sions of a book of this character have been necessary in order to keep it up-to-date. Thus a second edition was issued in 1934, and now a thoroughly revised third edition has been prepared by Professor Richtmyer's colleague, Professor Kennard, with the assistance of several of his associates on the Cornell University faculty.

The large amount of new material which had to be included if the text was to live up to its title, raised the difficult question of what to leave out. This matter has been admirably handled by Professor Kennard on the general principle that those topics in classical physics which are adequately treated in available general texts might well be skeletonized to the parts essential for future reference or omitted entirely. Thus the historical introduction has been abbreviated from 77 pages in the first edition to 50 pages, and the chapter on electromagnetism has been confined to those topics which are requisite for the subsequent developments and which are not usually contained in general texts. A short chapter on relativity has been added, but the greater part of the book is devoted to quantum phenomena, their description and their explanation. The chapter on x-rays has been completely revised, that on nuclear phenomena greatly extended, and a new chapter on cosmic-rays has been added.

In spite of these extensive revisions the character of the book has remained true to the standards set by Professor Richtmyer in the original edition. No attempt is made to give a logically complete development of modern theoretical physics. Rather the reader's attention is focussed on the phenomena of modern physics and on the understanding of modern concepts. Simple derivations of theory—often only for special cases—are given, but all complicated analyses are omitted. In this way the reader is led rapidly to the forefront of modern physics without having his attention distracted by mathematical details, which his awakened interest may lead him to fill in at a later time.

The book is designed as a textbook rather than as a treatise or a reference work. The large number of diagrams and photographic reproductions and the excellent printing add greatly to its usefulness. It is safe to predict that this revision will be welcomed with the same enthusiasm which greeted the earlier editions.

LEIGH PAGE

SPECIAL ARTICLES

DEMONSTRATING THE PRESENCE OF SULFONAMIDES IN THE TISSUES[1]

A PREVIOUS communication[2] reported the development of new vehicles (penetrasols) which increased the penetration of a large variety of substances through grossly intact skin (for example, protein allergens, iron, mercury, bismuth and the sulfonamides). One of the methods used to demonstrate the penetration of sulfonamides into and through the skin was excision of the area of inunction and the use of a newly developed histochemical technique, which produces a color reaction with sulfonamide in the tissues. We herewith report the details of this new method, since we believe that it may find many general applications.

I. ANESTHESIA AND EXCISION

Before excising the skin, general ether anesthesia was used in the guinea pigs; and local procain anesthesia in the human subjects. In the latter, the procain solution was injected around and well away from the site of the sulfonamide inunction and eventual biopsy. This is an indispensable precaution, as the presence of injected procain solution, or even fluid alone, could produce erroneous results. The line of excision was made outside and at considerable distance from the site of inunction, in order to avoid displacement of the inunction-material with the scalpel.

II. FIXATION

The usual liquid fixatives can not be employed, as the sulfonamides might be washed out or displaced by the liquid. Dry formaldehyde *gas* was therefore chosen as the fixative. Paraformaldehyde (trioxymethylene) powder is spread over the floors of small glass beakers. These beakers are roofed either with a piece of gauze fixed by a rubber band or by a small glass lattice. The biopsy specimens are spread flatly on these sieves. The small beakers are then placed in a larger glass jar, the floor of which has also been covered with a layer of paraformaldehyde powder. The top of the jar is closed with a well-fitting glass cover. The large jar containing the smaller beakers is kept tightly closed throughout the period of fixation by means of several windings of adhesive tape around the line of closure. (The layer of paraformaldehyde powder on the floor of the small beakers and of the larger jar should be about 1 cc in depth.)

At room temperature, fixation takes place in two to twenty-four hours, depending on the size and thickness of the specimens.

Microtome cutting is best carried out immediately after fixation; but we have, on occasion, obtained

[1] This research was made possible by a grant from the Wallace Laboratories, Inc., New Brunswick, N. J.

[2] Herrmann, Sulzberger and Baer, SCIENCE, 96: 451, 1942.

good sections as long as ten to fourteen days after fixation, despite some visible shrinkage and discoloration of the gross specimen.

