is known as "the phase-boundary potential." Leaving aside the ambiguity in the magnitude of this potential which has concerned many investigators and even compelled some theoreticians to consider this potential as inadmissible (5), an electrical state can arise which is associated with the sharp discontinuity in concentration of the mobile species and the space charge due to asymmetry in the positive and negative ions. The partitioning effect thus *must* be taken into account for a complete and physically meaningful solution of the Poisson-Boltzmann equation throughout the region of the membrane phase. This analysis has been carried out by Verwey and Niessen for the equilibrium state (1966. *Biophys. J.* **6**:371).

For those investigators who are studying the electrical properties of Rudin and Mueller films (phospholipid-hydrocarbon films) similar considerations are particularly relevant.

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The Electrical Conductance of Semipermeable Membranes

Dear Sir:

I had the privilege of reading the two papers of Dr. Bruner on the electrical conductance of semipermeable membranes (1965. *Biophys J.* **5**:867, 887). In addition, I had the opportunity to study the preprint of Dr. Bruner's third paper of this series and reflect on Dr. Mauro's comments regarding the assumptions invoked in these papers (1966. *Biophys. J.* **6**:367). Basically, Dr. Bruner's development begins with the combination of Poisson's equation with classical equation for ion current density. Since subsequent developments in his procedure have led to differences of opinions about the validity of some of the author's assumptions, it may be appropriate to point out certain relevant difficulties that one has to overcome in theoretical development of this kind. In my opinion, only then, we will be able to resolve satisfactorily these differences of opinions.

The recognition of discontinuity in dielectric constant at the interface compels one to include (and not discard) the image potential terms and corrections to local activity coefficients (see Buff and Stillinger. 1963. *J. Chem. Phys.* **39**:1411) to the classical equations, in the determination of concentration profiles in inhomogeneous boundary regions. Our understanding of equilibrium state of such inhomogeneous fluid regions is far from complete and one should appreciate the efforts of Levine, Bell (1960. *J. Phys. Chem.* **64**:1188), and Loeb (1951. *J. Colloid Sci.* **6**:75), before introducing assumptions regarding the continuous or discontinuous nature of activity or concentration of species across the interface and extension of analysis to steady states.

The assumption (*e*) of paper I, which states that the mobilities of ions in surrounding solution are infinite, which enables the author to conclude that $\Gamma^{\pm} = 0$, is not reasonable since surrounding solutions have finite conductance.

Since none of the above-mentioned difficulties are insurmountable, the initial task should be to understand equilibrium state of inhomogeneous fluid boundary interfaces before analyzing transport processes in these systems in order to preserve consistency.

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An Effect of Cell Shape on Apparent Volume as Determined with A Coulter Aperture

Dear Sir:

The Coulter counter (Coulter, 1956) has been used extensively for the determination of total concentration of cells in suspension culture, as well as for the determination of volume distribution spectra. Gregg and Steidley (1965) have studied the relationship between particle volume and Coulter pulse amplitude and have shown that particle shape affects the apparent volume as determined from the increase in aperture resistance. We have recently observed anomalies in the determination of both cell number and cell volume for synchronized mammalian cells in suspension culture caused by failure of daughter cells to separate after mitosis is complete. That this leads to a low value of cell concentration is obvious; the effect on the volume spectrum is more subtle.

Chinese hamster ovary (CHO) cells were selectively removed from glass-grown monolayer cultures by a modification of the method of Robbins and Marcus (1964), using a mechanical shaking machine (Tobey, Anderson, and Petersen, 1967). The detached cells (shaken off in low calcium F-10 medium) were set up as suspension cultures in spinner flasks and were studied during the ensuing division wave.

The selected populations were initially almost entirely mitotic (mitotic fractions ranging from 0.85–0.98) and were very tightly synchronized, as evidenced by an abrupt drop of the mitotic fraction to less than 0.05 during the first 20 min in suspension culture. Total cell count rose much more slowly and depended strongly on the intensity of mixing (repeated pipetting) to which the suspension was subjected. The earliest separation of daughter cells which could be obtained was about 10 min after completion of the mitotic wave—a measure of the minimum temporal difference between the dissolution of the mitotic apparatus and cell separation, fiducial marks in cell life cycle analysis (Tobey et al. 1966). With minimal pipetting, a population could be obtained with over 80% of the cells as unseparated pairs (as determined by visual scoring of microscopic fields) for periods of some 40 min following mitosis.

Cell volume distribution spectra were measured during this period using a 90 × 200 mi-