meters of isolated mitochondria and also of intact infected cells decayed beginning from 10th h p.i. At this time infected cells exhibited only slight morphological differences to controls in light microscopy. Since mitochondria play important roles both in energy supply and also in regulation of cellular ionic environment progressive functional impairment may contribute to cytopathic damage due to viruses.

Studies on Semliki-Forest-RNA Containing Polysomes

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The genomic RNA of Semliki-Forest-Virus (SFV) sediments at 42S. In the infected cell several viral RNAs can be found: a 42S-, a 26S- and a double stranded 20S RNA among others. Polysomes were extracted from SFV infected chick embryo fibroblast cultures with a hypotonic medium containing Triton-X-100 and cycloheximide to prevent ribosomal run-off; they were then isolated by centrifuging through layers with high sucrose concentrations. Such polysomes are highly active in cell-free systems with sap from uninfected chicken embryos. The different parameters of the cell-free system have been studied. If the viral RNA has been labelled in vivo in the presence of actinomycin to inhibit labelling of host RNA, all individual polysomal peaks contain label, though the main RNA is always the 26S-RNA. Therefore, as opposed to polysomes from uninfected cells, different spacings between ribosomes on this single type of RNA have to be assumed. The inhibition of over-all protein synthesis in infected cells in vivo cannot be explained by a polysomal defect: equal concentrations of polysomes in uninfected and in infected cells (irrespective of time after infection up to 14 h) yield in vitro the same amount of incorporation. A better explanation for inhibition of protein synthesis in vivo is given by the observation of an increasing amount of single ribosomes and a decreasing amount of polysomes in extracts dependent on duration of infection. Under condition of EDTA-desintegration of polysomes to ribosomal subunits in vitro, the 26S polysomal RNA sediments with the 40S ribosomal subunit.

Some Ultrastructural Aspects of the Replication of Coxsackievirus A9 in CV-1 Monkey Kidney Cells

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A lot of biochemical data concerning the replication of Enteroviruses are available but only little is known about the morphological structures involved in the synthesis of the virus progeny. The presented study deals with the ultrastructural changes in monkey kidney cells (CV-1) infected with Coxsackievirus A9. Giant polysomes containing about 35 ribosomes appear at 1 h p.i. in infected cells. According to biochemical data (SUMMERS et al., Virology *31*, 427–435, 1967; JACOBSON et al., Proc. natn. Acad. Sci. USA *61*, 77–84, 1968) they probably contain the transcript of the whole genetic information of the virus. After 4 h p.i. considerable alterations in the ultrastructure of the nucleus become obvious. At the same time areas of membrane bound vesicles arize in the cytoplasm in place of the Golgi zones. In parallel with the appearance of the vesicles the polysomes disintegrate. Different functions were attributed to the vesicles, e.g. they were thought to be the site of virus RNA synthesis (CALIGUIRI et al., Virology *42*, 100–111, 1970). The fact however that they appear after the extensive formation of the giant polysomes suggests that they are at least not the only site of viral RNA synthesis. Titration experiments show a sharp rise in infectious virus after 5 h and 7 h p.i. for intracellular and for extracellular virus respectively. Various granular and fibrillar components in the cytoplasm are though to consist of virus material but no virus progeny is detectable in their neighbourhood. All of the newly synthesized viruses are found in cell protrusions. Not until late in the replication cycle groups of virus particles are located in the inner part of the cell too. Most of the viruses are arranged in parallel linear arrays which are separated from each other by thin fibrils. Virus cristals were never observed.

Anucleate Cells: their Preparation and their Reaction to Poliovirus Infection

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Virus-host cell interactions were studied in poliovirus-infected, enucleated HEp-2 cells. Cytochalasin B causes spontaneous enucleation of mammalian cells at low percentages, which can be considerably enhanced by ultracentrifugation of whole monolayers. Some cell lines (like HEp-2 cells), however, do not adhere firmly enough to a carrier to be centrifuged. Therefore, a new method for mass enucleation was developed: S-shaped density gradients, containing colloidal silica, were adjusted to have their shallow part between the density of the cytoplasm and that of the nuclei. More than 70% of HEp-2 cells could be enucleated by ultracentrifugation in such gradients, containing cytochalasin B. Purification up to 97% was carried out by centrifugation at low speed through a second, preformed gradient. Purity of the anucleates was tested in Giemsa-stained smears as well as by ^{3}H-thymidine and ^{3}H-uridine uptake. The viability of the enucleated cells, tested by ^{3}H-leucine incorporation was comparable to that of normal cells. For further details see BOSSART et al., Expl. Cell Res. *96*, 360–366 (1975). In enucleated cells, the amount and kinetics of virus-induced RNA synthesis was measured by ^{3}H-uridine incorporation, the number of RNA synthesizing cells being determined by autoradiography. In contrast to ^{3}H-uridine uptake, the ^{3}H-leucine incorporation showed no virus-induced peak. Accordingly, virus yield was low and no virus-induced redistribution of lysosomal enzymes occurred. The available data indicate, that protein synthesis is the limiting factor of virus growth in the polio-HEp-2 system.

Comparative Study of the Hemolysin Produced by Different Serotypes of *Clostridium tetani*

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The hemolysins produced by 9 toxigenic (Tulloch Types I to IX) and 2 non-toxigenic (types III and V) strains of *Clostridium tetani* have been partially characterized. The supernatant of a 24 h culture (Brain Heart Infusion