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Short communication

Merkel cell polyomavirus infection occurs during early childhood and is transmitted between siblings

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ABSTRACT

Merkel cell polyomavirus (MCPyV) is thought to be the etiological agent of Merkel cell carcinoma, but little is known about its distribution and modes of transmission. We conducted seroepidemiological surveys in more than 1000 individuals, from two populations from Cameroon. Overall MCPyV seroprevalence was high in both populations (>75% in adults). Data from the first population, comprising mainly children, indicated that MCPyV infections mostly occurred during early childhood, after the disappearance of specific maternal antibodies. Results from the second family-based population provided evidence for familial aggregation of MCPyV infection status. We observed significant sib–sib correlation (odds ratio = 3.42 [95% CI 1.27–9.19], $p = 0.014$), particularly for siblings close together in age, and a trend for mother–child correlation (OR = 2.71 [0.86–8.44], $p = 0.08$). Overall, our results suggest that MCPyV infection is acquired through close contact, possibly involving saliva and/or the skin, especially between young siblings and between mothers and their children.

1. Background

Merkel cell polyomavirus (MCPyV) has recently been identified as the probable etiological agent of Merkel cell carcinoma (MCC), an uncommon, but aggressive skin cancer of neuroendocrine origin [1–3]. Recent serological studies have indicated that this virus is responsible for a common human infection, at least in individuals of Caucasian origin. Indeed, a high specific seroprevalence of MCPyV, increasing rapidly with age since childhood and reaching 50–90% in adults, has been reported in blood donors and populations of patients living in European countries and the USA [4–8]. However, seroprevalence data are lacking for other populations, and the routes by which this virus is transmitted and acquired remain unknown.

2. Objectives

The goal of this work was to obtain new insight into the modes of distribution and acquisition of this human oncogenic polyomavirus from family-based epidemiological analyses in African populations.

3. Study design

This study was carried out on two populations from Cameroon, Central Africa, in which we had previously carried out

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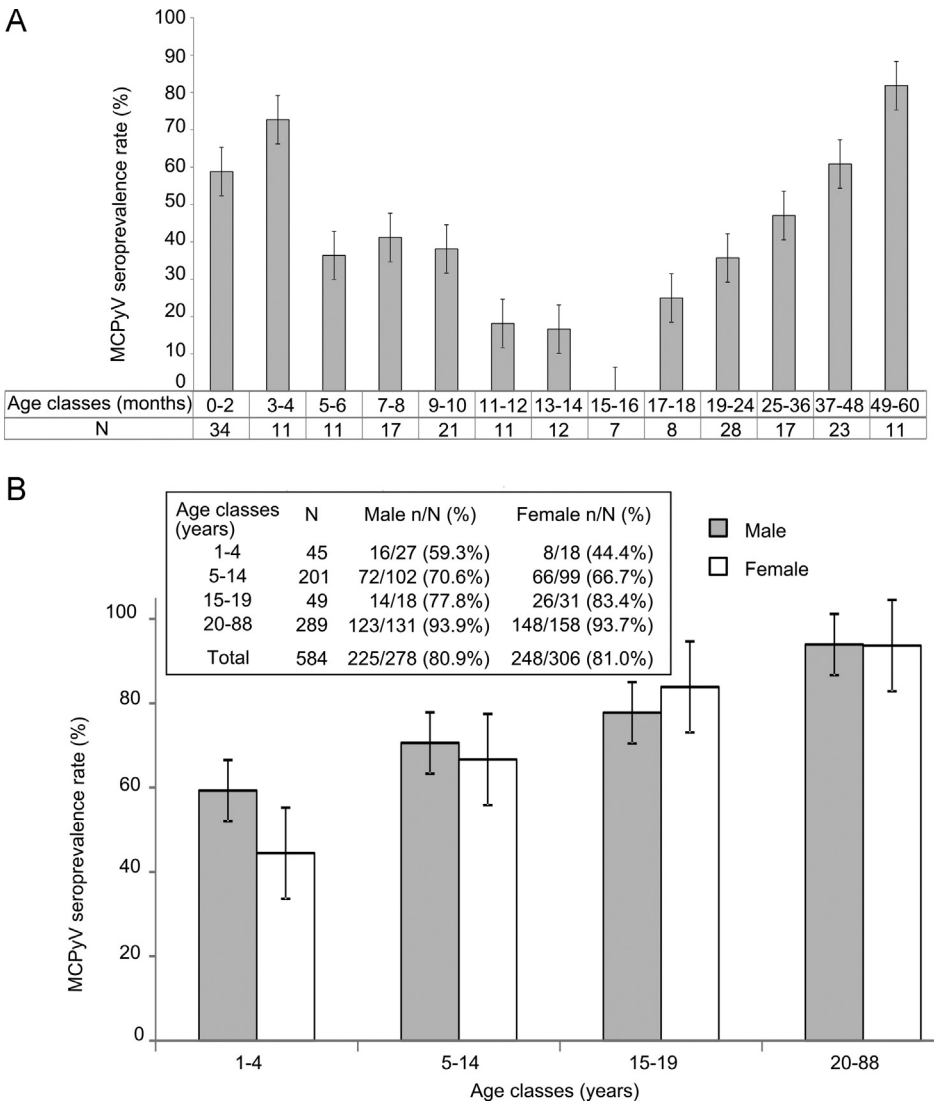

