Rotavirus Particles Can Survive Storage in Ambient Tropical Temperatures for More than 2 Months

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Typing and in vitro cultivation of rotavirus-positive human stool samples stored unintentionally at ambient tropical temperatures for 2 1/2 months showed that rotavirus is stable and may still be infectious in vitro. This indicates that stool specimen collection for rotavirus studies can be performed in areas and settings where reliable cold storage is not available. The retained infectivity of rotavirus particles underscores the need for safe systems for disposal of feces, in particular in developing countries where rotavirus is a major cause of childhood mortality.

Rotavirus is the most common agent causing acute dehydrating diarrhea in infants and children worldwide (5). Children living in developing countries constitute the major part of the approximately 450,000 deaths estimated to occur each year due to rotavirus infections (6). In developing countries, rotavirus diagnosis is primarily based on clinical manifestations. Nevertheless, as prevention through vaccination has become a realistic prospect within the near future (1), rotavirus disease and strain surveillance has intensified worldwide during the past decade(s).

It is recommended that feces containing rotavirus be stored at −20°C for later genotyping analysis (2) or cultivation (8). During a study of rotavirus diarrhea in Guinea-Bissau, West Africa (3), all laboratory-based research was disrupted for several months due to an armed conflict in June 1998. Already characterized rotavirus-positive stool samples, which were kept in cryotubes (Nunc A/S, Roskilde, Denmark) were left at ambient temperatures (30 to 35°C) for 2 1/2 months, as electrical power was cut shortly after the initiation of the conflict.

When the conflict ceased in August 1998, the 167 rotavirus-positive stool samples (3) were refrozen at −20°C before transportation to Copenhagen, Denmark, where they were stored at −20°C. Prior to the war, RNA from these rotavirus-positive samples was extracted and genotyped with a one-step multiplex reverse transcriptase PCR technique, as described previously (4). In Copenhagen, we extracted RNA and genotyped rotavirus from 15 randomly selected specimens that contained rotavirus of determinable VP4 and VP7 genotypes. There was a complete agreement with regard to the VP4 and VP7 genotypes of all the selected specimens. In addition, 11 specimens that initially had been difficult to type in Bissau, likely due to a small amount of viral particles per load or RNase in the stool, were cultivated in the MA104 cell line (8). Five of 11 (45%) were successfully cultivated, which is about the average success rate for cultivation of properly stored rotavirus-positive fecal samples (9).

We showed that rotavirus particles stored in feces are stable and may be infectious in vitro after 2 1/2 months of storage at ambient temperatures above 30°C. In a study of the stability of porcine rotavirus in feces (7), it was observed that rotavirus infectivity was maintained for a long period of time (more than 32 months) in stool specimens stored at a low temperature, i.e., 10°C. To our knowledge, no other study describing the survival of human rotavirus in stool at ambient temperatures has been published, and we hope that this unintended and unplanned observation is useful to other scientists working with rotavirus surveillance and genotypic characterization under conditions in which the power supply is unreliable.

While our observation is an encouragement for field epidemiologists studying rotavirus in settings with a weak infrastructure, it is not a recommendation to delay transport of specimens. Although we did not demonstrate clinical infections from rotavirus particles stored for months, the retained in vitro infectivity underscores the need for adequate and safe systems for disposal of feces in developing countries (7).

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