PLEOMORPHIC VIRUS-LIKE PARTICLES IN HUMAN FECES

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Summary
Pleomorphic fringed particles bearing some resemblance to orthomyxoviruses and coronaviruses were seen in 90% of stools from south Indian children and adults but not in stools from neonates. This finding may be related to the abnormalities of intestinal structure and function common in this region of India.

INTRODUCTION
Detection by electron microscopy of viruses, morphologically similar to reoviruses, in the stools from a large proportion of infants and children with non-bacterial gastroenteritis has been reported in several countries. Evidence suggests that these viruses are an important cause of gastroenteritis. Flewett et al. reported finding various isometric viral particles in the stools of children and adults with and without diarrhoea. Using similar techniques to study stool specimens in India, we found that many contained previously undescribed ultramicroscopic pleomorphic virus-like particles.

METHODS
Freshly passed feces was transported to the laboratory on ice and stored at -70°C till examination. An approximately 20% suspension (w/v) of stool in water was made using a mechanical blender. This suspension was centrifuged at 10,000 g for thirty minutes at 4°C. The supernatant was passed through a filter with a pore size of

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1200 rpm. 5 ml. of the filtrate was centrifuged at 25,000 g for ninety minutes. The supernatant was discarded and the pellet was resuspended in one or two drops of distilled water, placed on coated grids, negatively stained with ammonium molybdate, and examined with a Philips EM 200 electron microscope.

Stools were obtained from 6 neonates within the first week of life, 12 normal children, 29 adult controls, and 16 adults with chronic tropical sprue.

Planomorphy virus-like particles in human faeces.
(a) Rounded particle with the most commonly observed type of fringe composed of one layer of rounded knobs. (b) Kidney-shaped particle with double-layered fringe with a T-shaped projection attached peripherally. (c) Particle with double-layered fringe with a second knob-shaped projection attached peripherally. (d) Large, irregular, elongated particle with fringe apparently identical with that of particle (b).
RESULTS

Particles were not found in the stools from 6 neonates. However, pleomorphic fringed particles were found in stools from 10 of the 12 children, in stools from 27 of the 29 adult controls, and in stools from 14 of the 16 patients with tropical sprue. The particles varied in shape. Round (fig. a), oval or kidney shaped (see fig. b and c), and bizarre elongated forms (see fig. d) were seen. Particles of widely differing sizes and shapes were present in the same specimen. They ranged in size from about 100 nm for the smallest rounded particles up to about 400 nm for some of the larger ones. The elongated forms were about 100 nm in width and up to 800 nm long.

The fringes on the particles showed various morphological patterns. The most common pattern was made up of round or oval shaped knobs attached to the main body with a thin stalk (fig. a). In some T-shaped projections were attached peripherally to the knobs (see fig. b), while some others had a spike or a second small spherical structure similarly arranged (fig. c). The total width of the fringe in different particles ranged from 20 to 46 nm.

In the same stool specimen particles with some or all of these types of fringe could often be seen. No special type of particle was associated with the patients with tropical sprue. Attempts at growing these particles in tissue-culture and human fetal intestinal organ culture have not so far been successful.

DISCUSSION

The exact nature of these particles is not yet known. They show some morphological similarities to the orthomyxoviruses and some to the coronaviruses. However, the fringe morphology is not identical with electron micrographs of either of these groups of viruses. The possibility of their being mycoplasmas cannot at present be excluded, but on morphological grounds it seems improbable.

These submicroscopic particles are common in the community and are acquired after birth. Since they were found in nearly all stool specimens they are presumably being excreted most of the time by all the subjects examined. They do not seem to be associated with any specific disease state, since they were found in healthy children and adults and in
patients with chronic tropical sprue. Flewett et al., in their studies in the U.K., did not find particles like this either in patients with gastroenteritis or in controls. The absence of these particles in neonates and their widespread distribution in children and adults reflects the frequency of intestinal morphological abnormalities and malabsorption in the symptom-free population of southern India.1 This raises the question whether these particles are in any way causally related to these intestinal abnormalities. Further studies are in progress.

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