agents after the development of the collapse had no beneficial effect.

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Effect of FUDR on Interferon Production. (30032)

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Two recent papers present conflicting data concerning the need for new DNA synthesis for the production of interferon. Holmes *et al*(1) using iododeoxyuridine (IUDR) as an inhibitor of DNA synthesis, indicated that when DNA synthesis was stopped, interferon production was also blocked. On the other hand, Donikian and Stewart(2) found that IUDR increased the amount of interferon produced by chick embryo (CE) cells infected with irradiated influenza virus under certain circumstances, or had no effect under others. As part of a larger study on the control of interferon production, we have studied the effect of fluorodeoxyuridine (FUDR) on interferon production, using both an RNA virus and a DNA virus as stimulating agents, and these studies are reported here. Both FUDR and IUDR block the synthesis of DNA by inhibiting the conversion of deoxyuridylic acid to deoxythymidylic acid.

Experimental. Chickungunya virus was assayed by plaque method on primary hamster embryo cells, as previously described(3). Interferon was assayed by the plaque reduction method on CE cells using vesicular stomatitis virus as challenge. The titer of interferon is expressed as the reciprocal of the dilution of interferon that caused a reduction of 50% in number of plaques produced. Vaccinia virus was assayed by the fluid plaque method of Postlethwaite(4), using HeLa cells.

FUDR at concentrations of 3×10^{-6} to 1.8×10^{-5} molar inhibited DNA synthesis in CE cells by 90-95%, as measured by the incorporation of H^3-uridine into the DNA of the cells. Table I demonstrates the lack of effect of these concentrations of FUDR on

TABLE I. Effect of FUDR on Interferon Production by CE Cells Infected with Chickungunya Virus.

FUDR, molarity	Interferon titer		Virus titer $\log_{10}$ pfu/ml	
	No FUDR	With FUDR	No FUDR	With FUDR
3×10^{-6}	300	200	3.2	3.2
1×10^{-6}	100	100	3.4	4.0
3×10^{-5}	100	100	Not done	

FUDR, at indicated final concentrations, was added to the cells at the same time as the virus. Virus:cell multiplicity = 10. Virus was absorbed 30 min at room temperature, and cells were then washed 4 × with BME before incubation for 18 hr at 37°C. Interferon titers were determined on the fluid and virus on the frozen and thawed cells plus fluid.

TABLE II. Effect of FUDR on Growth of Vaccinia Virus in CE Cells and Production of Interferon by Such Cells.

FUDR	Interferon titer	Final titer of virus $\log_{10}$ pfu/ml
0	5	6.9
1.8×10^{-5}	10	5.5
1.2×10^{-5}	10	5.5
6×10^{-6}	8	5.8

Virus was added at a multiplicity of 8, at same time as FUDR. After 2 hr absorption at 37°, the fluid was removed, the cells were washed 3 times with BME, and new fluid with or without FUDR was added. After 24 hr at 37°, the whole culture was frozen and thawed 3 times and clarified by low speed centrifugation. Virus and interferon contents were determined in the supernatant fluid.

the production of interferon by these cells when infected with chickungunya virus, an RNA virus. Our strain of this virus grows little or not at all in CE cells, but gives maximum titers of interferon within 5-6 hours after infection.

Table II shows the effect of FUDR on the growth of vaccinia virus, a DNA virus, in CE cells and the production of interferon by these cells when infected with vaccinia virus.

It will be seen that vaccinia growth was inhibited about 95%, but that there was no inhibition of interferon production. The small differences in interferon titer seen in Table I are within experimental error.

Discussion and summary. FUDR had no effect on the production of interferon by CE cells, when the inducing virus was chickungunya virus, an RNA virus, or vaccinia virus, a DNA virus. The concentrations of FUDR used inhibited total DNA synthesis and growth of vaccinia virus by about 95%. Thus, under our experimental conditions, it may reasonably be concluded that little or no new DNA synthesis is needed for the production of interferon. Holmes *et al*(1), working with mumps virus, report that iododeoxyuridine inhibits the production of interferon without any gross toxicity to the cells. They conclude that new DNA synthesis is needed for interferon production. It is difficult to reconcile their findings with ours, unless the rather high levels of IUDR they used exerted a deleterious action on the metabolism of the cells which was not revealed by morphological alterations. The data of Donikian and Stewart(2) do not suggest the need for new DNA synthesis for the production of interferon. It would appear, therefore, that DNA is functioning in its usual way as a template in the interferon producing cell.

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Effects of Endotoxins on Cultured Leukocytes. (30033)

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In vivo administration of endotoxin produces multiple hematologic, immunologic, and physiologic effects(1). It can result in an immediate granulocytopenia, followed by lymphopenia and then a transient granulocytosis. Repeated administration results in