In the Laboratory

Bone Marrow of Horses and Cattle

LOIS CALHOUN
Department of Anatomy, Michigan State College
East Lansing

In a recent cytological study of the bone marrow of 7 horses and 14 head of cattle the range and mean cell counts were determined (Table 1). These figures compare favorably with those of other investigators (*1–5*).

A satisfactory procedure for extracting the marrow is presented below:

The 11th rib in the cow or the 11th to 16th ribs in the horse, at a level just below the long muscles of the back, are the best sites at which to secure the marrow sample. Confine the subject in a stock or restrain it against the side of the stall. Brush the back and side of the animal with a grooming brush and wipe with a damp cloth. Shave or clip the hair over the area, wash with soap solution, apply iodine, and

TABLE 1

SUMMARY OF BONE MARROW DATA ON THE COW AND HORSE

Cells	Cow		Horse	
	Range	Mean	Range	Mean
Stem cell	0.0 – 5.0	2.14	0.4 – 3.4	1.6
Erythroblast	11.8 –42.8	30.26	8.0 –32.0	20.94
Normoblast	7.2 –39.2	21.69	5.0 –24.2	13.71
Total erythroid cells (E)	21.0 –72.2	52.66	19.0 –47.6	34.66
Promyelocyte	0.0 – 6.8	1.51	0.0 – 5.0	1.83
Neutrophilic myelocyte	10.4 –32.0	19.39	26.2 –56.0	38.06
Neutrophil	1.2 –12.2	5.73	1.8 –20.2	13.31
Eosinophilic myelocyte	1.8 –10.4	6.69	0.4 – 3.6	2.34
Eosinophil	0.0 – 7.6	1.92	0.2 – 1.2	0.60
Basophils (all)	0.0 – 1.0	0.34	0.0 – 1.0	0.60
Total myeloid cells (M)	19.6 –60.4	35.59	45.0 –71.6	56.74
Monocyte	0.0 – 7.6	2.64	1.2 – 4.8	2.46
Plasma cell	0.2 – 2.0	0.79	0.0 – 0.8	0.63
Lymphocyte	1.4 –16.8	6.68	2.0 – 5.6	3.91
Megacaryocytes in 300 sq. mm.	0–121	25.14	0–8	1.71
Mitoses/500 cells	0– 11	4.9	0–8	2.71
Myeloid-erythroid ratio (M/E)	0.27– 2.5	0.676	0.94– 3.76	1.64

anesthetize the skin, fascia, and periosteum with 2 per cent procaine hydrochloride. Using a hand drill equipped with a 3/32-inch jobbers' drill, bore into the center of the rib in order to hit the marrow cavity. The drill "gives" when it enters the cavity. Caution should be exercised not to miss the marrow cavity, because there is danger of entering the thoracic cavity. Remove the drill and insert a needle trocar with the same outside diameter as the drill. Remove the needle, attach an airtight syringe, and aspirate 1 cc. or less of marrow. Pure marrow is more viscous than blood and greyish red. The sample is diluted more or less with blood, depending upon the amount of blood aspirated with the marrow.

The operative wound heals without leaving any visible scar once the hair has grown over it.

Obtain blood samples at the same time the marrow samples are taken. It is possible to carry out routine pipette filling and smear technics at the side of the animal, but a better method is to use oxalate tubes and take the material to a laboratory for examination.

It is hoped that this technic and the data presented may encourage other investigators to include the marrow in routine hematological studies of both normal and disease processes of horses and cattle.

A more detailed report will be published elsewhere.

References

1. ACKERNECHT, E. *Virch. Arch.*, 1912, **208**, 396–414.
2. HJÄRRE, A. *Skand. vet. tdskr.*, 1943, **33**, 457–472.
3. HJÄRRE, A., and BERTHELSEN, H. *Proc. 13th int. vet. Congr.*, 1938, **1**, 259–272.
4. MARCATO, A. *Nuova Vet.*, 1941, **19**, 173–179. (Abstract in *Jb. vet. Med.*, 1942, **69**, 530.)
5. VARIĆAK, T. D. *Arch. wiss. prakt. Tierheilk.*, 1935, **73**, 461–475.

A Simple Vaporizing Device for the Attainment of Bactericidal Concentrations of Glycol Vapors in Air[1]

T. N. HARRIS and JOSEPH STOKES, JR.
The Children's Hospital of Philadelphia and
Department of Pediatrics, School of Medicine
University of Pennsylvania

It has been well established that bacteria and viruses retain their infectious properties when dried in air, and that such air-borne organisms can transmit respiratory infections over considerable intervals of space and time. Since this mode of transmission, as against direct contact and droplet transmission, is responsible for a considerable percentage of all respiratory infections, both sporadic and epidemic, and since it is not affected by the ordinary hygienic precautions even where they are applied, the disinfection of air in enclosed spaces is a major problem in the control of respiratory diseases.

The demonstration of the lethal effect of certain glycol vapors on air-borne bacteria (*6*) and influenza virus (*5*) obviously suggested another means of ap-

[1] The work described in this paper was done under a contract, recommended by the Committee on Medical Research, between the Office of Scientific Research and Development and the Children's Hospital of Philadelphia.

proaching the problem of disinfection of enclosed air besides that of ultraviolet irradiation.

