IN THE LABORATORY

Studies on an Artificial Kidney: I. Preliminary Results With a New Type of Continuous Dialyzer[1]

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A rational approach to the treatment of acute renal insufficiency demands the removal from the body of retained metabolic waste products. This has been attempted by plasmapheresis, exchange transfusions, diaphoresis, and purgation, all of which have been abandoned as impractical. Continuous lavage of the intestine has also been advocated (*12, 14, 16*), but such small amounts of nitrogen can be thus removed that its usefulness is doubtful (*15*). Peritoneal lavage has been successful in animals (*1, 4, 15*) and in man (*5, 6, 7*), but the attendant danger of infection, clogging of the draining tubes, and the necessity of almost continuous operation have forestalled wide application.

Abel, Rowntree, and Turner (*2*) in 1914 first devised an apparatus for the removal of diffusible substances from the circulating blood of living animals by dialysis. Although they realized the possible usefulness of the method in the treatment of uremia, only recently have Kolff (*9, 10*), Alwall (*3*), Murray (*13*), and others (*11*) made serious attempts to apply it clinically. Although differing in constructional details, all of their "Artificial Kidneys" are characterized by their large size and the use of long lengths (100′) of cellophane tubing, necessitating complicated procedures for assembly and sterilization.

Although the above methods have not yet been generally accepted for routine use, they have met with sufficient success to demonstrate that life can be prolonged during temporary acute renal insufficiency through the removal of retained metabolic waste products and the adjustment of acid-base, electrolyte, and water balance.

The present report describes preliminary results with a new design of a continuous dialyzer (Fig. 1) which consists of a variable number of units, connected in parallel, each unit consisting of a single sheet of Cellophane and two rubber pads, each $12'' \times 18'' \times \frac{1}{4}''$. The Cellophane is sandwiched between the two rubber pads, the inner surfaces of which are finely grooved. The grooves on one of the pads serve as small "vessels" through which blood is allowed to flow under arterial (or other) pressure. The grooves of the other pad (on the other side of the Cellophane) carry dialyzing solution moving in the opposite direction. Thus, a single kidney unit is formed which may be connected with as many similar units as desired by means of interconnecting passages provided for within the rubber pads themselves.[3] The desired number of units are held firmly between two flat steel plates drawn together at the edges with thumbscrews. The entire assembled apparatus, ready for use, may be sterilized by autoclaving. Each kidney unit presents a surface area for dialysis of 840 cm², contains 45 ml of blood, and, under optimal conditions, is capable of removing 0.5 gm of urea nitrogen/hr from a blood urea nitrogen level of 150 mg/100 ml. Preliminary experiments have indicated that a similar dialyzing area may be obtained with the "kidney unit" holding much less blood.

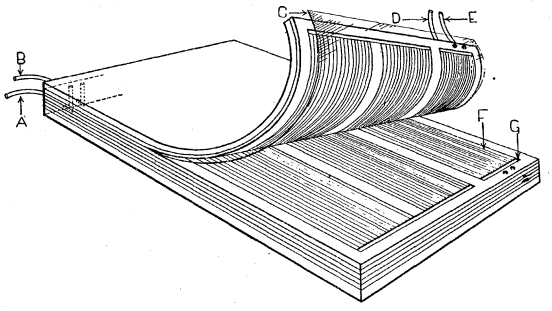

FIG. 1. Diagram of continuous dialyzer. The top rubber pad has been bent back to show the relative position of the Cellophane (C) and the fine grooves (F) in the bottom rubber pad (G). A, B, D, E are inlet and outlet tubes for blood and dialyzing solution.

