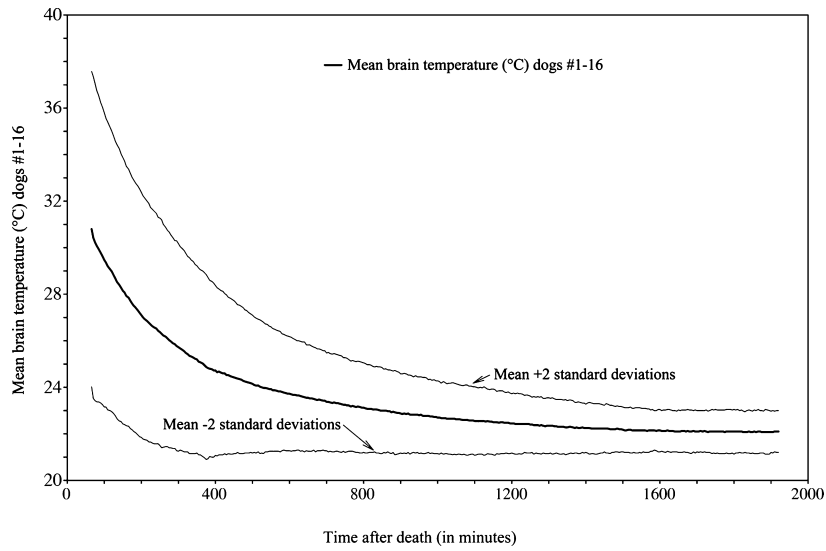


FIG. 3—Data logger mean brain temperature readings (°C) for all dogs every 5 min ($n = 16$).

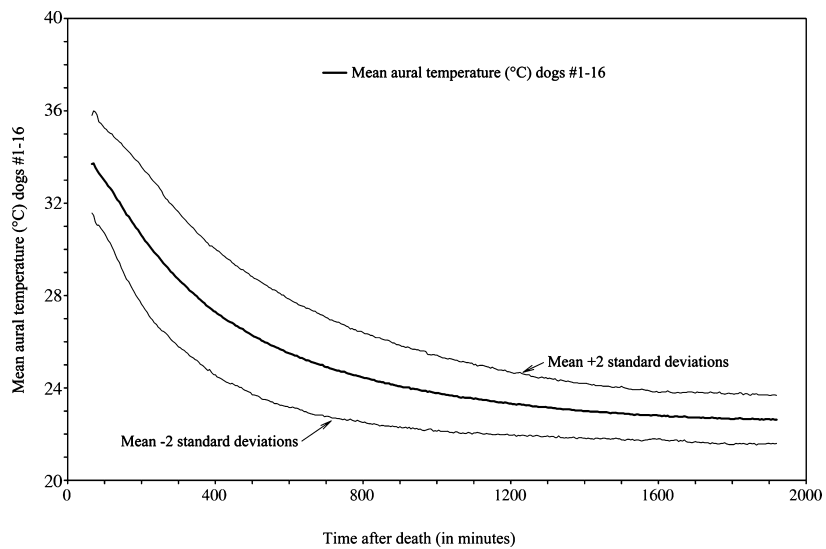


FIG. 4—Data logger mean aural temperature readings (°C) for all dogs every 5 min ($n = 16$).

TABLE 1—Comparison of temperatures of rectal, liver, brain, and aural sites with descriptive variables and variable mean (± 1 SD).

	Mean Temperature ($^{\circ}\text{C}$)			
	Rectal	Liver	Brain	Aural
	26.99 $^{\circ}\text{C}$ ± 1.25	27.83 $^{\circ}\text{C}$ ± 1.33	23.52 $^{\circ}\text{C}$ ± 1.04	25.00 $^{\circ}\text{C}$ ± 0.86
Mean body weight: 16.29 kg \pm 6.05	0.070	0.008*	0.001 [†]	0.002*
Mean body volume: 19.18 L \pm 8.46	0.025*	0.007*	0.003*	<0.001 [†]
Mean body mass: 0.87 kg/L \pm 0.12	0.079	0.263	0.623	0.041*
Mean hair coat density: 0.03 g/cm ² \pm 0.01	0.137	0.271	0.176	0.978

All the values in the Table indicate *p*-values.

*Significant at $p < 0.05$.

[†]Significant at $p \leq 0.001$.

TABLE 2—Comparison of mean of vitreous potassium ion concentrations by eye and collection.

