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Occurrence of viruses in human stools in the Ahaggar (Algeria)

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SUMMARY

From October 1977 to May 1980, 243 stools collected in sedentary and semi-nomadic populations of the Ahaggar (Algerian Sahara) were examined using immunoelectronmicroscopy and tissue culture inoculation. Immunoelectron-microscopy revealed the presence of rotaviruses in 8, coronaviruses in 26, adenoviruses in 5 and small round viruses in 4. Enteroviruses were isolated in tissue culture from 24 stools.

Rotaviruses were present in the Ahaggar but were associated with little acute enteric disease. The high frequency of coronaviruses both in gastroenteritis patients and in patients without disease was surprising. The prevalence of enteroviruses in this hyperarid zone was similar to or higher than that found in noticeably more humid countries.

Further systematic bacterial, viral and parasitic examinations are required to clarify the role of the above viruses in the aetiology of gastroenteritis in this region.

For more than ten years, research on human biology has been carried out in the Algerian Sahara. By 1969 we showed that enteroviruses do spread in this arid zone of Central Sahara. It seemed interesting to follow up this study in the Ahaggar, systematically studying the faecal viral carriage and the role played by certain viruses such as rotavirus in the aetiology of acute gastroenteritis.

We studied 243 stool samples collected from different places in the Ahaggar: in October 1977, mainly around Tamanrasset from 84 children of whom 50 were of fixed abode and 34 in families of semi-nomadic shepherds of whom the majority had come from Mali to the southern frontier of Algeria (Timiaouine–Tinzaouaten) to the area around Tamanrasset; in April 1978, around Tahifet and Tazrouk, from

Table 1. *Detection of viruses in faecal specimens according to the time of collection*

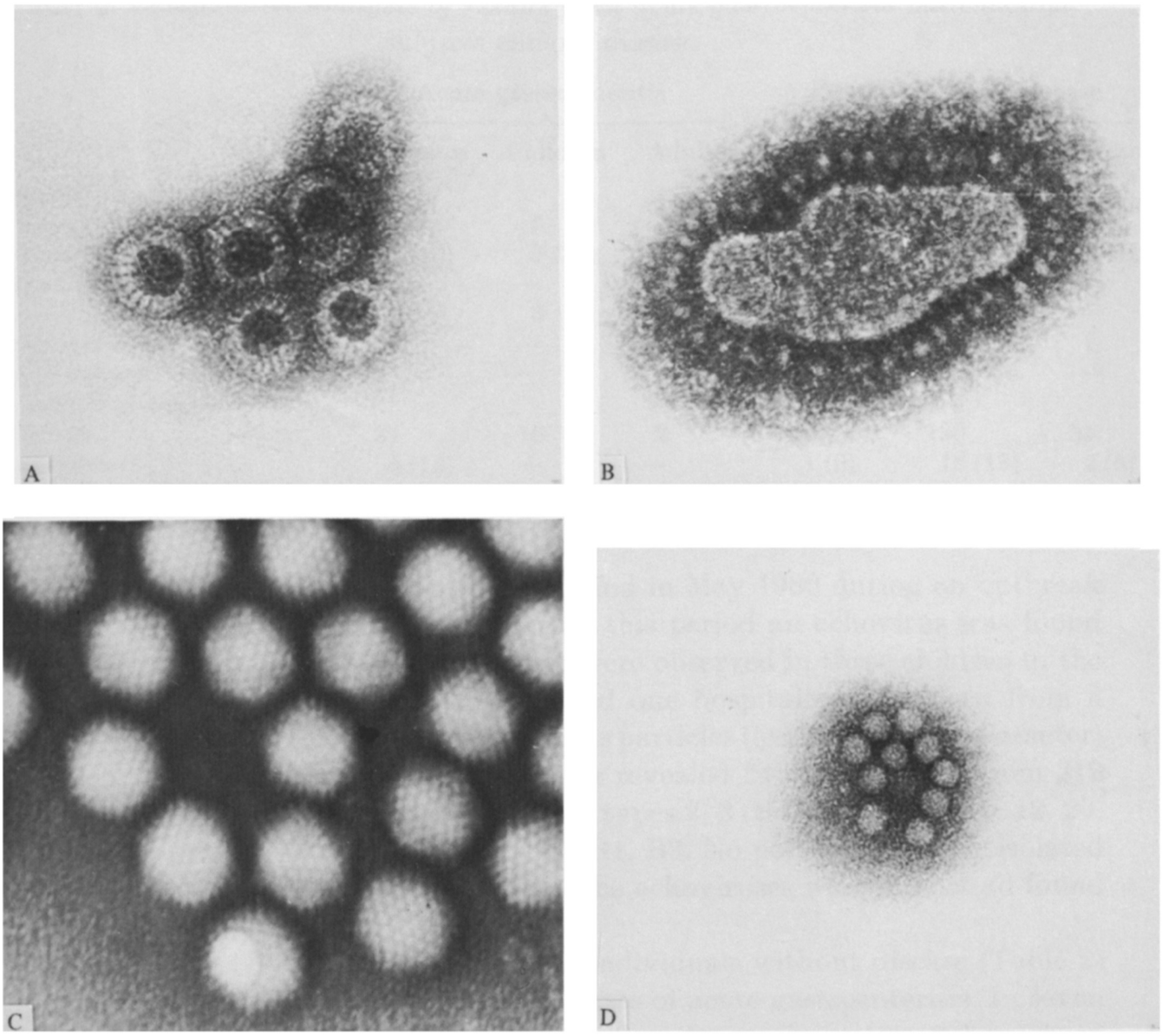
	Oct. 1977	Apr. 1978	Oct. 1978	May 1980	Total
Immunoelectronmicroscopy					
No. tested	65	13	66	21	165
No. positive (%)	13 (20)	3 (23)	14 (21)	13 (62)	43 (26)
Rotaviruses	7	—	1	—	8
Coronaviruses	6	2	5	13	26
Adenoviruses	—	—	5	—	5
Small round viruses	—	1	3	—	4
Tissue culture inoculation					
No. tested	74	54	70	21	219
No. positive (%)	7 (9)	8 (15)	6 (9)	3 (14)	24 (11)
Echoviruses	6	6	4	3	19
Coxsackie A viruses	—	2	—	—	2
Coxsackie B viruses	1	—	2	—	3

