

osmotic (10) experiments. In terms of the diffusion equation discussed above, when V is very small, a change in U will have no effect on E until U approaches V . This provides an explanation for the failure of cocaine up to 0.2% to produce a consistent reduction in the effectiveness of applied KCl in lowering the potential (Fig. 3).

For frog nerve evidence has been presented that the rise of resting potential in 5% $\text{CO}_2\text{-O}_2$ and its decline during anoxia are due to the uptake and release of K by the fibers, the associated changes in the K concentration of the extracellular spaces being the immediate major cause of the potential changes under these conditions (8, 9). A reduced permeability to K therefore would be indicated by a decrease in the magnitude of the potential changes in both cases. This has been observed under a variety of experimental conditions and with the lowest cocaine concentration employed (viz., 0.0005%).² Fig. 3, typical of a number of such experiments, is particularly instructive, inasmuch as it demonstrates not only that cocaine delays the decline of potential of a section of nerve rendered anoxic, but that the prevention of this depolarization is a sufficient condition for the maintenance of functional activity as judged from the maximum attainable spike heights observed at the stimulating electrode.

An examination of substances which resemble cocaine by virtue of a "stabilizing" action, e.g. procaine (1, 11), sulfanilamide and thiophene-2-sulfonamide (8, 9), recalls that Wilbrandt (12) employed similar basic compounds to reduce and reverse the negative charge normally present on collodion membranes. Such a mechanism of action appears attractive, inasmuch as chemical potentials obtained with nerve and muscle indicate that these and probably most animal cells have negatively charged surfaces (6). Some unpublished observations indicating that *p*-aminobenzoic acid enhances the ability of sulfanilamide to counteract the anoxic decline of the resting potential in frog nerve are readily explained on this basis.

These experiments therefore support the view that cocaine, at properly controlled concentrations, reduces the permeability of frog nerve and muscle to potassium. On this basis another tool is at hand for the analysis of the role of ionic movement in highly transitory phenomena. Thus, observations which have already been made of its action on the time constant of the excitatory process (Hill's "k," which is related to chronaxie) and on the afterpotentials in nerve suggest ionic transport under these conditions. These results and others, which cannot be included in this brief report, will be presented in greater detail subsequently.

References

1. BENNETT, A. L., and CHINBURG, K. G. *J. Pharm. exp. Therap.*, 1946, **88**, 72.
2. BISHOP, G. H. *J. cell. comp. Physiol.*, 1932, **1**, 177.

² In a recently published monograph (*Stud. Rockefeller Inst.*, 1947, **131**, 117), Lorente de N6 concludes that the reduced anoxic decline of potential in cocaine is due to a lowered respiratory rate, although Sherif (*J. Pharm. exp. Therap.*, 1930, **38**, 11) has shown that these concentrations of cocaine have no effect on respiration.

3. BOYLE, P. J., and CONWAY, E. J. *J. Physiol.*, 1941, **100**, 1.
4. CONWAY, E. J., and MOORE, P. T. *Nature, Lond.*, 1945, **156**, 170.
5. HILL, A. V. *Proc. roy. Soc. Lond.*, 1936, **B119**, 305.
6. HOBER, R. *Physical chemistry of cells and tissues*. Philadelphia: Blakiston, 1945.
7. NETTER, H. *Pflug. Arch. ges. Physiol.*, 1928, **218**, 310.
8. SHANES, A. M. *Biol. Bull.*, 1946, **19**, 211.
9. SHANES, A. M. *Yb. Amer. phil. Soc.*, 1946, 162.
10. SHANES, A. M. *Fed. Proc.*, 1948, **7**, 111.
11. TOMAN, J. E. P., WOODBURY, J. W., and WOODBURY, L. A. *J. Neurophysiol.*, 1947, **10**, 429.
12. WILBRANDT, W. *J. gen. Physiol.*, 1935, **18**, 933.