Comparison of Mean Potassium Ion Concentrations (mEq/L)		
1st collection* left eye 6.17	1st collection* right eye 5.79	$p = 0.11$
2nd collection [†] left eye 8.62	2nd collection [†] right eye 8.92	$p = 0.34$
1st collection* left eye 6.17	2nd collection [†] left eye 8.62	$p < 0.001$ [‡]
1st collection* right eye 5.79	2nd collection [†] right eye 8.92	$p < 0.001$ [‡]
1st collection* left eye 6.17	2nd collection [†] right eye 8.92	$p < 0.001$ [‡]
1st collection* right eye 5.79	2nd collection [†] left eye 8.62	$p < 0.001$ [‡]

*Vitreous humor collected at *c.* 1.5 h after death

[†]Vitreous humor collected at *c.* 7 h after death

[‡]Significant at $p \leq 0.001$.

all eyes at the second collection was 8.8 mEq/L (range 7.4–10.1, 0.90 mEq/L). The average rate of increase of potassium ion concentration was 0.5 mEq/L/h.

Discussion

Body Temperature Data

The rectal and liver temperatures at the start of data recording, 65 min after death, were almost identical at, respectively, 37.8 $^{\circ}\text{C}$ and 37.7 $^{\circ}\text{C}$. The brain and aural temperatures at the start of data recording were lower at, respectively, 30.8 $^{\circ}\text{C}$ and 33.7 $^{\circ}\text{C}$. We do not know why they were lower, but we suspect the brain and ear canal cooled more quickly because they are in a smaller “container,” the head, thus making them more susceptible to rapid temperature change.

This speculation is not completely satisfying because one would think that, not being completely contained within the cranium, the ear canal might cool even more rapidly than the brain. Given that the starting aural temperature (33.7 $^{\circ}\text{C}$) was higher than the starting brain temperature (30.8 $^{\circ}\text{C}$), the data do not support this speculation.

One could speculate further that, though the ear canal would seem to be more exposed to the ambient temperature, the depth of the probe inside the ear, *c.* 4 cm, would largely mitigate this effect. One could also speculate that, though the brain probe is inside the cranium, and is seemingly better insulated from the ambient temperature than the aural probe, the process of placing the probe in the brain may itself have cooled the brain tissue in and around the probe, thereby reducing the brain's temperature at the start of data recording. We can offer no other possible explanations for why the brain and aural temperatures were lower at the start of data recording.

The graphs included as Figs. 1–4 for rectal, liver, brain, and aural temperatures are the bedrock of this study. They are tools immediately available to forensic investigators when a dead dog is involved at a crime scene at or near room temperature of 21.5 $^{\circ}\text{C}$ (70.7 $^{\circ}\text{F}$) in still air. Illustration for the use of the graphs is discussed below.

Assume a dog is found dead in still air at approximately room temperature with a rectal temperature of 34.5 $^{\circ}\text{C}$. The mean time of death from our data (Fig. 1) would be 3.2 h. The range within 1 SD (68% of the observations) would be 2.1–4.7 h and for 2 SD (95% of observations) 1.0–6.2 h assuming an underlying normal distribution.

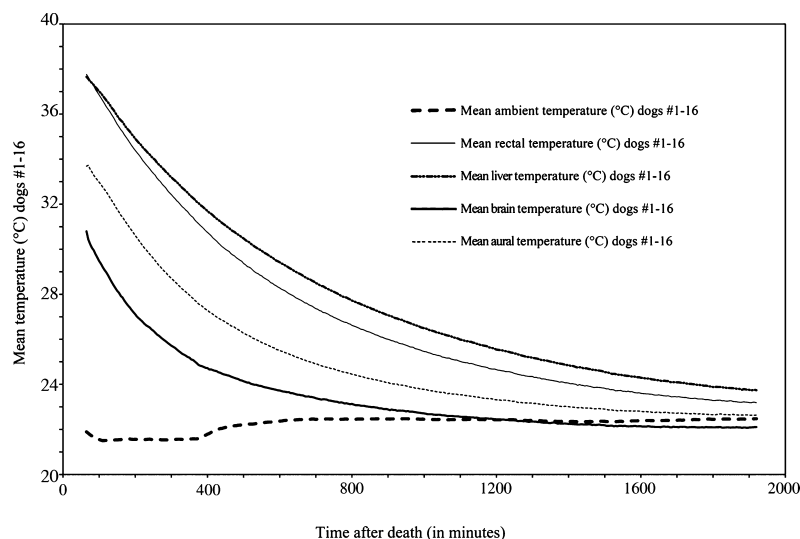


FIG. 5—Data logger mean temperature readings ($^{\circ}\text{C}$) by probe for all dogs ($n = 16$).

The usefulness of these graphs is limited to room temperature and still air, but other studies could produce similar graphs for other temperature and air conditions. The normal rectal temperature in dogs, the temperature used almost exclusively in clinical veterinary practice, is $38.9 \pm 0.5^\circ\text{C}$ (10). We cannot, based on this study, conclude with certainty which site—rectum, liver, brain, or ear—would be most reliable for measuring core body temperature in deceased dogs, though Al-Alousi et al. (11) in a human study suggested that the brain, followed in order by liver, rectum, and ear, is the most reliable.

