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Viral particles were detected in 30.1% of examined specimens. In 22% of cases Rotavirus were described\(^3\), and in one case, tubular forms were evidenced\(^4\). In 3.5% of cases Adenovirus were observed, in two cases surrounded by adenovirus-associated virus particles\(^5\).

Parvovirus\(^6\) 25-30 nm particles were observed in 2.5% of patients. Three cases of Parvovirus-Rotavirus associations could be described too. In one specimen large clusters of 15-20 nm particles which can probably be classified as Parvovirus\(^7\) were evidenced.

In 0.16% of cases typical Astrovirus could be described\(^8\). Well defined Coronavirus could not be detected, as demonstrated by immunoelectronmicroscopy techniques with specific rabbit induced antibodies. In some specimens (1%), however, there could be observed varying undefined particles with particular surface projections, similar to the Mixovirus ones\(^9\).

CYTOCHEMICAL AND AUTORADIOGRAPHIC OBSERVATIONS ON TOMATO BUSHY STUNT VIRUS-INDUCED MULTIVESICULAR BODIES

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Various hypotheses have been put forward on the nature and function of the multivesicular bodies (MVBs) specifically induced by tomato bushy stunt virus (TBSV) and other tombusviruses (diameter c. 30 nm, single-stranded RNA genome)\(^1\)-\(^3\). The MVBs are roundish cytoplasmic inclusions, made of one or more layers of vesicles surrounding an internal cavity variously subdivided and often containing fine fibrils. In the attempt to elucidate the nature of these structures and their possible role in virus replication we made cytochemical reactions and autoradiographic experiments on leaves of Gomphrena globosa systematically infected with TBSV.

**Cytochemical reactions:**
- a) specific staining of the plasma membrane with phosphotungstic acid-chromic acid\(^4\);
- b) specific staining of the chloroplast membranes with bismuth\(^5\);
- c) digestion with pronase after H\(_2\)O\(_2\) treatment\(^6\).

**Autoradiography:** the method of Caro and Van Tubergen\(^7\) was used after administration of H\(^3\)-uridine to the infected leaves.

The first results of our observations indicate that the membranes of MVBs are not directly originated from the plasma membrane or from the chloroplasts and that protein is not their main component.

1. Martelli and Russo, Adv. Virus. Res. 21 (1977) 175.
2. Appiano et al., J. Gen. Virol. 40 (1978) 277.
3. Russo et al., J. Ultrastruct. Res. 82 (1983) 52.
4. Roland et al., Stain Technol. 47 (1972) 195.
5. Hayat, in: Principles and Techniques of Electron Microscopy I (1970) 301.
6. Monneron, J. Microscopie 5 (1966) 67a.
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CYTOCHEMICAL IDENTIFICATION OF CALLOSE DEPOSITS AROUND PLASMODESMATA OF VIRUS-INFECTED PHLOEM CELLS

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In previous studies on the ultrastructural alterations caused by the RuPV strain of barley yellow dwarf virus (BYDV) on the host plants one of the noteworthy cytopathological features observed has been the presence of electron-transparent deposits around plasmodesmata of the infected cells. These deposits have been considered callose-like material because of their low electron density, general appearance and location between plasmalemma and cell wall, although they sometimes were more electron-dense than the callose present on the nearby sieve plates.

In order to identify this material some chemical treatments have been carried out directly on thin sections
using either KOH or SnCl₂ solution which are known as callose solvents in unembedded plant tissues (Currier, Am. J. Bot. 44, 1957, 478). Sections (60-80 nm thick), cut from either Epon-Araldite or Spurr's resin embedded BYDV-infected plants, were transferred with a Deer's loop (J. Microscopy 130, 1983, 115) to small drops of KOH at different concentrations or to SnCl₂ solution in HCl IN for 5 to 120 minutes. They were then picked up with a grid, immediately washed in distilled water, stained with U and Pb and examined under the electron microscope.

Results indicate that KOH 1 N treatment for at least 15 minutes removes the deposits from plasmodesmata, both in Epon-Araldite and Spurr's embedded samples, although the high pH of the solution tends to make sections opaque, diminishing their general contrast. However, lower KOH concentrations are not effective also after longer exposure periods. SnCl₂ treatments do not give reproducible results as they sometimes seem to be effective in removing the deposits and at other times they are not. In any case, when deposits are removed from plasmodesmata, either using KOH or SnCl₂, also callose on sieve plates is removed thus confirming the identity between the two substances.

**X-RAY MICROANALYSIS AND ULTRASTRUCTURE OF MAIZE LEAVES NATURALLY INFECTED WITH BARLEY YELLOW DWARF VIRUS**

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Barley Yellow Dwarf Virus (BYDW), an isometric virus of graminaceous plants, is a typical member of the luteovirus group, in being phloem-restricted, and aphid-borne but non-sap-transmissible. BYDW has been proven to be a cosmopolitan pathogen existing in several strains that vary in insect vector specificity.

Three cultivars of maize plants (W646, FR18 and B84) resulted positive after the serological test by enzyme-linked immunosorbent assay (ELISA), a useful direct quantitative assay for monitoring yields of BYDW, were used in our studies.

General symptoms, such as yellowing, redding and malformations of the leaves were observed. The ultrastructural observations on the infected tissues gave us the possibility to identify the virus particles in the phloem cells and to analyze the alterations of cell organelles caused by the virus infection.

Moreover, we report the data obtained by SEM X-ray microanalysis, performed on healthy and infected areas following standard and new methods of specimen preparation.

Particularly interesting are the low levels of K⁺ detected in reddish portions of infected leaves. The results of X-ray microanalysis are discussed with the ones obtained after TEM observations.

**POLYEDRAL VIRUSLIKE PARTICLES IN MYOCYTES AND NEUROGLIA OF PERIPHERAL NERVES IN OCTOPUS VULGARIS L. AFFECTED BY RUNGER'S DISEASE**

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The present work contributes knowledge to the area of viral tumors in Invertebrates. Rungger described a seasonal epidemic between young Octopus Vulgaris L. found at summer time in the Gulf of Naples when anthropic industrial and oil marine pollutions were at their peak, and treatment was not yet initiated. The morb consists of edema and nodular tumors in the tentacles and mantle and leads to apathy of the animal and self-multilation and appears to be fatal. Tumoral foci appeared to consist histologically of degenerated myocytes and connective tissue and to contain hexagonal particles measuring 140 x 100 nm approximately.

According to Rungger's data, 8.4% of young animals (about 100 g in body weight) were affected but the efforts to obtain experimental reinfection were not conclusive.

In the course of the present study, realized in 1973, three animals were found affected by mantellar microfoci alone and not yet by tentacular tumors; the microfoci were all localized on or nearby the nerves irradiating from the stellate ganglion on the ventral side of the mantle. Within the nodules myocytes appeared necrotic and virus-like