III. CUTTING

Frozen sections of 10 μ to 20 μ (average 16 μ) are cut with care to prevent the microtome knife from displacing the sulfonamide. The specimen should be kept in such a position on the freezing table that the epidermal surface is never directed toward the knife, but always toward the operator. (The tendency of the thin epidermis of animal skin to separate as a thin lamella before the cutting knife can be overcome by placing the specimen sagittally, at an angle of 90 degrees to the edge of the knife.)

IV. CHEMICAL REAGENTS AND PROCESS

After trial of many standard reagents and solutions for demonstrating sulfonamides, we found that for our purposes the most suitable reagent and solution was Ehrlich's reagent, *i.e.*, p-dimethylaminobenzaldehyde, as used by A. E. A. Werner[3] and others for quantitative analyses of sulfonamides in biologic fluids. The optimal reactions were obtained with the following solution:

P-dimethylaminobenzaldehyde	1.0 g (grams)
Absolute alcohol	95.0 cc
Concentrated HCl	5.0 cc

The aldehyde must be as pure as obtainable, with little color or, at most, a light buff shade. Freshly prepared solutions are best, but a solution not over two or three weeks old may suffice, if it has been kept in ground-glass-stoppered, amber-colored flasks.

The frozen sections, obtained in the manner described, are placed directly on a clean slide, by means of a dry camel's-hair brush. Two drops of the above p-dimethylaminobenzaldehyde solution are dropped on the section. After three to five minutes, one to two drops of absolute alcohol containing 5 per cent. concentrated HCl are added. From this point on the procedure must be carried out as quickly as possible. The sections are dried by careful absorption of excess fluid with small pieces of filter paper or before a slowly rotating fan or both. (Warming is not advisable.) When dry, the sections are covered with a high concentration of dammar resin in xylene, as follows:

Dammar resin	10.0 g
Xylene	10.0 g

The cover slip is then superimposed. Air bubbles under the cover slip must be removed; then the edges are sealed with liquid paraffin melting at 56° C.

[3] A. E. A. Werner, *Lancet*, 136: 18, 1939.

This method demonstrates the sulfonamide in the tissues as a colored precipitate, ranging from lemon-yellow to orange, depending on the concentration of the drug.

This colored reaction-product bleaches out, particularly rapidly in the presence of air. Even with all precautions to expel air bubbles, with the thick dammar resin employed and with the paraffin sealing of the edges, microscopic examinations and microphotographs should be made as early as posible, and never more than three to four hours after the reaction has occurred.

V. CONTROLS

Several controls are indicated in order to make sure that the yellow to orange color is actually due to a chemical reaction between the sulfonamides and the reagent, and not due to artefacts or to the presence of preformed yellowish or orange material. These controls include:

(1) Examination of sections of tissue which has been exposed to sulfonamide, using all the procedures outlined, with the exception of the treatment with the p-dimethylaminobenzaldehyde solution.

(2) Examination of sections of tissue which has not received sulfonamide inunction and which is known to be free of sulfonamide; or, in the case of systemic administration of the drug, of tissue from the corresponding organ of an individual who has not received the drug. These sections are, of course, to be processed and treated in exactly the same way as the specimen being investigated for presence of sulfonamides, including treatment with the solution of p-dimethylaminobenzaldehyde.

Comparative examination of these control sections and the section being investigated will reveal the presence or absence of (a) preformed colored material and (b) material other than sulfonamide which might give similarly colored reaction-products with p-dimethylaminobenzaldehyde (primary aromatic amines, urobilinogen, urea, etc.).

Our studies indicate that the method just described will be of value in defining the presence, route and localization of sulfonamides in many different tissues (for example, skin, urethra, other mucous membranes, tonsils, bone marrow, etc.); but is not likely to be useful in tissues such as those of the liver, gall bladder, etc. Of course, the localization of the colored material and the intensity of the color can be expected to give only indications of quantity and site, rather than absolute values.

