

## PROPAGATION OF GOAT POX VIRUS ON MONOLAYER CELL CULTURES ( \* )

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*With 4 figures*

Although the majority of different members of poxviruses have been carefully studied, little is known about goat pox and no detailed report on the disease and its causative agent is, so far, available.

The cultivation of *goat pox virus* (GPV) in cell cultures reported in the present paper makes available the most reliable and economic source for large scale production of vaccine to control the disease.

### Materials and Methods

Virus: GPV used in this experiment was lymph and lyophilized pulp, Gorgan strain, isolated in Iran in 1956 and kept at  $-65^{\circ}$  C after two in vivo passages until used.

The strain is highly virulent and antigenic. Subcutaneous inoculation of  $\frac{1}{2}$  ml. of the virus adsorbed on aluminium gel (1/200 proportion) establishes a solid immunity in indigenous goats and causes generalization and a high mortality in susceptible imported animals.

Tissue cultures: Primary sheep kidney (SK), kid kidney (KK) and kid testis (KT) cells have been used throughout the experiment. Trypsinized cells were suspended in Earle's medium containing 10% unheated fresh calf serum, 10% lactalbumin hydrolysate (5% sol.), 2% phenol red (10% sol.), and penicillin and streptomycin 100,000 I.U. and 100 mgr. respectively.

Cultures were kept at  $37^{\circ}$  C. Complete monolayer was formed on the 3rd—5th day.

Cultivation of virus: 100% dense cultures in medicine bottles were infected with 2 ml. of infectious material diluted  $10^{-1}$  and kept at  $37^{\circ}$  C for 3 hours. Bottles were tilted from time to time to allow the inoculum to reach all cells and avoid the monolayer drying.

After the adsorption period the inoculum was discarded, cells were washed gently with phosphate buffered saline and 12 ml. of VM 3 (5) were introduced into each bottle. Infected cultures were transferred to a  $37^{\circ}$  C incubator, and checked daily together with non-infected controls and the variations recorded.

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(\*) Zbl. Vet. Med. B. 13, (1966), 334.

A weak cytopathogenic effect (CPE) appeared 10 days after infection with the original material. This period diminished to 72 to 48 hours when serial passages were continued (Figs. 1 — 3).

### Results



Fig. 1. Normal Kid Kidney Cell's (Control)

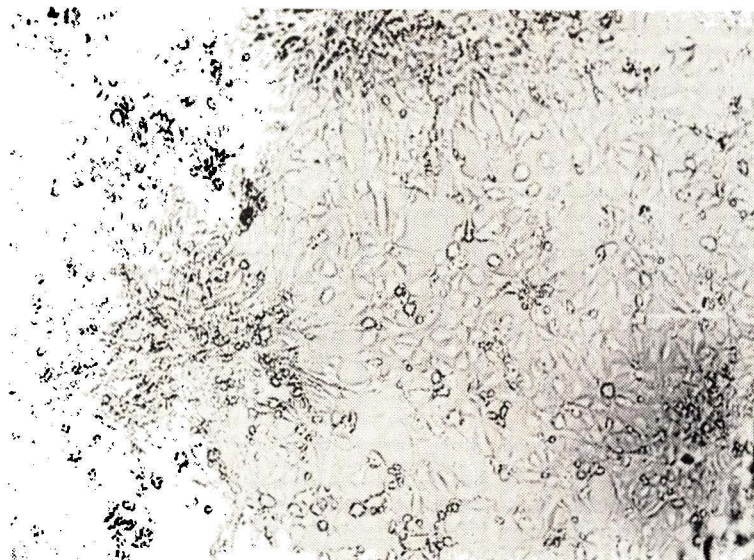


Fig. 2. CPE after 48 hours



Fig. 3. CPE after 120 hours

Titration of virus: Ten-fold dilutions of the 5th KK and KT passages were separately prepared in VM 3 (virus medium 3) and inoculated in 0.2 ml. amount in each of 6 culture tubes. Infected cells were examined daily for CPE. The infectivity titer calculated according to KARBEN reached  $10^{5.2}/\text{ml}$ .