Loss of Radioactivity From Barium Carbonate Samples¹

PETER E. YANKWICH

*Department of Chemistry and Radiation Laboratory,
University of California, Berkeley*

Several investigators have reported observations which lead them to conclude that barium carbonate samples containing C^{14} can lose rapidly rather significant fractions of their activity by exchange with atmospheric carbon dioxide through the agency of moisture. In many experiments it is desirable to store counting samples for possible checking after weeks or months, and any rapid loss of activity by exchange or any other process would make such a practice hazardous in so far as experimental accuracy is involved; other consequences of rapid exchange loss are legion and need not be detailed here.

Two current papers report the results of purposeful investigations of this exchange phenomenon. One of these (2) deals only with exchange loss when solutions of sodium bicarbonate (pH 8.8) are evaporated to dryness in a variety of atmospheres. It was observed that even the small concentration of carbon dioxide in laboratory air caused losses as high as 60% in a single evaporation.

Most C^{14} radioactivity determinations of solid samples are made on barium carbonate, and it is to be expected that this solid can lose activity by exchange with carbon dioxide, through carbonic acid and its ions, though the rate of this exchange should be inappreciable in the absence of water. Though several estimates have been made as to the rate of loss of activity from precipitates of barium carbonate, only one paper has been published which contains the results of experimentation designed to explore the problem (1).

In that study, thick deposits of barium carbonate were prepared by filtering a slurry of freshly precipitated solid through a retentive filter paper; the samples were dried by drawing air through the filter after washing the mass with acetone and ether. All samples used in the investigation were of "infinite thickness" with regard to the C^{14} beta radiations. Several samples thus prepared were subjected to atmospheres of (1) moist carbon dioxide,

¹ This paper is based on work performed under contract No. W-7405-Eng-48 with the Atomic Energy Commission in connection with the Radiation Laboratory, University of California, Berkeley.

(2) moist carbon dioxide-free nitrogen, and (3) dry carbon dioxide. The experiments were carried out in a desiccator, the bottom of which was covered with a layer of acidified water during the "moist" experiments. Exchange was observed only in the first case; after 65 hrs, 37.4% of the activity had disappeared from the solid and was recoverable from the gas phase.

Somewhat different results have been obtained in these laboratories with samples prepared by evaporation-settling technique (3). Samples are prepared by the evaporation of 95% alcohol from an alcohol-barium carbonate slurry; after the evaporation is complete, the samples are dried for a few seconds at 160° C. (The barium carbonate used in these experiments was dried at 115° after precipitation from an ammonium nitrate-sodium hydroxide solution; washing on the filter was carried out with distilled water only.)

(I) Four different types of experiment were performed, the first of which was a repeat of the work just described. Two plates (deposits of solid on aluminum), 3.7 and 5.6 mg/cm² thick, were stored in a sealed desiccator, the bottom of which was covered with distilled water; the gas space was swept with a moderate stream of tank carbon dioxide for 15 min before the vessel was sealed. After 172 hrs the plates were removed and counted; the activity loss in both cases was $0.9 \pm 1.0\%$. Concentrated hydrochloric acid was added to the water layer in an amount necessary to produce an approximately 3N solution; the samples were put back into the vessel, which was swept again with carbon dioxide and sealed. The samples were removed after 150 hrs and their radioactivities determined; the lighter sample lost an additional $1.2 \pm 1.0\%$ of its original activity, while the heavier plate apparently gained $0.2 \pm 1.0\%$. The atmosphere during the second part of this experiment was sufficiently acid to etch the exposed portions of the aluminum sample mounts.

These experiments were repeated with samples, prepared by filtration technique (1), which were about 8 mg/cm² thick. In carbon dioxide moistened by distilled water one sample lost $33.0 \pm 1.1\%$ of its activity in 56 hrs. A similar sample prepared from the same batch of barium carbonate lost 36% of its activity when stored in CO₂ moistened by acidified water for 56 hrs. During 56-hr "acid" storage, similar samples prepared from batches of barium carbonate precipitated under various other conditions lost amounts of their original activities which varied from 5 to 40%, all losses occurring without important changes in weight.