Satisfactory results from the application of chemical disinfection of air to the control of air-borne cross-infection in human beings have already been published by the authors (*4*) and others (*2*). It was shown that the total rate of occurrence of respiratory infections was markedly decreased in a group of subjects in whose air supply a bactericidal concentration of glycol vapor was maintained. Devices for vaporization of glycols had been developed by the Research Corporation and by Bigg (*1*). It was felt in this laboratory, however, that the development of as simple a device as possible might favor wide application of chemical disinfection of air to the control of respiratory cross-infections.

For operation in relatively small enclosed spaces it was decided to make a small, portable unit, such as would maintain adequate concentration of glycol in a volume of air not so great as to involve problems of dispersal and distribution of the glycol vapors to be liberated by the device. For somewhat larger enclosed spaces it was thought better to use an appropriate number of such units rather than a single, more powerful one.

The apparatus, as developed, depends on the use of wicks and an ordinary electric light bulb. The wicks stand in a reservoir of glycol, with the upper ends at the level of the incandescent bulb. The glycol rises in the wicks by capillarity, and the radiant heat in the vicinity of the bulb causes the glycol at the top of the wick to evaporate. This is in turn replaced by additional glycol rising in the wick. The wicks are made of Fiberglas[2] in order to avoid carbonization by the higher temperature at the top. Although some heat is conducted down the wicks, the temperature of the reservoir does not rise above 50° C. Finally, a circular opening around the socket of the incandescent bulb admits air which, on being heated by the bulb, rises in a continuous upward stream, carrying off the glycol vapor.

The apparatus (Fig. 1) consists of a cylinder cut out at the center. The latter is open at the top and bottom, and in this space there is a socket for an electric light bulb. The top of the container, or glycol reservoir, is perforated by holes, 1 cm. in diameter, arranged in two circles concentric with the instrument itself. Through these holes pass the wicks of braided Fiberglas, which are somewhat frayed at both ends for greater efficiency. The wicks, 15 cm. long and terminating near the widest part of the incandescent bulb, are fixed in their vertical position by being passed through a similarly perforated plate near the lower end of the reservoir. An approximately hemispherical dome containing a shell of glass-wool insulation, 10 cm. in height, surmounts the apparatus, and a hole 10 cm. in diameter is cut out at the very top for egress of the glycol vapor and of the warmed air which carries it off. A float gauge and a glare shield complete the apparatus.

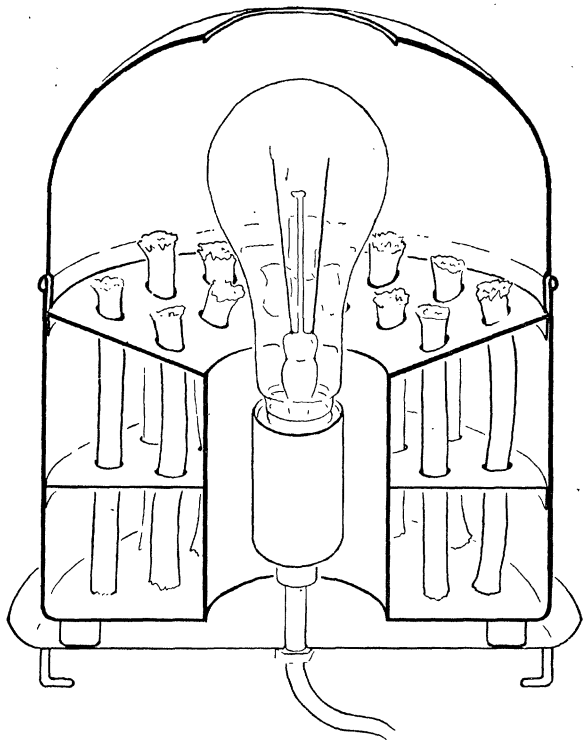

FIG. 1

The criterion of simplicity of construction and operation has been satisfied by the absence of any moving parts or valves. With respect to flexibility of operation, the output of glycol vapor in unit time can be regulated coarsely by changing the wattage of the bulb or quite finely (to a 4 per cent difference) by removing one or more wicks from the vaporizer. This flexibility is of importance in adjusting the device to the enclosed space in which it is to be used, in terms of volume of air, rate of air exchange, and activity of air currents.

With propylene glycol in use, the characteristics of this vaporizer are as follows: The rate of evaporation of the glycol, using a 100-watt bulb and all the wicks, in a room of about 20° C., is approximately 38 cc./hour. At a conservative estimate, it is calculated that this will supply 200 cubic feet/minute of outside air with glycol vapor to a concentration well above the bactericidal threshold. Stated in terms of volume

[2] The authors wish to express their thanks to Walter R. Sykes, of the Owens-Corning-Fiberglas Corporation, and to F. N. P. Supplee, a Philadelphia horticulturist, whose experience with Fiberglas wicking was of great use in the development of the vaporizer.

and rate of air exchange, it would supply a volume of 1,300 cubic feet with 9 complete air changes per hour. With the vaporizer at room temperature, the vapors can be demonstrated above the apparatus within 2 minutes of turning on the switch of the incandescent bulb. When the apparatus was used in a room of 1,260 cubic feet, the glycol was dispersed to all corners of the room with sufficient rapidity to reach bactericidal concentration within 30 minutes, in the absence of any human activity except that of the operator.

