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The Eye as a Chemical Indicator of Environmental Temperature at the Time of Death

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ABSTRACT: Vitreous humor chemistry profiles were reviewed on 133 autopsied cases in which death occurred outdoors during a six-year period, to determine whether environmental temperature at the time of death influenced chemistry values obtained at autopsy. The glucose concentration and total carbon dioxide content varied inversely with temperature. Values were significantly higher in the winter than the summer months. The mean glucose level was higher in deaths caused by cold exposure than in other deaths occurring in the cold, but individual cases could not be distinguished on the basis of chemistry values. Potassium levels tended to be slightly lower in winter. It was noted that chemistry studies could be used to help determine whether a body found outdoors in winter actually died in a different, warmer environment.

KEYWORDS: pathology and biology, vitreous humor, glycolysis, postmortem chemistry, hypothermia, temperature effects

It was recently observed that under certain circumstances, the environmental temperature at the time of death can influence the extent to which the human vitreous humor glucose level decreases postmortem [1]. Bray et al [1] performed vitreous humor chemistry studies on a group of air crash victims who became immersed in near-freezing water, and observed that the mean glucose concentration of these individuals was significantly higher than that of a comparison group in which death did not occur in a cold environment. The results suggested that chilling of the eye at the time of death could inhibit glycolysis, slowing the normal post-mortem fall in the glucose level. This hypothesis was subsequently confirmed in an experimental setting [2].

These findings were the stimulus for an extensive review of vitreous humor chemistry data obtained at autopsy, to see if evidence existed of an effect of environmental temperature on chemistry values. The results of this study are presented in this paper.

Materials and Methods

The autopsy records of the Office of the Chief Medical Examiner of the District of Columbia were reviewed for the period December 1977 through April 1983, with the exception of the spring and fall months of 1981 and the fall of 1982. All cases in which vitreous humor chemistry profiles were obtained on individuals who were known to have died outdoors, or in a setting exposed to outdoor temperatures, based on available history and scene investigation, were selected for study. Cases were excluded if the vitreous humor glucose level was higher than 200 mg/dL. A total of 133 cases were accepted for study. All chemical analyses

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during this period were performed by the same laboratory, using an autoanalyzer. Glucose measurement was by a hexokinase method.

Actual environmental temperatures at the time of death were not known for any of these cases. In many cases, even the time of death was not known with any accuracy. It was therefore decided to obtain a rough estimate of temperature at the time of death by using National Weather Service reports of average daily temperatures for the Washington, DC area. The average daily temperature was found for each case for the day the body was pronounced dead.

There were a total of 63 cases during the winter months of December, January, and February in which death took place outdoors, or in an unheated building or vehicle. In 29 of these cases, death was judged to have been caused by cold exposure, on the basis of the available history, scene investigation, autopsy findings, core temperature, and toxicologic studies. The highest average daily temperature of the day a body was pronounced dead in this group was 5°C (41°F). Of the 34 other deaths during the winter months, 28 occurred on days with an average temperature equal to or lower than 5°C (41°F). To be able to compare chemistry values obtained in the hypothermia cases to those of individuals who died in the cold from other causes, only these 28 cases were used in calculating mean and range values.

Causes of death were similar through the year. Of the 34 nonhypothermia deaths in winter, 11 were caused by trauma, 10 by cardiac disease, 3 by acute alcohol intoxication, and the remainder by a variety of causes. There were also a total of 34 cases for the months of June, July, and August, in 10 of which death resulted from trauma, 9 from cardiac disease, 6 from drug overdose, and 3 from drowning with or without trauma. The spring and fall months showed a similar incidence of causes of death.

For purposes of comparison, a group of "room temperature" cases was assembled by collecting two consecutive series of vitreous humor chemistry profiles obtained on adults who died in heated homes or apartments, or in emergency rooms or on hospital wards, during the winter months of 1980-81 and 1981-82, for a total of 70 cases. Twelve of these individuals died as a result of trauma; the remainder from a variety of diseases or the complications of drug or alcohol use.

As is typical for cases in which bodies are found outdoors, the true postmortem interval was not known in the majority of cases. To evaluate the relationship between chemistry values obtained at autopsy and the length of time that elapsed between pronouncement of death and the autopsy, this interval was determined for all cases. The mean and range values proved to be similar for all seasons of the year: for all winter cases, the mean was 14.3 h (3 to 26); for summer, 14.0 (3 to 28); for spring and fall, 13.8 (2.5 to 30); and for the room temperature group, 15.4 h (3 to 50). The portion of the interval during which the body was in a morgue cold room was not determined.

Results

Means and ranges of the data for the various groups are shown in Table 1. Results for each solute will be presented individually.