(II) An experiment was performed to give information on the exchange rate when the heat-dried solid was in contact with two solvent media. Two plates, both 4.65 mg/cm² thick, were carefully freed of loose particles and placed in shallow dishes. One was covered with 95% ethanol saturated with carbon dioxide, and the other was covered with distilled water, also saturated with carbon dioxide (this gas is about three times as soluble in ethanol as in water). After 28 hrs the samples were carefully removed from the dishes and dried in air at 100°. Radioactivity determinations showed that both samples lost only $0.3 \pm 1.0\%$ of their activity in this time.

(III) In a third test, small portions of carbon dioxide-saturated ethanol and water, and carbon dioxide-free ethanol and water, were evaporated repeatedly from heat-dried barium carbonate plates of various thicknesses.

TABLE 1
ACTIVITY LOSS FROM BaCO₃ PLATES—REPEATED LIQUID EVAPORATION

Liquid	Typical Result			Dependence on sample thickness
	Thickness (mg/cm ²)	Liquid volume (ml)	Activity loss (%)	
95% ethanol Dist. H ₂ O	4.65	11.0	0.0 ± 0.6	None observed Loss/ml decreases with increasing thickness
	0.42	15.0	$0.8 \pm 0.1/\text{ml}$ of liquid	
95% ethanol + CO ₂ Dist. H ₂ O + CO ₂	4.82	21.0	0.2 ± 0.5	None observed Loss/ml decreases with increasing thickness
	0.41	9.1	$0.8 \pm 0.1/\text{ml}$ of liquid	

The plate was freed of loose particles and wet with 0.25 ml of the solvent or solution to be investigated, then dried in air at 100°. The plate was then cooled, another portion of liquid added, and the cycle repeated; at suitable intervals the sample radioactivities were measured. The results are summarized in Table 1. The small difference in the rate of loss between the two aqueous media prob-

TABLE 2
LONG-TERM LOSS OF ACTIVITY FROM BaCO₃ SAMPLES

Conditions of storage	Plate thicknesses (mg/cm ²)	Mean % loss (9 wks)*
Uranium glass	1.2 ± 1.2
BaCO ₃ —shellac bound	1.0 ± 1.2
Covered Petri dishes	1.65, 2.20	2.9 ± 2.0
Office-type room	0.53, 3.06	2.6 ± 1.2
Outdoors (protected only from rain)	1.07, 2.90	15.2 ± 4.2
Stream of wet outdoor air	0.63, 3.29	3.6 ± 1.3
Drying oven at 130° (air)	1.35, 2.14	3.6 ± 1.4
Stream of wet CO ₂	1.75, 2.87	8.0 ± 1.4
Organic synthesis laboratory, low CO ₂ , relatively acid air	1.03, 2.77	6.0 ± 1.3
Combustion room, high CO ₂ , relatively acid air	0.66, 1.88	12.1 ± 1.6
Sample storage book; somewhat better sealed than Petri dishes	1.25, 2.86, 3.10	1.7 ± 1.3 (20 wks)

* Error includes counting error and sensitivity loss.

ably is due to very rapid loss of dissolved carbon dioxide from the evaporating solution of carbon dioxide in water; the rate of loss with this solution probably would be considerably higher if the evaporation were carried out under a constant pressure of carbon dioxide.

(IV) In one of the papers cited (1) a loss of 2.7% in 6 days is reported for a sample stored in a covered Petri dish. A fourth set of experiments has been performed to shed more light on this long-term exchange loss. Pairs of heat-dried barium carbonate plates of different thicknesses were stored over a period of 9 weeks in different controlled and uncontrollable atmospheres. At intervals of 10 to 14 days the plates were collected and their radioactive strengths determined; at the end of the test all samples were reweighed, but no significant weight changes were observed. The sensitivity of the counter was checked with standards of uranium glass and of shellac-bound barium carbonate. The data obtained are collected in Table 2.

