to be indicative of suboptimal conditions in the tissue culture system used in the above mentioned investigations.

It is possible that the reasons for these discrepancies are connected with physiological differences between "attached" and "unattached" cells. Observations made in this laboratory indicate that these differences may be important at least in the case of chick embryo fibroblasts. When cell suspensions were prepared with the aid of 0.25% trypsin, either from chick embryos or from monolayers, the cells failed to attach to the walls of the glass vessels in the absence of serum, retained their globular form, and did not show any obvious metabolic activities such as production of acid in the presence of glucose. In the presence of small amounts of either horse or calf serum (0.1%-0.2%) the same cells attached themselves to the glass walls, took on the typical shape of fibroblasts, and caused acidification of the medium in the presence of glucose, even after removal of the serum by means of washing, and addition of medium from which serum was excluded.

In the system described by Levine and Sagik, although serum was present in the media, other conditions were such that the cells remained in suspensions, *i.e.* in "unattached" form during the period of virus multiplication. It might be that these conditions led to a shortened latent period and lowered virus yield.

Summary and conclusions. The multiplication of cytopathogenic strain of Newcastle disease virus in monolayers of chick embryo cells was studied. The latent period in the growth cycle of this virus was found to last 8-9½ hours, followed by a period of rise lasting 3-4½ hours. The average yield of virus per cell in the tissue culture was found to be approximately 300.

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A Method for Continuous Intravenous Infusion of Dogs Confined to Cages.* (23431)

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The availability of intravenous fat emulsions satisfactory for clinical use has necessitated reevaluation of the efficacy of complete parenteral alimentation. The ideal conditions for investigation of this problem in dogs required that normal animals, unrestrained but confined to metabolic cages, receive intravenous fluids at a constant rate throughout each 24 hour period of study. To fulfill these conditions, it was necessary to devise a method for continuous administration of fluids which would protect the intravenous tubing against mechanical injury resulting from the dog's activity. The swivel developed by Jacobs(1) for the continuous infusion of dogs confined to a room was unsuitable for our work; how-

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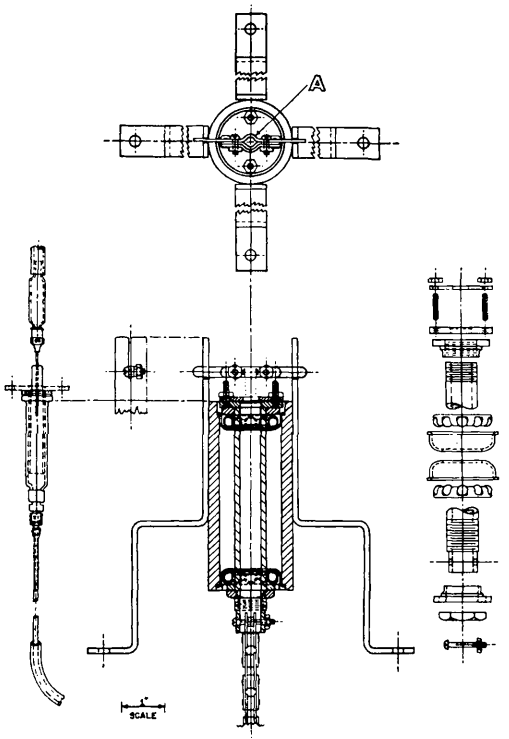

FIG. 1. Individual portions of the swivel assembly drawn to scale.

ever, its modification provided the basis for a simplified swivel which satisfactorily fulfilled the requirements for our studies.

Description of swivel assembly. Fig. 1 represents the individual sections of the swivel assembly drawn to scale. The principle of the assembly is establishment of continuity of the intravenous tubing between the dog and the fluid reservoir by a joint which permits fluid to flow without leakage while the tubing between the joint and the dog is free to rotate without twisting. A 2 cc Leur Lock syringe, its plunger converted into a hollow cylinder by removal of both ends, fulfills the requirements for such a joint. An 18 gauge needle, introduced through a rubber plug which seals the upper end of the plunger, is connected by an adapter to the intravenous tubing from the infusion bottle. A 13 gauge needle attached to the Leur Lock fits snugly into the end of the tubing which connects the dog with the syringe.

The swivel consists of an inner cylinder, free to rotate within an outer cylinder, that is mounted at a convenient distance from the top of the cage. The syringe portion of the assembly, placed within the inner cylinder of the swivel, is supported by the lip of the syringe barrel, which rests upon the top of the inner cylinder. To prevent air from entering the system between the walls of the plunger and the barrel of the syringe, a small vaseline pack is wrapped around the portion of the plunger extending above the barrel. A metal disc is slid over the plunger and bolted to the inner cylinder to hold the vaseline gauze in place and to fix the syringe barrel to the inner cylinder. The plunger of the syringe is immobilized by the screw clamp A (Fig. 1) which is fixed to the stationary outer cylinder of the swivel.

