A more marked effect was obtained when isolations were made in a continuous human embryo lung cell line—L132[9], obtained from the American Type Culture Collection. These cells were maintained in 2 per cent foetal calf serum in Eagle's medium in roller tubes at 33° C. Nasal washings collected from volunteers who had been inoculated with the B814 organ culture strain induced a maximum cytopathic effect in L132 cell cultures about 5 days after inoculation, but the effect often regressed later. The cytopathic effect can be passed serially: sera obtained from volunteers after recovery from colds induced by the B814 virus were at least four times more effective in inhibiting this cytopathic effect than sera obtained from the same volunteers before inoculation. Of eight volunteers inoculated three developed colds, and the cytopathic agent was isolated from each of them and also from two symptomless volunteers. The cytopathic effect could not be passed if tissue culture fluids were treated with ether or acid. 5-Bromodeoxyuridine (BUDR) at 25 μg/ml. did not inhibit the cytopathic effect in L132 cells, although it reduced the titre of a DNA virus (vaccinia)—by 10^5 TCD_{50}, compared with a control titration. The titre of poliovirus type 1 in these cells was not affected by this concentration of BUDR. Organ culture fluids which had been shown to contain infectious B814 virus also produced the cytopathic effect in L132 cells, indicating that the virus had been propagated in tissue culture.

There is preliminary evidence that three other ether labile viruses, previously cultivated only in organ culture—LP, EVS and MR isolates[10]—have now been propagated in this way. Fig. 1 shows normal L132 cells in a stained roller tube culture and Fig. 2 shows a tube of these cells inoculated with a second tissue culture passage of LP virus. Both tubes were rolled for 5 days at 33° C. The LP and EVS viruses have morphology typical of a "coronavirus" (J. D. Almeida, personal communication). Isolates of B814 and EVS viruses will inhibit plaque production of 229E virus in monolayers of L132 cells in plastic Petri dishes, presumably as a result of some sort of viral interference. This inhibition is reversed by mixing the isolate with convalescent human serum at room temperature before the cells are inoculated. After passage in L132 cells, LP virus has become adapted to diploid lung fibroblast W1-38 cells.

Influenza virus C (strain Jhb/1/66)[11] can also be isolated in the L132 cell line from infectious nasal washings and can be passed serially. Furthermore, the cells support the growth of rhinoviruses (E. J. Stott, personal communication) and are sensitive to respiratory syncytial virus.

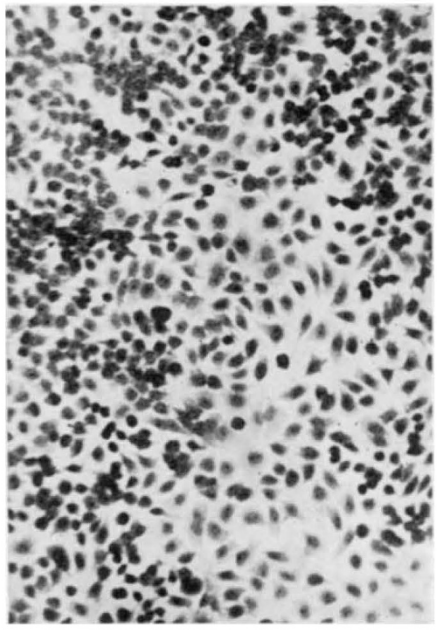

Fig. 1. Normal L132 cells in a stained roller tube culture.

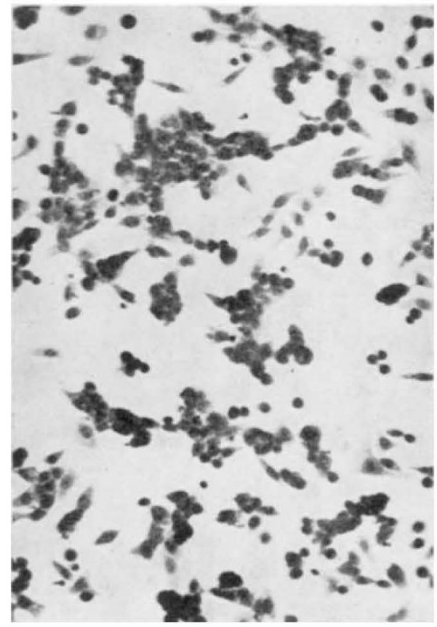

Fig. 2. L132 cells inoculated with a second tissue passage of LP virus.

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Received November 1, 1968.

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Occurrence and Determination of Inositol in the Oviducts of Turkey and Hen

At the junction of uterus and vagina of the turkey a few scattered tubular glands have been observed, Spermatozoa are stored there[2] and retain their fertilizing ability for about 45 days[2]. Turkey semen collected for insemination, however, has to be used within an hour of ejaculation, because fertility decreases considerably after that time.

Weighed segments of parts of the turkey oviduct (isthmus, uterus, vagina and junction of uterus and vagina) were treated in a Bühler homogenizer with water for 1 min. The solutions obtained were deproteinized with alcohol. After centrifugation, they were evaporated and the residue was dissolved in a known quantity of water. Adequate amounts were submitted to electrophoresis at *p*H 1·9 followed by chromatography in a solvent system of butanol, acetic acid and water (4 : 1 : 5). The resulting paper chromatograms were sprayed with

benzidine–sodium metaperiodate reagent, an ammoniacal solution of silver nitrate and 2,3,5-triphenyltetrazolium chloride. We found that only small amounts of glucose were present, but rather large quantities of a non-reducing carbohydrate. We took this to be inositol, because it had the same R_F value and gave the same orange-brown colour with the silver nitrate reagent as pure inositol.