After considerable preliminary work, 5 bilaterally nephrectomized dogs were successfully dialyzed. Using heparin as an anticoagulant,[4] the blood was taken from a cannula in the femoral artery, passed through the dialyzer, and returned to the femoral vein. The dialyzing solution was prepared to contain the same electrolyte concentration as extracellular fluid—namely (in meq/liter), sodium, 142; potassium, 5; calcium, 5; magnesium, 3; and chloride, 113; bicarbonate, 31; lactate, 8; phosphate, 3—and sufficient dissolved carbon dioxide to keep

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[4] In order to retard blood clotting further, the rubber pads are coated with a nonwetting film of silicone resin, for which we are indebted to W. R. Collings and W. Pedersen, of the Dow Corning Corporation, Midland, Michigan.

the pH at 7.3–7.4. The results have been summarized in Table 1.

In all cases the blood urea nitrogen was appreciably decreased, and significant amounts of urea were removed. In those animals in which the acid-base balance was disturbed, there was a tendency for return to normal. For example, dog No. 20 had repeatedly vomited, and before dialysis the plasma chloride was 86 meq/liter and the CO_2 content 92 volumes %, indicating a moderately severe metabolic alkalosis. Following dialysis the values were restored to 97 meq/liter and 68 volumes %, respectively. The animals showed no apparent harmful effects from the procedure, and in several cases clinical improvement was noted. However, we have not yet been successful in keeping the animals alive for a significantly longer period than the controls.

The apparatus is also useful for many problems in laboratory dialysis. For example, during the preparation of renin (*8*) from hog kidneys, a solution of the active principle (volume, 8 liters from 3.5-kg kidneys) is dialyzed for 3 days in 3″ Visking Cellophane tubes in order to remove trichloroacetic acid and other salts.

TABLE 1

REMOVAL OF UREA FROM NEPHRECTOMIZED DOGS BY EXTERNAL DIALYSIS

Dog No.	Hrs after nephrectomy	Wt. of dog (kg)	No. of kidney units	Blood urea nitrogen (mg/100 ml)		Hrs of dialysis	Urea removed (gm)
				Before dialysis	After dialysis		
14	72	10.5	6	152	54	7.0	18.8
16	60	8.6	4	216	57	5.6	23.1
17	72	8.0	4	...	88	4.5	21.8
19	84	25.0	8	184	81	5.6	38.0
20	60	16.4	8	179	46	7.6	31.7
20	96	15.0	8	130	80	2.5	...

Using 6 units of the present dialyzer, the same efficiency of dialysis was accomplished in 20 hrs. If more units had been used, the time would have been proportionately shorter. The dialyzing water was cooled by passing through copper coils immersed in an ice bath, and the dialysis was carried out at 2° C without the necessity of working in a large, cold room.

In summary, a continuous dialyzer has been constructed which should find applications both as a laboratory tool and as an "Artificial Kidney." Advantages include a large dialyzing surface relative to the volume of contained solutions, efficient dialysis because the fluids on both sides of the Cellophane are in a thin film and rapidly moving countercurrent to one another, and easy sterilization of the completely assembled unit. The apparatus shows promise for the treatment of acute renal insufficiency.

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Sensitivity of the Ninhydrin Reaction in Paper Partition Chromatography[1]

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The method of Consden, Gordon, and Martin (*1*) for the qualitative separation of amino acids by partition chromatography on filter paper is being used by the authors in studies of the amino acids in the blood and tissues of insects. This technique is particularly adaptable to physiological investigations with insects because it requires only micro quantities of the substances to be analyzed. Most of the free amino acids present in 25 microliters of insect blood can be qualitatively identified by this method.

In order to estimate quantitatively the free amino acids present in insect blood and tissues, and in order to determine the quantities of blood or tissue extracts necessary to identify qualitatively all of the amino acids present, the sensitivity of several amino compounds to the ninhydrin reaction was determined. This was done after pure amino compounds had been run in the separation chambers under the same conditions as the biological fluids being analyzed. Measured quantities of freshly made, pure solutions were placed on the filter paper, run 28 hrs in water-saturated phenol in one dimension, dried, run for 60 hrs in the second dimension in a water-saturated 1:1 mixture of γ-collidine and 2,4-lutidine, and again dried. The temperature was held between 23° and 26° C during the runs. The paper was then sprayed with a 0.1% solution of ninhydrin in normal butanol and heated in an oven for 5 min between

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