

the Montgomery estate; 500 rose bushes from Bobbink and Atkins; 700 packets of seed selected and purchased by Stanley Ranger, and cannas, iris, dahlias, chrysanthemums, peonies and other plants for display purposes from a number of growers.

Gifts of money included \$3,140 for the permanent

endowment, \$410 for current expenses and \$1,942 for restricted purposes.

The library, which is the largest combined horticultural and botanical library in America, now numbers 43,429 bound volumes, of which more than 1,000 were added in 1933.

SCIENTIFIC APPARATUS AND LABORATORY METHODS

AIDS IN THE STUDY OF DENTAL CARIES WITH ALBINO RATS

RECENT reports¹ on the study of dental caries in the rat indicate that in such studies macroscopic examinations of the teeth were made only at autopsy. It is obvious that periodical examinations of the teeth made while the animal is still alive would be very advantageous, as then the course of the lesion could be followed.

Such observations have been carried out in this laboratory for the past two years, using the following procedure: The assistant holds the rat as indicated in the diagram (Fig. 1). With the rat grasped and

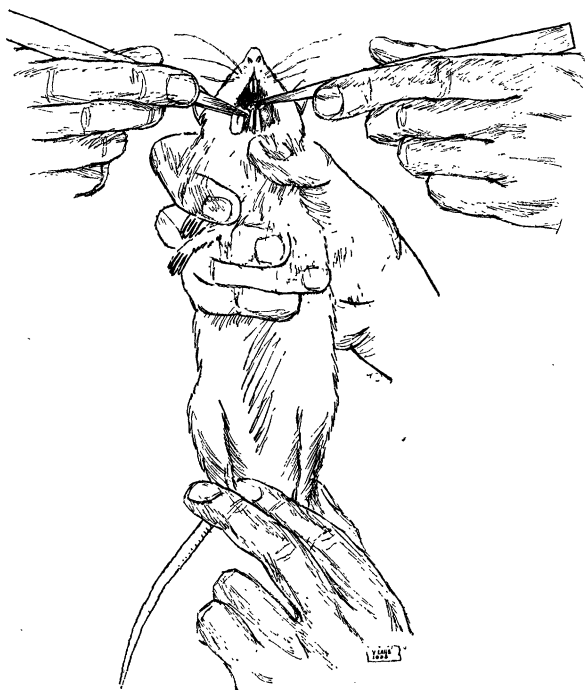


FIG. 1.

held in this position there is no danger to the assistant or the observer. The observer, wearing a Murphy head light and a Beebe binocular loupe, holds a pair of 3½ inch blunt forceps in each hand. Assuming that the left side of the rat's mouth is to be examined,

¹ H. Klein and E. V. McCollum, *Jour. Dent. Res.*, 13: 69, 1933; M. C. Agnew, R. G. Agnew and F. F. Tisdall, *Jour. Amer. Dent. Assoc.*, 20: 193, 1933; D. E. Shelling and D. E. Asher, *Jour. Dent. Res.*, 13: 363, 1933.

the observer opens the animal's mouth by using the forceps in the right hand. The forceps in the left hand is now used to pull out the rat's tongue gently. The forceps in the right hand is now used to push the left cheek of the animal aside, thus permitting examination of the teeth on the upper and lower jaws. For observations on the opposite side this process is, of course, reversed.

The question of inorganic elements in blood serum in relationship to dental caries has also assumed importance recently.² It is, of course, desirable in such studies to obtain the maximum amount of serum in order to permit accurate determinations.

We have used the following modification of the method described by Kramer and coworkers:³ With the animal lightly anesthetized by ether, the skin is removed from the neck region by cutting with scissors.

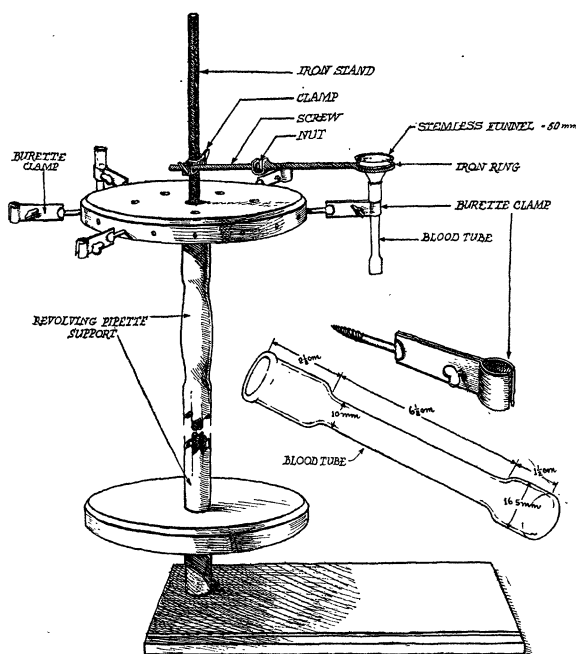


FIG. 2.