To confirm this, inositol, glucose, fructose and sorbitol were treated with tri-sil, and then the trimethylsilyl derivatives were investigated by gas chromatography (F and M 402, glass column, liquid phase 2·5 per cent *SE*-30, column temperature 180° C, detector temperature 190° C, flash heater temperature 230° C). Fig. 1 shows that the carbohydrates separated well. We then eluted the carbohydrate from the paper chromatogram with water, and the solution was evaporated under nitrogen in a small vial. The residue was treated with tri-sil and the derivative formed gave a peak with the same retention time as the tri-sil derivative of pure inositol. It is clear therefore that inositol is present in the oviduct of the turkey hen.

Samples of a known solution of inositol were submitted to chromatography. After elution (using Whatman 3MM paper and trimethylsilylation) some of each sample of inositol was injected into the gas chromatograph. The amounts injected were plotted against the areas of the peaks and gave a straight line (Fig. 2).

Table 1. CONTENT OF INOSITOL IN VARIOUS SEGMENTS OF TURKEY AND HEN OVIDUCT (MG/100 G TISSUE)

	Turkeys				Hen
	1 day insem. egg	3 days insem. no egg	16 days insem. egg	30 days insem. egg	Insem. egg
Vagina	5·9	9·0	25	3·9	5·5
Junction uterus–vagina	6·9	10·0	25	14·0	25
Uterus	11·0	22·5	7·5	11·0	10·5
Isthmus	4·0	7·9	17·0	3·9	0·35
Magnum					0·35
Infundibulum					25

A few results obtained with the same procedure using extracts of turkey oviduct are given in Table 1. It is sometimes possible to obtain small amounts of liquid from the uterine lumen—it also contains inositol, and on one occasion, as much as 24 mg per cent was recorded. Similar experiments have been carried out using the hen oviduct. Inositol seems to occur in all parts of the oviduct. One set of values is given in Table 1. Preliminary experiments have also shown that inositol occurs in the uterus of the dog and cow. Experiments are in progress to determine the parts of the cells in which inositol is formed.

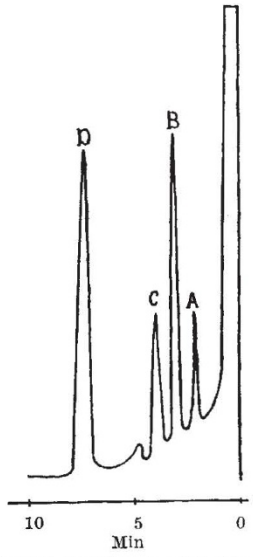

Fig. 1. Gas–liquid chromatographic separation of a mixture of trimethylsilyl derivatives of pure fructose (*A*), glucose (*B*), sorbitol (*C*) and inositol (*D*).

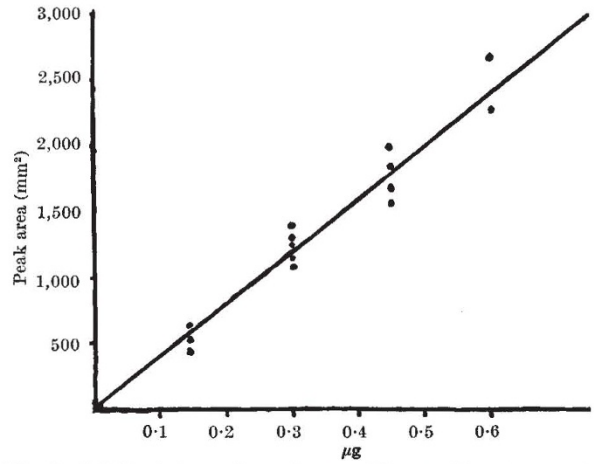

Fig. 2. Relation between the peak areas of the gas chromatograms in mm² and the quantities of inositol injected (μg).

Several investigators have mentioned the significance of inositol for the storage of cells[3–5] and we wondered whether this compound has a similar function in relation to spermatozoa in the oviducts of turkeys and hens.

Examination of turkey and hen spermatozoa and seminal plasma showed that inositol was present in both the plasma as well as in the spermatozoa. This suggests that the presence of inositol influences the motility and perhaps the fertilizing ability of the spermatozoa. Comparable experiments have therefore been carried out with turkey semen diluted with Tyrode solution with and without inositol (2,000 mg/l.). In all cases the motility of the spermatozoa in the semen and inositol decreased many hours after that of the spermatozoa without inositol. The influence of inositol on fertilizing ability is being investigated. The presence of inositol is evidently beneficial to the storing qualities of turkey semen.

We thank Mr H. J. Beudeker for assistance with the inositol determinations.

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Received September 11, 1968.

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Effect of Protamine Sulphate and Environmental Temperature on Mouse Sarcoma 180

IT has been shown that protamine and its derivatives inhibit the growth of Landschutz ascites tumours and of sarcoma 180 in mice[1], and that they can retard the growth of malignant tumours in man[2,3]. Protamine probably does this by inhibiting a tissue thromboplastin which is produced by malignant cells and which aids invasiveness[4]. Metabolic and pharmacological processes are influenced by changes of temperature, and so it seemed worthwhile