It is apparent from these data that some barium carbonate samples, at least those which are heat dried, can

be stored for long periods without serious loss of activity. Exchange may not be a trivial matter with samples prepared by other methods, because of particle size distribution, particle surface condition, etc.; each circumstance at least requires observation of active samples for long periods.

References

1. ARMSTRONG, W. D., and SCHUBERT, J. *Science*, 1947, **106**, 403.
2. LESLIE, W. B. Experimental use of C^{14} . Atomic Energy Commission, MDDC 674.
3. YANKWICH, P. E., ROLLEFSON, G. K., and NORRIS, T. H. *J. Chem. Phys.*, 1946, **14**, 131; DAUBEN, W. G., REID, J. C., and YANKWICH, P. E. *Anal. Chem.*, 1947, **19**, 828. See also: HENDRICKS, R. H., BRYNER, L. C., THOMAS, M. D., and IVIE, O. *J. phys. Chem.*, 1943, **47**, 469.

IN THE LABORATORY

A Piston Recorder for Small Volume Changes

FOSTER N. MARTIN, JR.

*Department of Pharmacology,
Tulane University of Louisiana
School of Medicine*

The apparatus here described was designed for use in recording small volume changes, associated with pressure changes, such as occur in the eye of a cat following administration of vasoconstrictor drugs.

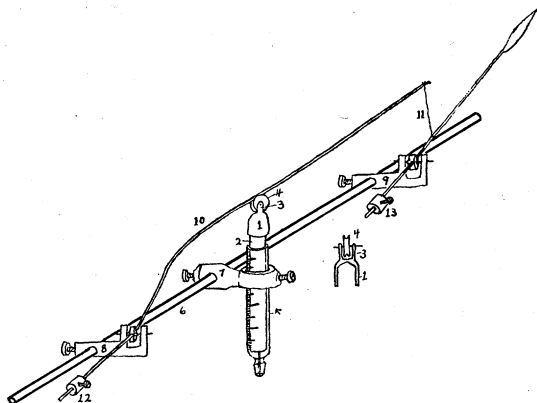


FIG. 1

The basic part of the apparatus (Fig. 1) is a piston made from a portion of a tuberculin syringe. A brass cap (1) was drilled to fit the cut-off plunger (2) of the syringe and then turned on the lathe until very thin. A stem about one-third the diameter of the cap and 5 mm long was left projecting at the top. The projecting stem

was sawed and filed into a U-shaped fork (3) and fitted with a small, grooved pulley (4) which was held in place by a small pin of stainless steel wire. This cap was placed on the plunger (2), which was then inserted in the syringe barrel (5). The latter was cut shorter than the plunger to prevent the cap from striking it. This piston assembly was fixed to a small brass rod (6) by means of a clamp (7) made of brass and drilled vertically to fit the syringe barrel and horizontally to fit the rod.

Lever supports (8, 9) were made to fit the same rod, supplied with levers of aluminum wire, and fixed one on each side of the piston clamp. One aluminum wire lever (10) was bent to fit and placed in the groove of the pulley (4) on the piston cap. A thread (11) was used to attach the first lever to the second to magnify the movements.

Adjustment of the magnification of the movement could then be made by altering the distance between the first lever clamp (8) and the piston clamp (7) and also by varying the position of the thread (11) connecting the two levers. The weight of the levers may be nullified by using small cylindrical brass counterweights (12, 13) fastened by set screws on the short ends of the levers.

An hydraulic system is necessary for smooth operation of the piston assembly. Water is not satisfactory, since the small amount lost by evaporation from the piston will show as a gradual fall in the base line of the record. Mineral oil is perhaps best for filling the piston. As used for recording intraocular pressure changes, a small mercury manometer made of fine-bore glass tubing was interposed between the piston and the eye, oil being used on the piston side of the manometer and aqueous solutions on the other.

With a piston made of about one-third of a tuberculin syringe volume, changes up to about 0.2 cc may be recorded, giving an excursion on the record of 10-20 cm or