Nevertheless, employing this method in our studies of percutaneous absorption of locally applied sulfonamides in various penetrasol-type vehicles, we have been able to demonstrate that certain of these vehicles favor the concentration of the drug in horny and follicular structures, while others permit penetration

and more or less equal distribution through all the cutaneous tissues. Details of these studies on different types of skin-penetrating vehicles, as well as further discussion of the histochemical method itself, will be published shortly.[4]

GEORGE M. MACKEE
FRANZ HERRMANN
RUDOLF L. BAER
MARION B. SULZBERGER

SKIN AND CANCER UNIT,
NEW YORK POST-GRADUATE SCHOOL AND HOSPITAL,
COLUMBIA UNIVERSITY

ETHER AND METABOLISM IN THE CEREBRAL CORTEX[1]

JOWETT and Quastel[2] have concluded that anesthetic concentrations of ether do not inhibit by more than the experimental error the respiration of the gray matter of the cerebral cortex of the rat or guinea pig. That another metabolic process might be affected by ether was suggested by an observation made on slices of cerebral cortex from cats under ether anesthesia.[3] Measured in phosphate medium, the lactic acid output was greater in slices from these animals than from controls. The present report is concerned with the effect of ether applied *in vitro* on the oxygen uptake and lactic acid output by cat cerebral cortex slices.

The procedure was as follows. First, the oxygen uptake in Ringer-phosphate-glucose medium in six vessels with oxygen in the gas phase, was observed for 45 minutes. Second, a quantity of medium with or without ethyl ether dissolved in concentrations up to 40 mg per cc was added to each vessel through the venting plug by means of a syringe fitted with a long needle, to make a final concentration of up to 9 mg per cc. Third, oxygen uptake was followed for from two to six hours more. The details of the manometric technique were as reported elsewhere[4] except that the shaker speed was 80 cycles per minute. Fourth, metabolism was stopped by tipping in HCl solution from the side arm, and the concentration of ether in the medium was estimated manometrically with a temperature change of 1.5° by the method of Jowett.[5] Fifth, the lactic acid that accumulated in each vessel was determined by the method of Barker and Summerson.[6] In the control vessels the lactic acid content was higher at the end of the experiment than at the end of the first 45 minutes.

Data for 15 cats and the standard errors of the means are given in Table 1, in four groups according to ether concentration. In the first group the concentration was such that it would not produce unconsciousness if present in the blood.[7] In the second group the ether concentration would produce full anesthesia. The higher concentrations would be lethal. Oxygen uptake is recorded as a percentage of the initial rate for each vessel corrected for any change in rate with time in the control vessel, to which an addition of medium without ether had been made. Lactic acid is reported as the difference between the amounts in the vessel containing ether and in the control vessel, divided by the number of hours of exposure to ether.

TABLE 1

EFFECT OF ETHER ON THE OXYGEN UPTAKE AND LACTIC ACID OUTPUT BY SLICES OF CEREBRAL CORTEX OF THE CAT IN RINGER-PHOSPHATE-GLUCOSE MEDIUM

Ether concentration			Oxygen uptake						Extra lactic acid output	
Range	Mean		1st hour		2nd hour		3rd hour			
mg/cc	mg/cc	M × 1000	c.*/o.	Per cent. of control	c.*/o.	Per cent. of control	c.*/o.	Per cent. of control	c.*/o.	mg/g/hr
0.025–0.4	0.2	2.7	6/14	97 ± 1.85	6/14	95 ± 2.09	3/3	90 ± 6.46	6/14	4 ± 0.93
0.7 –1.6	1.3	17.5	9/15	100 ± 1.34	9/15	101 ± 1.29	5/11	100 ± 2.00	9/15	3 ± 1.19
2.0 –5.3	3.7	50	7/14	103 ± 1.73	7/14	106 ± 1.89	5/8	111 ± 3.63	7/14	5 ± 0.62
6.7 –9.6	8.4	113	6/9	107 ± 6.1	6/9	86 ± 8.8	6/9	43 ± 6.9	6/9	14 ± 2.75

* Number of cats/number of vessels.

[4] MacKee, Herrmann, Baer and Sulzberger, *Jour. Clin. and Lab. Med.*, 1943 (in press).

[1] Aided by a grant from the Milton Fund of Harvard University to Henry K. Beecher.

[2] M. Jowett and J. H. Quastel, *Biochem. Jour.*, 31: 1101, 1937.

[3] H. K. Beecher and F. N. Craig, *Jour. Biol. Chem.*, 148: 383, 1943.

[4] F. N. Craig and H. K. Beecher, *Jour. Neurophysiol.*, 6: 135, 1943.

[5] M. Jowett, *Biochem. Jour.*, 31: 1097, 1937.

[6] S. B. Barker and W. H. Summerson, *Jour. Biol. Chem.*, 138: 535, 1941.

[7] H. W. Haggard, *Jour. Biol. Chem.*, 59: 783, 1924.