Glucose

As shown in Fig. 1, there is an inverse relationship between the vitreous humor glucose level measured at autopsy and the estimated environmental temperature at the time of death. This relationship is evident despite the wide scattering of values found in all temperature ranges. Causes for this wide scatter will be discussed below. Of interest is the fact that low values were never seen in the winter cases (no glucose concentration below 34 mg/dL in the range 5°C [41°F] or colder), and high values were absent in the summer group (none above 95 mg/dL at temperatures over 13°C [55°F]).

The relationship between the mean glucose concentration and the mean environmental

TABLE 1—Vitreous humor chemistry values.^a

Number of Cases	Average Temperature, °F ^b	Glucose, mg/dL	CO ₂ Content, meq/L	Potassium, meq/L	Sodium, meq/L	Chloride, meq/L	Urea Nitrogen, mg/dL
Hypothermia cases (29)	27.9 (16-41)	83.8 (45-185)	13.4 (1-25)	9.0 (5.5-26.7)	143.8 (129-155)	123.8 (108-134)	32.8 (1-195)
Other deaths in cold (28)	32.4 (3-41)	73.4 (34-160)	11.2 (3-20)	9.6 (6.1-27.0)	145.7 (129-172)	124.6 (108-148)	31.7 (5-252)
Spring and fall (36)	60.4 (44-80)	45.6 (16-95)	10.0 (3-15)	9.5 (5.7-20.4)	142.5 (104-154)	122.1 (106-134)	20.2 (7-100)
Room temperature cases (70)	68	39.9 (5-145)	9.4 (3-21)	10.9 (6.1-30.8)	141.6 (117-164)	121.2 (85-149)	26.4 (4-212)
Summer (34)	79.2 (65-90)	32.7 (1-75)	9.1 (5-16)	11.2 (5.5-28.5)	142.1 (134-157)	121.7 (104-131)	14.5 (4-42)

^aValues in parentheses are ranges.

^b $t^{\circ}\text{C} = (t^{\circ}\text{F} - 32)/1.8$.

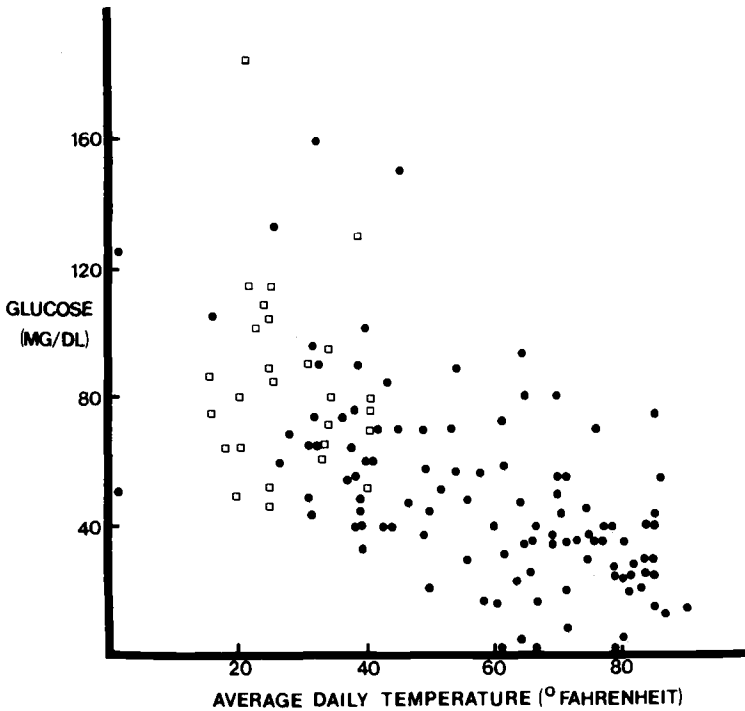


FIG. 1—Vitreous humor glucose concentrations in 133 outdoor deaths, plotted against average temperature on the day of death (see text). Open boxes indicate deaths caused by cold exposure.

temperature for the various groups is shown graphically in Fig. 2. Room temperature is assumed to be 20°C (68°F). A virtually linear inverse relationship is observed. The mean glucose concentration for the group of hypothermia cases is significantly higher than that for other deaths in the cold ($p < .01$ by two-way t test). The mean glucose level of either winter group is much higher than the mean values measured during other seasons or at room temperature ($p < .0005$).

