

must have appeared excessive in relation to rejuvenating a bull for only a breeding season or two of effective service. Not so in our own era of artificial insemination and of egg implantation into foster mothers, when the reproductive power of a progenitor is spread over a much larger herd or flock.

Voronoff's method should prove of especially timely importance for prolonging the usefulness of high-priced livestock, which, shipped abroad under the various ECA, Point Four, and private (e.g., IBEC) programs of aid to underdeveloped countries, may become prematurely sterile in an adverse climatic environment.

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Potentiometric Measurements in Colloidal Systems

THE letter by Karol J. Mysels (1) in some ways clarifies the issues raised by Jenny *et al.* (2), yet in other respects it may cause further confusion. Dr. Mysels at the time of writing probably did not have available the detailed criticisms by Marshall (3), and by Peech and McDevitt (4) of a paper by Coleman, Williams, Neilson, and Jenny, entitled "On the Validity of Interpretations of Potentiometrically Measured Soil pH" (5).

It seems now to be agreed that a proper criterion as to the condition under which the KCl liquid junction potential is small is that the KCl solution shall be concentrated in comparison with the colloidal system (all contributing ions included). Thus, since saturated KCl (3.5 *M*) bridges are normally employed, we may dismiss from consideration all colloidal systems having ionic concentrations below, say, 0.1 molar. More concentrated systems, such as beds of granular cation exchange resins, call for special consideration. The Teorell-Meyer-Sievers theory can well be applied to such cases when a proper steady state has been reached (see below).

As Mysels points out, a crucial factor in concentrated colloidal systems is the contact with the KCl bridge. However, he seems to have missed one important aspect; namely, that the salt solution should make contact with a representative cross section of the system as a whole. Valid potentiometric interpretations depend greatly on this condition being fulfilled. Different results may readily be obtained in coarse granular systems, depending upon the kind of contact attained.

This was well illustrated here some months ago in

an experiment performed by Wm. J. Upchurch. A column of 60-mesh cation exchange resin IR 120 (potassium-saturated) was supported on a sintered glass filter disk. The pores of the latter were filled with saturated KCl solution, with a reservoir under the filter connected to a side tube. By raising the level of KCl in the side tube it could be brought into contact with a complete cross section of the exchanger. After this was done, a saturated calomel electrode was inserted in the side tube. Then a second saturated calomel electrode of the Beckman type was pushed into the upper part of the column of exchanger after the manner of Jenny *et al.* Thus we apparently had the simple system, Calomel | Sat. KCl | K Exchanger | Sat. KCl | Calomel, which of course should give zero potential under proper conditions. This Beckman type of electrode, however, furnishes KCl for the liquid junction by a very slow gravity flow down an asbestos fiber in a glass capillary. The conditions were evidently very far from ideal because a maximum potential of 35 mv was observed, which slowly came down to zero over a total time of 10 days.

When electrodes of improved design were employed, this time interval was greatly reduced; but any kind of small-bore, upturned tip that depends on diffusion to make contact requires hours for the true steady state to be reached. Granular systems thus require special design of the KCl junctions in order to give a truly representative contact in a short time.

Colloidal suspensions of particles $< 1\mu$ naturally do not cause difficulties of this order of magnitude. Nevertheless, instantaneous readings cannot always be relied upon, and the KCl should be given time to form a true boundary. In our experience 15–20 min amply suffice, where the KCl makes contact at the end of a well-defined tip of $\frac{1}{2}$ –2-mm cross section.

The "perched" potential at first obtained with the resin exchanger evidently includes the average work done in moving an ion from the granular system to a layer of water interposed between the KCl and the granules. Because the granular system contains water as well as resin, this "perched" value does not represent the total phase potential of the resin against water; it will be somewhat less, depending on the porosity.

Finally, as regards the potentials to be expected when saturated KCl bridges come into true steady state contact with concentrated colloidal systems, the Meyer and Sievers theory indicates relatively low values compared with the phase potentials and "perched" potentials just discussed. If the effective cationic concentration *A* of the resin continuous phase is taken as equal to that of the ions in saturated KCl, then the Donnan potential according to the Meyer and Sievers theory is about 12 mv. Higher values than this could only arise if *A* effectively exceeded 3.5 *M*, and in view of the distribution of ionizing sites in exchange materials this would be barely possible. The liquid junction potential in the interior of the resin is proportional to the difference in mobility between the ions of the salt and can therefore be made

vanishingly small when the ions are equal in this respect. Thus with KCl the only potential of significance when a steady state boundary has been achieved is of the kind evaluated above.

The limiting case derived from the Meyer and Sievers theory by Mysels corresponds precisely to the experiment described above, which gave rise to the "perched" potential. The steady state potential between saturated KCl and any exchange material is likely to be much smaller.