Fig. 1. Age-dependent Merkel virus seroprevalence in the two studied populations. (A) Age-dependent Merkel virus seroprevalence in 196 children aged 0–5 years from Yaoundé, Cameroon. Bars, 95% CI of the seroprevalence rates. N is the number of individuals per age class. (B) Age-dependent Merkel virus seroprevalence in 584 Bantou aged from 1 to 88 years from the village in the Ntem Valley, South Cameroon. Bars, 95% CI of the seroprevalence rates.

epidemiological studies searching for intrafamilial transmission of the human herpes virus 8 (HHV-8), the etiological agent of Kaposi sarcoma [9,10]. The first population, from Yaoundé, consisted mostly of children. The second consisted of villagers living in an isolated rural area of Southern Cameroon in which it was possible to establish familial relationships with full pedigrees. This survey was performed after authorization had been obtained from the local authorities and from the National Ethics Committee in Cameroon. In France, it was approved by the *Comité de Protection des Personnes* (N° 11-02-02). Each participant was provided with information about the study and informed consent was obtained from adults or from the parents of minors.

An ELISA detected specific antibodies against MCPyV. More specifically, anti-VP-1 antibodies were detected with MCPyV-like viral particles (VLPs) generated in insect cells, as previously described [7,11]. Briefly, microplates (Maxisorp, Nunc) were coated overnight at 4 °C with 100 ng/well of purified MCPyV VLPs. Plasma samples were tested at a 1:100 dilution and human IgG binding was detected by peroxidase-conjugated anti-human IgG (Southern Biotech, Clinisciences, Nanterre, France) diluted 1:20,000. A cut-off value for positive samples of 0.2 was used as previously determined [7,11].

4. Results

In the first population, which comprised 458 individuals (229 male and 229 female subjects), 68% of whom were children, the overall seroprevalence of antibodies directed against MCPYV VP-1 was 59%, with no significant difference ($p = 0.25$) between male (57%) and female (62%) subjects (Supplementary Figure 1). We further investigated the age at which the virus was acquired, by focusing on young children. Seroprevalence was quite high, at about 60–70%, from birth until the age of 4 months (Fig. 1A). Interestingly, this seroprevalence is very similar to that observed in women of child-bearing age (~70%). Seroprevalence then decreased with age, reaching 0% at 15–16 months of age, although there were only seven children in this age class. Seroprevalence then rapidly and steadily increased, beginning at 17 months of age, to reach about 60–80% in children aged 4–5 years. This pattern of seroprevalence in young children is consistent with the presence of maternal antibodies in very young children. These maternal antibodies then progressively disappear and infection is rapidly acquired in most children, beginning from the age of about 16 to 18 months.

Table 1
Distribution of pairs according to the familial relationship and MCPyV serological status.