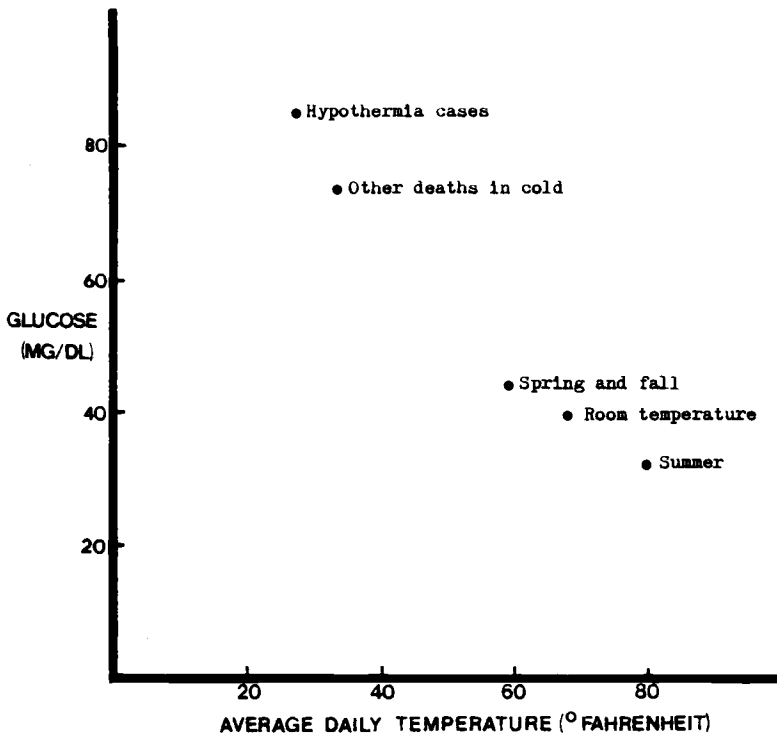


FIG. 2—Mean vitreous humor glucose concentrations of the various groups, plotted against the mean of average daily temperatures.

Least-squares regression lines were computed for the relationship between the glucose concentration and the time interval between pronouncement of death and autopsy. These regression lines are shown in Fig. 3. The gentle downward slope of these lines indicates that the glucose level declines slowly during this interval at all temperature ranges. For example, the data indicate that a delay of 24 h in performing an autopsy on a body found outdoors in winter would tend to result in a glucose value at autopsy approximately 17 mg/dL lower than that which would have been measured the previous day.

In an attempt to minimize the possible effects of antemortem illness on observed postmortem chemistry values, the subgroup of individuals who died suddenly as a result of trauma, asphyxia, drowning, poisoning, or suicidal drug ingestion was also studied. Only those cases were included in which the sodium, chloride, and urea nitrogen concentrations of the vitreous humor revealed no evidence of antemortem chemical imbalance. There were a total of thirteen such cases in the winter group, fourteen in the spring and fall, twelve in the summer, and ten among the room temperature deaths. The mean interval between pronouncement of death and autopsy was comparable for these cases. The mean glucose concentration of the winter cases was 60.2 mg/dL, for the spring and fall 48.1 mg/dL, for the summer 28.1 mg/dL, and for the room temperature cases 30.8 mg/dL. These values are lower than the means for the groups as a whole, except for the slightly higher average for the spring and fall deaths. However, the mean glucose concentration for the cold weather cases is still significantly higher than the mean level in the spring and fall ($p < .01$), and much higher than the room temperature and summer values ($p < .0005$).