Dr. Mysels' letter fulfills a useful purpose by drawing attention to the fact that we must make up our minds what kind of junction we are concerned with in any particular experiment. It seems to the writer that a junction with a representative cross section of the system under investigation is desirable. Conclusions drawn from such measured potentials refer then to the system as a whole. The colloid chemist's task, in general, is the interpretation of such systems.

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A Practical Laboratory Method for Mass Culturing Bacteria¹

BACTERIOLOGICAL investigations involving an elucidation of the chemical constituents of bacteria are frequently impeded by lack of suitable facilities for growing sufficient quantities of cells. The method described below requires no unusual laboratory apparatus, and it is mobile, inexpensive to construct, and applicable to mass cultivation of a variety of microorganisms for research purposes. The method has been used here for the past two years for the cultivation of both pathogenic and nonpathogenic bacteria. Good yields have been realized in a minimum of time, without undesirable degenerative changes occurring in the cells, and with no environmental contamination.

The culture container is a 7-gal, cylindrical Pyrex jar resting on a rubber-cushioned sheet-iron base (Fig. 1). Four tierods project from the base through matching holes drilled in the $\frac{1}{4}$ " aluminum cover. The latter has a collar welded to its under surface around which is fitted a $\frac{1}{4}$ " rubber gasket. A tight seal around the lid is obtained by tightening the tierod nuts on top of the cover with a wrench.

The cover has holes suitably drilled in it to receive a mercury seal, air and alkali tubes, glass and calomel electrodes, thermoregulator (glass rod type), heating element (300-w Calrod), thermometer (25°-45° C), mercury manometer, and siphon tube, all mounted in rubber stoppers (Fig. 2). A housing consisting of a 2 $\frac{1}{2}$ " copper pipe nipple with flanges on either end

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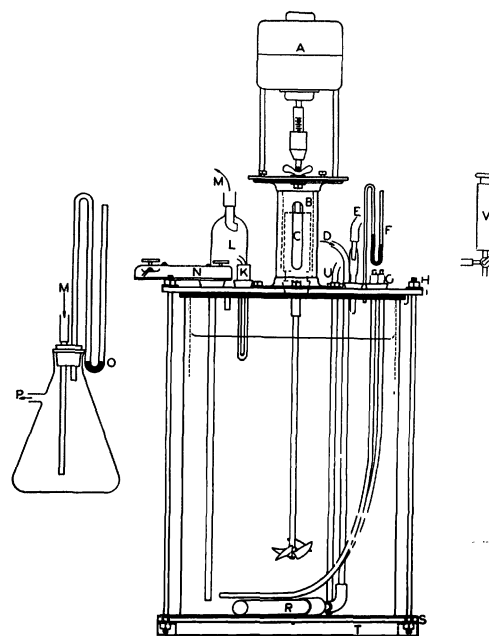


FIG. 1. Schematic representation of culture unit. A, motor; B, motor support; C, mercury seal; D, air "in" tube; E, to alkali reservoir; F, mercury manometer; G, heater; H, tierod cover nut; I, lid; J, rubber gasket; K, electrode; L, froth trap; M, exhaust air; N, thermoregulator; O, relief mercury manometer; P, to water pump; R, filter candle; S, rubber-cushioned base plate; T, wooden base; U, siphon tube (to sampler, for introduction of medium ingredients, for harvesting); V, sampler (30-ml syringe with 2-way Luer stopcock).

is bolted to the center of the cover to support the stirrer motor (1/20 hp, 1400 rpm). The latter is mounted as indicated (Fig. 2) to allow for proper alignment with the stainless steel stirrer through the mercury seal.

In operation, the thermometer and electrodes are sterilized separately in a quaternary ammonium salt and maintained in tubes of sterile broth. Cotton plugs are placed in the cover holes occupied by these, the motor removed, and the otherwise assembled apparatus is placed on its side in an ordinary autoclave and sterilized. After sterilization, the thermometer and electrodes are aseptically inserted into the cover, the motor attached, and the proper amounts (about 24

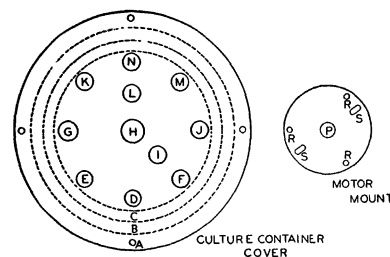


FIG. 2. Culture container cover. B, rubber gasket; C, collar; holes for: A, tierods; D, E, electrodes; F, air out; G, air in; H, heater; I, mercury seal; J, manometer; K, siphon; L, alkali; M, thermometer; N, thermoregulator. Motor mount: O, taped holes for leveling screws; Q, slots for motor mount bolts; P, drilled holes for stirrer shaft.