However, we suggest that, considering both the accuracy of measurement and ease for the forensic investigator, the rectal temperature be used in dogs. This is because of the difficulty of obtaining liver or brain temperatures, and also because, as mentioned, aural and brain temperatures in dogs drop quickly immediately after death. Also note that the postmortem rate of reduction of rectal temperature in the dog was $0.5^\circ\text{C}/\text{h}$. This is very similar to estimates for humans of $0.6\text{--}0.8^\circ\text{C}/\text{h}$ (12).

This study considered some factors that might affect the rate of body temperature reduction. These included sex, body mass, body weight, body volume, and hair coat density. Results indicated that sex does not affect the rate of body temperature reduction. Table 1 summarizes the other factor effects and suggests that the forensic investigator ignore the effects of body mass and hair coat density, but realize when using Figs. 1–4 that as body weight and body volumes increase, the rate of temperature reduction decreases.

This study did not consider every factor that might be involved in the postmortem reduction of body temperature. Some of these include rain, sun, wind, time of day, breed of dog, shape of the dog's head, dog's body type, temperature of the surface on which the dog was found, how much wax was in the dog's ear, and others. This list could go on interminably. Evaluating all these factors was far beyond this study's scope. Nevertheless, recognizing these possible limitations, the graphs are still useful for dead dogs at room temperature in still air.

Vitreous Humor Potassium Ion Concentrations

We found an average vitreous potassium ion concentration of 6.0 mEq/L immediately after death. This is essentially identical to the 5.92 mEq/L of Crowell and Duncan (9) found antemortem. Normal serum potassium ion concentration in dogs is 3.5–5.3 mEq/L (10). Interestingly, the average increase in potassium ion concentration in this study of dogs (0.5 mEq/L/h) was exactly identical to the 0.5 mEq/L/h reported in humans (13).

We are reasonably confident that right and left eye vitreous humor potassium ion concentrations are virtually identical at the same time after death, and that potassium ion concentrations increase after death (Table 2). The two vitreous humor measurements from this study cannot be used to plot over time the postmortem increase in potassium ion concentration. Two points are simply not enough. Another study should be done to plot more

points over time. Nevertheless, this study adds the useful results that left and right eyes have the same potassium ion concentration and that the concentration increases after death.

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References

1. American Veterinary Medical Association. U.S. pet ownership and demographics sourcebook. Schaumburg, IL: American Veterinary Medical Association, 2002.
2. Cox RJ, Mitchell SL, Espinoza EO. CompuTOD, a computer program to estimate time of death of deer. *J Forensic Sci* 1994;39:1287–99.
3. Hadley BM, Robbins LW, Beffa DA. Estimating time of death of deer in Missouri: a comparison of three indicators. *J Forensic Sci* 1999;44:1124–30.
4. Johnson BC, Maguire LA, Anderson DR. Determining time of death in mule deer by using potassium levels in the vitreous humor. *Wildlife Soc Bull* 1980;8:249–52.
5. Woolf A, Gremillion-Smith C. Using vitreous humor to determine time of death: problems and a review. *Wildlife Soc Bull* 1983;11:52–5.
6. Hood C, Daoust P, Lien J, Richter C. An experimental study of postmortem ocular fluid and core temperature analysis in incidentally captured harbour porpoise. *NAMMCO Sci Publ* 2003;5:229–42.
7. McLaughlin BG, McLaughlin PS. Equine vitreous humor chemical concentrations: correlation with serum concentrations, and postmortem changes with time and temperature. *Can J Vet Res* 1988;52:476–80.
8. McLaughlin PS, McLaughlin BG. Chemical analysis of bovine and porcine vitreous humors: correlation of normal values with serum chemical values and changes with time and temperature. *Am J Vet Res* 1987;48:467–73.
9. Crowell WA, Duncan JR. Potassium concentration in the vitreous humor as an indicator of the postmortem interval in dogs. *Am J Vet Res* 1974;35:301–2.
10. Merck & Co., Inc. The Merck manual of diagnosis and therapy. Whitehouse Station, NJ: Merck Research Laboratories, 1999.
11. Al-Alousi LM, Anderson RA, Worster DM, Land DD. Multiple-probe thermography for estimating the postmortem interval: I. Continuous monitoring and data analysis of brain, liver, rectal and environmental temperatures in 117 forensic cases. *J Forensic Sci* 2001;46:317–22.
12. Saferstein R. *Criminalistics: an introduction to forensic science*, 8th edn. Upper Saddle River, NJ: Pearson Education, 2004.
13. Agrawal RL, Gupta PC, Bhasin S, Nagar CK. Determination of the time of death by estimating potassium level in the vitreous humor. *Indian J Ophthalmol* 1983;31:528–31.

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