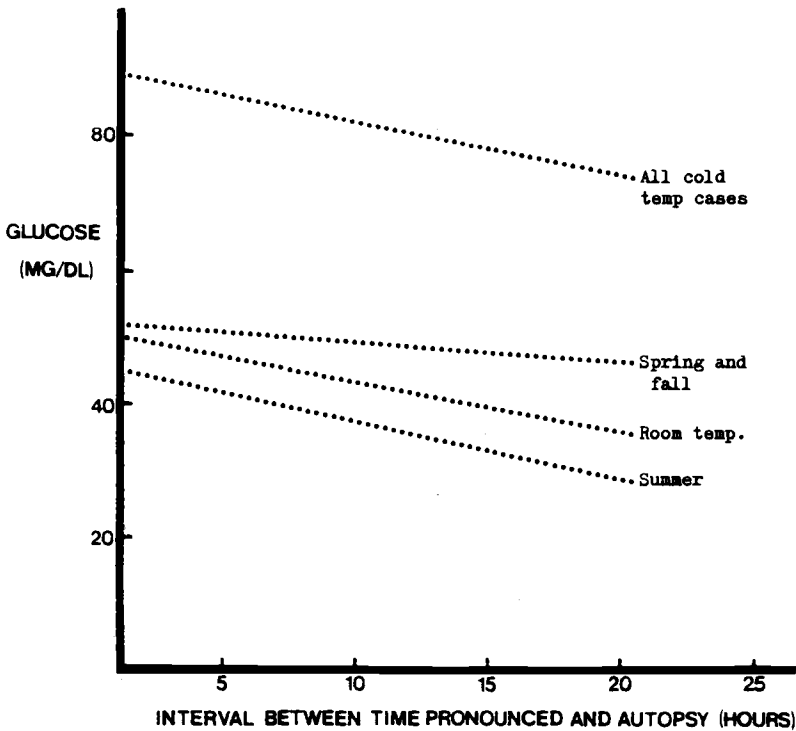


FIG. 3—Least-squares regression lines for the relationship between the vitreous humor glucose concentration and the interval between pronouncement of death and autopsy for the various temperature groups.

Carbon Dioxide

The data show that the total carbon dioxide content of the vitreous humor at autopsy is also inversely related to environmental temperature at the time of death. The values tend to be widely scattered, in much the same manner as the glucose levels (individual data points not shown).

It was noted that the total carbon dioxide content was always very low in cases in which the vitreous urea nitrogen level was markedly elevated, apparently as a result of the associated systemic acidosis. Cases with urea nitrogen levels greater than 50 mg/dL were therefore excluded in assessing the effect of environmental temperature on the carbon dioxide (CO₂) content. After elimination of these few cases, the mean CO₂ content for all cold temperature deaths was found to be higher than the mean for spring and fall deaths ($p < .01$), and much higher than the mean of the room temperature or summer cases ($p < .0005$). This was also true for the subgroup of cases of individuals dying from causes other than antemortem illness: the mean for winter cases (11.8 meq/L) is higher than that for spring and fall (10.5 meq/L), and significantly higher than the mean of the room temperature or summer cases (7.7 and 9.2 meq/L, respectively, $p < .005$).

As with the glucose data, least-squares regression analysis revealed a slow decline in the total carbon dioxide content of the vitreous humor with increasing time interval between pronouncement of death and autopsy in all temperature ranges. The decline over 24 h was in the range of 1 to 3 meq/L (data not shown).

Potassium

There is a tendency for potassium concentrations to be lower in the cold weather groups than in other temperature ranges, but the differences of means do not reach statistical significance. Since true postmortem intervals were generally not known, it was not possible to determine for these cases whether environmental temperature affected the rate of rise of the potassium level postmortem. Least-squares regression lines of potassium concentration versus time interval between pronouncement of death and autopsy had similar slopes in all temperature ranges (data not shown).

Other Solutes

The concentrations of sodium, chloride, and urea nitrogen appeared not to be influenced by environmental temperature at the time of death. The lower mean urea nitrogen levels of the summer and the spring and fall groups simply reflect the absence of any markedly elevated values in those groups.

Discussion

The findings clearly demonstrate an inverse relationship between environmental temperature at the time of death and the vitreous humor glucose level and carbon dioxide content measured at autopsy. Two different factors apparently combine to produce this result. The first is the effect of temperature on the rate of anaerobic glycolysis. If glycolysis is slowed by chilling, diminished lactic acid production will tend to preserve the carbon dioxide content of the vitreous humor by sparing bicarbonate ions. The second factor that contributes to the inverse temperature effect is the presence of many elevated glucose values in the group of cold weather cases. This will be discussed below.

A number of studies have shown that the rate of glycolysis is markedly affected by temperature. Arthus [3] was apparently the first to make this observation. He reported in 1891 that blood kept at 37°C became virtually devoid of glucose after 24 h, while specimens held at 10°C retained an average of 60% of their original glucose after ten days. Subsequent studies have shown that inhibition occurs at the level of the phosphorylation of glucose by hexokinase [4].

The effects of temperature on the postmortem chemistry of the vitreous humor have been studied by using enucleated eyes. Bito and Salvador [5] incubated rabbit eyes at 37°C, and found that the glucose concentration of the vitreous humor fell rapidly, often reaching zero within 4 h. Glycolysis was markedly inhibited at an incubation temperature of 3°C. The rate of rise of the potassium concentration was also diminished at that temperature. It has been shown that the majority of intraocular glycolysis is carried out by the retina, creating an ascending gradient of glucose concentration between the retina and the lens [6].

Two groups of investigators have examined the effect of temperature on the postmortem chemistry of the eye in situ. Bannister et al [7], in a study of induced hypoglycemia in chicks, found that the glucose level fell rapidly, and to roughly the same extent, in chicks kept at room temperature or at 4°C during the first hour postmortem. The rate of glycolysis was slowed at 4°C thereafter. Schoning and Straffuss studied vitreous humor chemistry values in dogs kept at 4, 20, or 37°C for up to 48 h after death. They found that the glucose concentration fell by more than 50% within 3 h in all three groups, and declined more slowly from then on. No temperature effect was seen. Potassium levels rose more slowly at lower temperatures.