25 adults and 34 children, almost all semi-nomadic; in October 1978, from the nomadic children in the Tamanrasset boarding-school; in May 1980 from the children of semi-nomads settled in two villages in the north of the Ahaggar, Ifragh and Mertoutek, during an outbreak of gastroenteritis.

The stools, in sterile pots, were kept at 4 °C before transport to Toulouse (never more than 15 days). They were treated as soon as they arrived at Toulouse in the Microbiology Laboratory at the Centre Hospitalier Universitaire de Purpan. Two tests were used, immunoelectronmicroscopy (IEM) and tissue culture inoculation, but the small amounts of stool available meant that it was not possible to do both tests on all specimens. Suspensions of faeces in phosphate-buffered saline were frozen to −25 °C, and after thawing centrifuged at 5000 *g* for 30 min; 0·1 ml of supernatant fluid was placed in two wells of microtitre plates containing a 1 % agar suspension with 4 % pooled human gamma globulin (obtained from the Centre Français de Transfusion Sanguine). Carbon-formvar-coated grids were floated on the supernatants in the wells, and the plates were incubated for about 30 min at 37 °C. The grids were stained with 3 % phosphotungstic acid (pH 6·0) and examined in a Siemens electron-microscope (Elmiskop).

The samples were also inoculated into tissue cultures following antibiotic treatment, according to standard techniques. The tissue cultures used were: primary monkey kidney, human fetal diploid cells (WI 38 in 1977 and 1978, MRC5 in 1980), continuous passage cell lines (Hela and Vero in 1977 and 1978, Hela and RD in 1980). The isolated viruses were typed according to Lim and Benyesh-Melnick's microneutralization technique (1960), using antiserum pools supplied by the World Health Organization. Certain rare types such as coxsackie A15 and A16 were typed in the World Health Organization Reference Laboratory at Lyon.

