sults are different. Instead of assuming a yellow color, the cells are decolorized except the droplets which may remain stained red at least for a short time. The cells, into which NH_4Cl has thus penetrated, are still able to carry out amoeboid movements under favorable conditions. NH_4Cl behaves, therefore, in a manner similar to HCl; in both cases the acid entering the cells causes the granules to give off their stain. On the other hand, the amoebocytes differ in their behavior towards NH_4Cl from certain plant cells, into which, according to the observation of Jacobs, NH_4OH (NH_3) penetrates rapidly while the acid remains in the surrounding medium; thus, in an acid solution the interior of the cell assumes an alkaline reaction owing to the much greater power of penetration on the part of ammonia as compared to HCl or other inorganic acids (or of the H ions). Apparently associated with the delicate structure and lability of the amoebocytes is their great permeability to substances to which many other kinds of cells seem to be impermeable or much less permeable.

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Presence of a growth stimulating substance in the yolk of incubated hens' eggs.

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It has been shown by Burrows[1] that body cells can grow independently only when they are crowded together into narrow stagnant confines. These conditions are important because this growth depends on the accumulation of growth stimulating substance or substances to a certain concentration. This substance or substances has been called the archusia. The cells cannot re-

[1] Burrows, M. T., and Jorstad, L. H., "On the Source of Vitamin A in Nature" and "On the Source of Vitamin B in Nature." To appear in the *Am. J. Physiol.* in May or June, 1926.

tain it as it is soluble in the circulating fluids of the body. It can be extracted from actively growing tissues, and when added in sufficient quantities to the medium of a tissue culture it causes growth in cells not already containing a sufficiency of it for their growth.

Burrows finds that the blastomeres of the chick and frog embryos cannot form under the same conditions sufficient stimulus for their growth when crowded into a culture medium. For them to grow they must obtain an extra supply of stimulus from other sources. This made it seem certain that the egg must either contain a large quantity of stimulus, or such must be liberated early in its development. The failure for these blastomeres to grow under the same conditions as those suitable for the cells of older embryos, is to be related to the presence of yolk in the blastomeres. This inhibiting action of the yolk has been further associated with the presence in the fat and proteins of the yolk of a lipoid substance which has been called the ergusia. The source of the stimulus for the blastomeres has not been determined. The question arises may it not be contained in the yolk, but its action is overshadowed by the lipoids present there. Carrel and Baker[2] have shown that the lipoid of the egg inhibits growth in the tissue culture. It is possible that a stimulus is present in the yolk and that it diffuses more readily than the lipoid and thus becomes active in the embryonic cells independently of these lipoid substances. Previous work on the addition of yolk to a tissue culture has not shown that it has any stimulating value. In these cases the yolk was added to the medium about the cells while in the embryo it must diffuse through limiting membranes to the cells.

I have recently shown[3] that the growth stimulating substances in embryonic tissue extracts are capable of passing through a colloidin dialysing membrane of a texture sufficiently fine to prevent the passage of proteins in quantities large enough to be recognized by the biuret reaction. It became of interest to determine whether the dialysate of the egg yolk may not be rich in the stimulating substance.

The method used for the estimation of growth is essentially the

[2] Carrel, A., and Baker, L. E., *J. Exp. Med.*, 1925, xlii, 143.

[3] Wright, G. Payling, "On the Dialysability of the Growth Stimulating Substances Contained in Extracts of Embryonic Tissues." To appear in the *J. Exp. Med.*

same as that described by me, the yolk taking the place of the embryo tissue extract in the dialysing vessel. The yolk used in the experiments was taken from eggs which had been incubated for 7 or 8 days. The cells upon which it was tested were emigrant cells from heart fragments of 10 to 11 days incubation. The saline used was the same as that described previously. The estimation of growth was based upon the relative numbers of mitotic figures in experimental and control cultures.

The following figures are averages for experiment and control cultures:

	Experiment.	Control.
Expt. 1 average	127	14
Expt. 2 average	114	14

It would appear from this that at the 7th or 8th day of incubation the yolk contains vigorous growth stimulating substances which, when freed from certain of the yolk constituents, are capable of producing great mitotic activity in artificially cultivated heart cells. It may be of interest in this connection to note that in the course of development the yolk-sac entoderm is separated from the yolk proper by the perilecithal space, a locality free from the droplets of fat of which the greater part of the yolk is composed. This supports the possibility that the growth inhibition is associated with the presence of the yolk fat droplets.