Type of pair	Number of pairs[a]				Total	Odds ratio[b] [95% CI]	*p*
	−, −	−, +	+, −	+, +			
1. Father–child	3	9	42	105	159	1.03 [0.18–5.91]	0.97
2. Mother–child	6	6	57	175	244	2.71 [0.86–8.44]	0.08
3. Child–child							
All children	52	50	83	218	403	3.42 [1.27–9.19]	0.014
Age difference <7 years	39	34	41	128	242	5.00 [1.63–15.33]	0.005
Age difference ≥7 years	13	16	42	90	161	1.60 [0.4–6.44]	0.12

[a] MCPyV status of the first (father for pair 1, mother for pair 2 and oldest sibling for pair 3) and second members of a pair (+ = seropositive, - = seronegative).
[b] Familial odds ratio estimated with the second-order estimating equations approach.

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Our second analysis was based on a familial sample collected from an isolated village located in Southern Cameroon [10]. We specifically studied a total of 584 individuals (278 male and 306 female subjects) aged from 1 to 88 years (median age: 19 years). The overall MCPyV seroprevalence was 81%, with no significant difference ($p = 0.94$) between male (80.9%) and female (81%) subjects. Seroprevalence increased rapidly with age, from 53% in children aged 1–4 years to 94% in adults over the age of 20 years (Fig. 1B). Various age coding schemes were tested to account for this highly significant age effect ($p < 0.00001$), and the best-fit was obtained for a model in which the logarithm of age was considered as a quantitative variable. The 584 subjects were clustered into 65 families, each with two to 25 members, and the second step of the analysis was to estimate the familial aggregation of serological status for MCPyV (infected or non-infected). The correlation between spouses (i.e. the father and mother of the family) could not be estimated, as almost all adults were seropositive. We therefore estimated the following three familial correlations: father–child, mother–child and sib–sib. Familial odds ratios (ORs) were estimated with the second-order estimating equations approach (EE2), and adjusted for age (by a logarithmic transformation) as previously described [12]. All EE2 analyses were carried out with a program described elsewhere [13], and the results are shown in Table 1. No significant correlation ($p = 0.97$) was found between father and child (OR = 1.03, 95% confidence interval [0.18–5.91]), but a trend for a high OR (OR = 2.71 [0.86–8.44], $p = 0.08$) was found between the mother and her children. Finally, MCPyV infection status was significantly correlated ($p = 0.014$) between siblings, with an OR of 3.42 [1.27–9.19]. Furthermore, the OR was higher for pairs in which the two siblings were closer in age (age difference of less than 7 years, to give a balanced number of sib–sib pairs in each subgroup), in which it reached 5.0 [1.63–15.33] ($p = 0.005$), than for sibling pairs with an age difference ≥7 years (OR = 1.6 [0.4–6.44], $p = 0.12$).

5. Conclusions

This study clearly demonstrates the high seroprevalence of antibodies directed against MCPyV in children living in Central Africa. This confirms previous findings of a high level of serological evidence of MCPyV exposure in children living in the USA and Europe, extending these findings to subjects of African origin [4–8]. Indeed, the few previous studies, carried out in children, reported a MCPyV seroprevalence of 25–50%. Moreover, our epidemiological data indicate that most primary MCPyV infections (at least 60–70% of these infections) occurred during early childhood, before the age of 6 years. Our data also strongly suggests that mother–fetus transmission is not a major route of MCPyV transmission [14]. Lastly, the correlations in serological status for MCPyV infection observed within families strongly suggest that MCPyV is transmitted between (especially if they are close in age) and, probably, from mother to child. This pattern is similar to that found in this population for HHV-8, which is thought to be transmitted through close contacts involving saliva [10]. MCPyV is present in several human tissues and in urban sewage, but this virus is found principally in the saliva and on the skin [15–17]. Overall, our analyses suggest that MCPyV can be transmitted through close interpersonal contact involving saliva and/or the skin, particularly between young siblings and between mothers and their children.

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Competing interests

All authors: No reported conflicts.

Ethical approval

The study received administrative and ethical clearance in Cameroon, from the research division of the Ministry of Public Health and from the National Ethics Committee. In France, it was approved by the institutional review board (the *Comité de Protection des Personnes*), the consultative committee relating to the processing of information in scientific research (No. 11.176) and the Commission nationale de l'informatique et des libertés (DR-2011-398).

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