The highest titer of virus was obtained at 96 to 120 hours when the cells showed 80—100% CPE. Then the titer fell gradually so that 216 hours incubation of virus caused no degeneration in infected cells (Fig. 4).

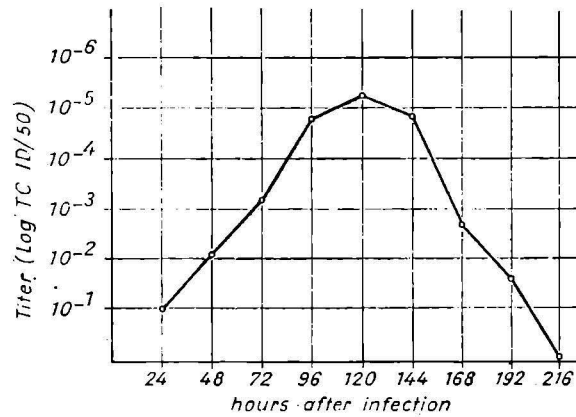


Fig. 4. Growth curve of Goat Pox Virus in Kid Kidney cells

### Discussion

Goat pox is one of the major contagious diseases of the goat in Asian and some African countries.

In nature, only goats are infected. Sheep and other animals living with infected goats never contract the disease (1 and 4).

The possibility of an antigenic relationship between goat and sheep pox viruses has been debated in the past (2 and 3) and requires further investigation.

Mass vaccination is the only means of controlling the disease. The application of formolized lymph or tissue vaccines has been of much value but live-modified strains of virus would probably be the most effective prophylactic to control the disease.

#### **Acknowledgement**

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#### **Summary**

An Iranian strain of *goat pox virus* has been successfully adapted to sheep kidney, kid kidney and kid testis cell cultures. Infected cells were completely degenerated at 120 hours. The highest titer of virus was obtained between 96 and 120 hours.

Multiplication of virus in tissue culture produces a large supply of material for further investigation as well as for vaccine production.

#### **Zusammenfassung**

##### **Die Züchtung von Ziegenpockenvirus auf einschichtigen Zellkulturen.**

Ein Stamm Ziegenpockenvirus aus dem Iran konnte erfolgreich an Schafnieren-, Ziegenlammernieren- und Ziegenlammernierenzellkulturen adaptiert werden. Die infizierten Zellen waren in 120 Stunden vollkommen degeneriert. Der höchste Virustiter wurde zwischen 96 und 120 Stunden beobachtet. Die Vermehrung des Virus in Gevebekulturen liefert ein grosses Angebot an Material für weitere Untersuchungen sowie auch für die Herstellung von Vaccine.

#### **Résumé**

##### **La multiplication du virus de la variole de la chèvre sur des cultures de cellules à couche unique**

Une souche de virus de la variole caprine d'Iran put être adaptée avec succès à des cultures de cellules rénales de mouton, de cellules rénales et de cellules testiculaires caprine. Les cellules infectées présentaient en 120 heures une dégénérescence complète. On observa le titre de virus le plus élevé entre 96 et 120 heures. La multiplication du virus en culture de tissu donne une grande quantité de matériel utilisable pour d'autres recherches ainsi que pour la préparation de vaccins.

## Resumen

### El cultivo del virus de la viruela caprina en culturas celulares monoestratificadas

Una cepa de virus variólico caprino del Irán se pudo adaptar con éxito en culturas celulares de rinones ovinos, de rinones de cabritos y de testículos de cabritos. Las células infectadas estaban degeneradas por completo al cabo de 120 horas. El título vírico máximo se observó entre las 96 y 120 horas. La multiplicación del virus en cultivos hísticos brinda una gran oferta de material para subsiguientes investigaciones, así como también para la elaboración de una vacuna.

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