² H. Klein and E. V. McCollum, *SCIENCE*, 74: 662, 1931; M. C. Agnew, R. G. Agnew and F. F. Tisdall, *Jour. Amer. Dent. Assoc.*, 20: 193, 1933; D. E. Shelling and D. E. Asher, *Jour. Dent. Res.*, 13: 363, 1933; I. Neuwirth and P. Brandwein, *Jour. Dent. Res.*, in press.

³ B. Kramer, M. J. Shear and J. Siegel, *Jour. Biol. Chem.*, 91: 271, 1931.

Using forceps, the muscles in the neck region are separated in the mid-line until the trachea is exposed. The internal carotid artery is now isolated and a ligature passed under it. The animal is now held head downward and, using the ligature as a guide, the artery is cut and the blood permitted to flow into the tube as shown in the arrangement in Fig. 2. We have found that the tube illustrated permits obtaining more serum than is ordinarily possible in the average centrifuge tube.

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THE SILVER IODIDE TEST FOR HYDROCYANIC ACID

It is not infrequently desirable to ascertain the presence of hydrocyanic acid in faint traces, with the use of a delicate, fairly rapid and simple test. The ready recognition of this compound is of use occasionally to agriculturists, toxicologists, health officials, fumigators of ships and buildings, and to the coroner, as well as to numerous scientific investigators engaged in various types of work.

Recently the writer has had occasion to make tests for traces of HCN in connection with current studies of cyanogenetic glucosidases present in certain marine organisms. With the cooperation of Dr. C. E. ZoBell of this institution, a number of marine bacteria and fungi are being studied with reference to the capacity of these forms to hydrolyze the cyanogenetic glucoside amygdalin. Similar studies are being made of the alimentary tracts of certain marine invertebrates.

Guignard's test, which consists of the suspension of sodium picrate papers over the solution or culture under investigation, is very delicate, but according to the numerous writers, not highly specific for HCN. This test, however, is employed as a preliminary one in our laboratories to indicate the presence of free HCN in various microorganism cultures or enzyme systems. If the compound is present, the bright yellow color of the alkaline picrate paper is converted, through various stages of yellow-orange, orange and red orange to the brick color of the reduced compound, sodium picramate.

In order to confirm the presence of HCN, and to demonstrate that it, and not other substances, is responsible for the reddening of the papers, an additional delicate and specific test has been developed, based, in principle, upon the quantitative method for determining HCN described and used by Roe¹ and later by Bishop.² This method, used in determining

the amount of HCN present in certain leaves, kernels, and pure cyanogenetic plant glucosides, consists in aerating the solution containing the digest, and conveying the air thence into dilute potassium hydroxide to catch all of the HCN, aeration being continued until all of the compound has been carried over. The alkaline cyanide solution is finally titrated for CN⁻, using the general method of Liebig, with very dilute silver nitrate solution, employing potassium iodide as an indicator. With proper lighting and a black background the end point can be determined very sharply as the first permanent faint cloud of AgI appears, indicating that all of the CN⁻ has been taken up in forming the complex salt potassium argenticyanide KAg(CN)₂.

The procedure used in our own work, for confirming the presence of HCN indicated by Guignard's test, consists in a modification, perhaps better termed an inversion, of Roe's method.

To a thoroughly cleaned test-tube are added first one drop of 5 per cent. KI solution, and one drop of .001 M AgNO₃ (solutions as dilute as .00025 M can be used if only the faintest traces of HCN are suspected), and then one cubic centimeter of 5 per cent. KOH solution. A faint bluish cloud of AgI is now present. Air is drawn through the system or culture which is being tested for HCN, and thence into the alkaline silver iodide suspension. If HCN is present, the KCN formed dissolves the AgI, rendering the solution perfectly clear. The test is specific for HCN, since H₂S, NH₃, HCNS (NaCNS), and other bacterial catabolites were found to exert no visible effects upon the silver iodide, which is soluble only in such non-volatile compounds as Na₂S₂O₃, NaCl, etc. It is important that the reagent be prepared freshly for each test, since AgI does not remain in the colloiddally dispersed condition described under the circumstances for longer than a day at most. The limit of delicacy is about 1 part HCN in 2,000,000, when .00025 M AgNO₃ is used under ordinary laboratory conditions. Without doubt the perception of even fainter clouds of AgI with the use of special nephelometric instruments would render the test still more delicate.

An attractive feature of this method, besides its specificity, delicacy and simplicity, is that, should a positive test for HCN be found, quantitative determination of the compound can be made forthwith if desired by merely continuing the aeration without altering the system or interrupting the process, thus making use of the chemical method perfected by Roe.

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¹ J. H. Roe, "The Estimation of the Hydrogen Cyanide Content of Amygdalin by the Aeration Method," *Jour. Biol. Chem.*, 58: 667, 1924.

² L. R. Bishop, "The Estimation of Cyanogenetic Glucosides," *Biochem. Jour.*, 21: 1162, 1927.