Viruses were demonstrated by IEM in 43 of 165 stools (26 %) (Table 1). The types of particles found are shown in the Plate. Rotaviruses were rarely found: almost all in October 1977, in three children from the nomads' dwellings and four from people of fixed abode at Tamanrasset. Coronaviruses were the most common viruses found: these were found during each sample period, always in the



The four types of virus seen by IEM, rotavirus (A), coronavirus (B), adenovirus (C) and small round virus (D). All magnifications ×150000.

(*Facing p.* 172)

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Table 2. *Comparative analysis of results from acute gastroenteritis patients and subjects without disease*

	Acute gastroenteritis			Patients without disease		
	Neonates	Children	Adults	Neonates	Children	Adults
Immunoelectronmicroscopy						
No. tested	23	9	2	16	103	12
No. positive (%)	3 (13)	3 (33)	1 (50)	3 (19)	32 (31)	1 (8)
Rotaviruses	2	—	—	1	5	—
Coronaviruses	1	3	1	1	20	—
Adenoviruses	—	—	—	—	4	1
Small round viruses	—	—	—	1	3	—
Tissue culture inoculation						
No. tested	31	10	2	11	130	35
No. positive (%)	4 (13)	—	—	1 (9)	18 (13)	2 (6)
Enteroviruses	4	—	—	1	18	2

semi-nomads. The highest incidence was found in May 1980 during an outbreak of gastroenteritis and in three cases during this period an echovirus was found concurrently. Non-cultivable adenoviruses were observed in three children in the nomads' school, an adult semi-nomadic and one hospitalized newborn from a non-nomadic family. In four cases, small virus particles (less than 35 nm diameter) were seen. The inoculation of tissue cultures revealed 24 enteroviruses from 219 stools (11 %) (Table 1). They were echovirus types 2, 3 (the commonest) 9, 12, 20, 27, 33 and coxsackie virus types A15, A16, B1, B3. No polioviruses were isolated during this study. Like the coronaviruses, the echoviruses were almost all found in the semi-nomadics.

Most of the specimens (80 %) were from individuals without disease (Table 2) and viruses were detected in only 11 of 34 cases of acute gastroenteritis. In seven of these cases the viruses (two rotaviruses, five coronaviruses) may have been responsible for the disease (Table 2). Parasites (*Giardia intestinalis* or *Entamoeba histolytica*) were found in nine of the 28 stools from gastroenteritis patients which were appropriately tested but in only three did they coincide with viruses (all echoviruses).

The results indicate that the frequency of occurrence of rotaviruses in the Ahaggar was low. However, studies by Hieber *et al.* in the U.S.A. (1978), Schnagl, Holmes & Mackay-Scollay (1978*a*) in Australia, and Maiya *et al.* (1977) in India, have shown that the incidence varies with climate or degree of humidity. The low incidence could be explained by the drought or because we took no samples in the winter when the rotavirus incidence is known to be higher in most countries (Flewett, 1977). Moreover samples were taken during an epidemic of diarrhoea only in the last period (May 1980).

We were surprised by the frequency of coronaviruses in the Ahaggar, especially in May 1980. During this period, as in the study by Caul and Clarke (1975), the sampling was done while there were numerous cases of gastroenteritis in the two villages concerned. Particles resembling coronaviruses were found in the stools of

5 out of 9 subjects with diarrhoea, and 8 out of 11 controls. However, in the latter the history did not exclude with certainty gastrointestinal disturbances in the preceding few days. These results are comparable to those of Schnagl, Holmes & Mackay-Scollay (1978*b*) and Schnagl, Morey & Holmes (1979) in Australia, and to those of Mathan *et al.* (1975) in India. We also observed the atypical particles which they noticed.

Although the Daira of Tamanrasset is a hyperarid zone, a prevalence of enteroviruses (13·5%) comparable with or higher than that of noticeably more humid countries was seen (Koornhof *et al.* 1979; Mathur *et al.* 1978).

We emphasize that we have no clinical data on the epidemiology of neonatal acute gastroenteritis in the Ahaggar, and winter samples would be necessary to determine the role of rotaviruses.

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