The findings of the present study are in better agreement with the results of experiments using enucleated eyes than with those employing intact animals. This discrepancy can be readily explained if the factors involved in producing a temperature effect on the metabolism of the eye are taken into account. The most important factor must be the rapidity and degree of retinal cooling during the early postmortem period. In those experiments in which animals were placed in the cold after death, considerable glycolysis occurred before the retina began

to cool. In contrast, most humans who die in a cold environment have been exposed to the cold before death, and some chilling of the eye and its surrounding tissues has already taken place. This is true by definition of death from hypothermia.

The influence of the antemortem environmental temperature provides an explanation for some of the lowest glucose values observed in the series of cold weather cases. As an example, four of the deaths were the result of accidental or suicidal falls from apartment buildings, in which the decedents went directly from warm to cold environments at the time of death. The glucose levels were 34, 48, 55, and 90 mg/dL in these cases. The last value is that of an elderly man, who may have had some degree of glucose intolerance, causing his higher level.

The degree of coldness of the environment and its physical nature must also determine the extent of any cold effect. Many of the cold weather deaths in the present series took place at temperatures below those used in the intact animal studies. Death in cold water, as in the case of the air crash victims, results in very rapid heat loss from the eye. Inhibition of glycolysis might begin within minutes after death in such an environment.

In general, the effect of chilling is to place a limit on how far the glucose concentration and carbon dioxide content fall postmortem. Above that lower limit there is wide variation in individual values. Most of the variation in glucose levels is probably caused by variability in antemortem blood glucose concentrations. In some cases, undiagnosed diabetes mellitus or milder degrees of glucose intolerance may be present. However, the temperature correlation could probably be improved in spite of antemortem variation if the actual temperature at the time of death were known.

As mentioned, many of the glucose values measured in the cold weather cases are equal to or higher than normal blood glucose concentrations. High values are somewhat more common in deaths caused by cold exposure. Published reports have shown that many patients suffering from hypothermia are hyperglycemic at the time of hospital admission [9]. This may be related to the elevated urine catecholamine levels and glycosuria that have been observed in some hypothermia cases at autopsy [10, 11]. The overlap observed between chemistry values obtained in hypothermia cases in this series and values measured in winter deaths from other causes unfortunately makes vitreous humor chemistry studies of little value in diagnosing death from cold exposure.

Among individuals who were judged to have died from cold exposure, it was found that 59% had blood alcohol levels at autopsy of 0.05g/dL or higher. There was no correlation between the blood alcohol concentration and any vitreous humor chemistry values. There was also no evident relationship between the presence of fatty change in the liver and the concentration of glucose or any other solute. In three cases in which pancreatic fibrosis was grossly evident at autopsy, the vitreous humor glucose values were 105, 115, and 125 mg/dL.

Hypoglycemia has been reported to occur in some alcoholics suffering from cold exposure [12]. There were no very low glucose values in the present series of hypothermia deaths, but another investigator has encountered such cases.² If antemortem chilling of the eye is known to have occurred, it might be possible to reliably diagnose hypoglycemia on the basis of the vitreous humor glucose level.

Another interpretation of a low glucose concentration measured in a body found in the cold is that death actually occurred in a different, warmer environment, and the body was subsequently placed where it was found. The following case from the files of the District of Columbia office provides an example of a potential criminalistic application of vitreous humor chemistry studies in winter deaths. The body of a young woman was found outdoors on a winter morning. Examination of the body indicated that the decedent was a narcotics addict. Toxicology studies subsequently revealed that she had died from an overdose of heroin. From her clothing, the absence of personal effects, and the presence of nearby tire marks, investigators concluded that the body had been dumped from a car. Vitreous humor chemis-

²Dr. John Coc, personal communication.

try studies performed at autopsy later that day showed a glucose concentration of 30 mg/dL, a carbon dioxide content of 7.0 meq/L, and a potassium concentration of 12.9 meq/L. These values, which are not representative of an outdoor death in winter, support the conclusion that the woman died indoors.

Since the eye is directly exposed to the environment, it is not surprising that environmental temperature appears to influence postmortem chemistry values. However, in the communication previously cited, Dr. Coe has reported finding no difference between the glucose levels of a group of traumatic outdoor deaths in winter and those of a group of indoor deaths. Elevated glucose values were found in many, but not all hypothermia deaths. Further research will be required to clarify these temperature effects.

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