A typical series of tests of the apparatus gave the following results: a 1:10 dilution of an 18-hour broth culture of Group C streptococci was sprayed into the air of a room $9 \times 9 \times 15.5$ feet, and the organisms were dried by mixing the output of the spray with many volumes of room air in a large bottle. The window was opened 1½ inches, and the relative humidity was 32. After 10 minutes, the bacterial spray was discontinued, and one Petri plate was exposed for 5 minutes near each corner of the room, at table height. The glycol vaporizer was then attached to the electrical outlet. At various intervals thereafter the spraying of bacteria and the exposure of plates were repeated identically as in the case of the controls above. All plates were incubated for 48 hours at 37° C., and colony counts were then made. In this series of experiments the table supporting the bacterial spray and glycol vaporizer was near corner D and farthest from C (Table 1).

TABLE 1

CONTROL COLONY COUNTS AND COUNTS AT INTERVALS AFTER CONNECTING THE VAPORIZER TO THE ELECTRICAL OUTLET

Site	30 Minutes				60 Minutes			
	Exp. 1		Exp. 2		Exp. 3		Exp. 4	
	Control	With glycol	Control	With glycol	Control	With glycol	Control	With glycol
A	339	28	280	20	99	1	252	3
B	322	14	272	10	97	1	238	2
C	280	37	252	14	87	0	220	1
D	374	3	208	3	94	1	231	1

Similarly satisfactory results were obtained on spraying *Escherichia coli* and *Staphylococcus albus*. The concentrations of bacteria in air produced by spraying in these tests are, of course, very many times the concentrations found by similar means in any normal habitation.

The application of this vaporizer is not predicated on the use of any particular glycol. At present there is no general agreement among all workers in the field as to the glycol of choice, and it is entirely possible that those in current use, propylene and triethylene, may be superseded. Propylene glycol was used in these experiments and seems at present to be preferable for the applications for which the device was intended, *i.e.* small enclosed spaces not equipped with devices for regulating the rate of vaporization. The most important reason for this is that the wider range of concentration between bactericidal and precipitation thresholds in the case of propylene glycol obviates the necessity for special regulatory instruments.

Finally, no attempt was made, in developing this device, to provide means of controlling the relative humidity. Recent work by Hamburger, Hurst, Robertson, and Puck (*3*) has shown that, although the bactericidal effect of triethylene glycol vapor is greater in the presence of relative humidity above 40, it is only somewhat lower at a relative humidity of 18–30 per cent. Work done with propylene glycol in this laboratory is consistent with these findings. This minimum relative humidity is not higher than is necessary as a general hygienic measure and should be maintained by some means which is a part of the heating or ventilating apparatus. Hamburger, *et al.* suggest for the purpose steam caps in steam-heated interiors. In the apparatus described here the technical problems of simultaneous vaporization of glycol and water would defeat the original purpose of producing as simple a device as possible.

References

1. BIGG, E., and JENNINGS, B. H. *Ind. Hyg. Toxicol.*, 1944.
2. BIGG, E., JENNINGS, B. H., and OLSON, F. C. W. *Amer. J. publ. Hlth*, 1945, **35**, 788.
3. HAMBURGER, M., JR., HURST, V., ROBERTSON, O. H., and PUCK, T. T. *J. inf. Dis.*, 1945, **77**, 177.
4. HARRIS, T. N., and STOKES, J., JR. *Amer. J. med. Sci.*, 1942, **204**, 430; 1943, **206**, 631; 1945, **209**, 152.
5. HENLE, W., SOMMER, H. E., and STOKES, J., JR. *J. Pediat.*, 1942, **21**, 577; ROBERTSON, O. H., *et al. Science*, 1943, **97**, 142.
6. ROBERTSON, O. H., *et al. J. exp. Med.*, 1942, **75**, 593; *Science*, 1943, **97**, 142.

Fine-tapered Silver Electrodes for Physiological Work [1]

KENNETH D. ROEDER

Department of Biology, Tufts College

In the course of electrophysiological studies of nerve activity in insects (*1*) fine and pliable electrodes were necessary. It was found that 28 to 36 B- and S-gauge silver wire was too coarse, while finer gauges were subject to whip and vibration. Conventional saline-filled glass capillary electrodes had the disadvantage of high electrical resistance, while it was impossible to bend such electrodes during an experiment to conform with the short nerves and narrow operational fields encountered in insects. Various mechanical methods of tapering silver wire were tried without success until a simple and rapid electrolytic method was developed.

Leads are soldered to short lengths of No. 28 B and S soft-drawn silver wire, which are mounted in a glass

[1] The electrodes were developed in the course of work done under a contract between the Chemical Warfare Service and Tufts College.