A polyethylene catheter, introduced into the dog's external jugular vein through an incision in the side of the neck, is passed subcutaneously to the back of the neck where it emerges through a small stab wound and makes connection with the intravenous tubing

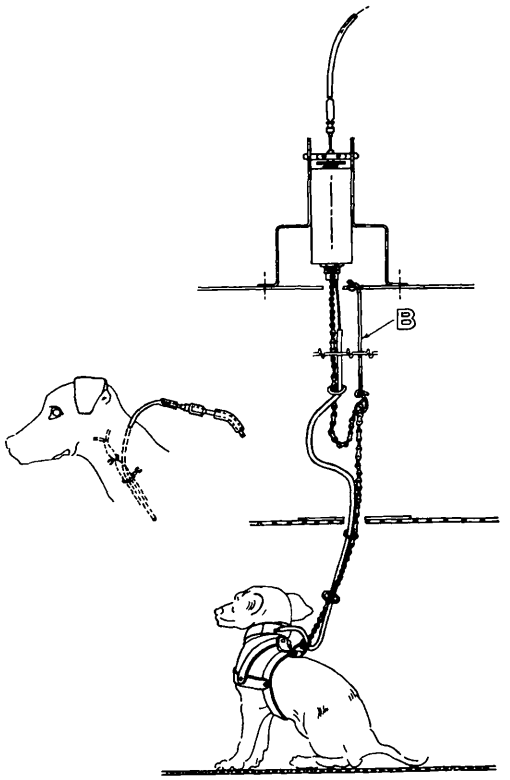

FIG. 2. A schematic representation of the dog's connection to the swivel.

from the swivel. A collar of tape which should not be applied tightly is used to secure the polyethylene catheter and its connection with the intravenous tubing.

The dog is placed in a loose-fitting harness which is connected to the inner cylinder of the swivel by a bicycle chain sufficiently long to allow the animal to lie down but which is shorter than the tubing between the dog and the swivel. Two points on the chain are connected by a piece of rubber tubing (B. Fig. 2) so that the excess chain and tubing are withdrawn from the cage when the dog is standing.

For protection, the intravenous tubing between the swivel and the dog is encased in heavy rubber tubing which is wired at multiple points to the bicycle chain except at the distal end where the excess tubing is allowed to accumulate. Slack in the tubing at this point eliminates the possibility of pull upon the connection between the intravenous tubing and the polyethylene catheter. When the dog lurches, his motion is thus stopped by the chain and no stress is placed upon this union.

Discussion. The swivel has been utilized in studies performed upon 10 dogs. These experiments ranged from 15 to 35 days in duration and were performed without mechanical difficulty. Only one experiment was terminated because the dog chewed the rubber tubing. The careful selection of gentle, quiet dogs will eliminate virtually all the difficulties one may expect with this mechanism. Two solutions were administered simultaneously and, in most experiments, rate of administration for each was controlled individually by glass stopcocks placed in the system between the fluids and swivel. Although this method was satisfactory, it required supervision because of fluctuations in rate of flow resulting from changes in the animal's position. More recent experience has demonstrated that rates of administration can be controlled with greater facility by the use of Sigmamotor infusion pumps.†

The effectiveness of the swivel was singularly dependent upon the bicycle chain. Its flexibility in one plane allowed the dog to stand or lie while its rigidity in all other directions provided an effective handle which rotated the swivel as the dog walked in the cage. Thus the bicycle chain was the link between the swivel and the dog which permitted ambulation without destruction of the intravenous tubing.

Summary. A method for continuous intravenous infusion of dogs confined to metabolic cages has been described. The method was free of mechanical problems and was entirely satisfactory in our experience. It can be recommended for any experiment requiring continuous intravenous infusion of dogs for indefinite periods of time.

† Sigmamotor, Inc., Middleport, N. Y.

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Role of Cations in Conglutination and in Formation of Properdin Zymposan Complex from Bovine Serum. (23432)

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Bovine serum is an inexpensive and convenient source of properdin(1). However, attempts to study the reaction of bovine properdin with zymosan were complicated by the conglutinating activity of bovine serum(1,2), which causes the clumping reaction known as conglutination. The studies reported here describe the methods developed to prevent conglutination.

Materials and methods. Sheep cells were preserved in Alsever's solution(3) containing penicillin and trisodium ethylenediamine tetraacetic acid(4). Standardized suspensions of sensitized sheep erythrocytes (EA), were