



Prevention of child-to-mother transmission of cytomegalovirus by changing behaviors: a randomized controlled trial

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Abstract

Background: To determine whether a behavioral prevention approach reduces child-to-parent transmission of cytomegalovirus.

Methods: Subjects were seronegative mothers whose child was less than 36 months of age and was shedding cytomegalovirus. Nonpregnant women were randomly assigned to three groups. Mothers in the education group (E) were given instructions about protective behaviors (frequent hand washing, wearing latex gloves) and risky behaviors to avoid (intimate contact with the child). Disposable diapers, liquid soap and latex gloves were provided. During biweekly home visits glove and soap use were monitored for an indirect objective measure of adherence to the protective behaviors. Throughout the study mothers self-reported the frequency they engaged in protective and risky behaviors. In addition to the procedures for Group E the adherence and education group (A) also received social reinforcement for adherence and problem solving for any perceived problems with the behavioral recommendations. The control group (C) received no intervention. A fourth group of pregnant women received an intervention equivalent to that of the education group.

Results: Eight of 17 women in Group C and 4 of 11 women in Group E seroconverted. For both E and Group C the average time from enrollment to infection was 4 months (range, 2 to 7 months). Two of 8 women in Group A seroconverted (1 at 3 months and 1 at 8 months). None of 14 pregnant women observed for an average of 8.4 months during pregnancy seroconverted.

Conclusions: These results suggest that intervention for pregnant women is effective because pregnant women will perceive a higher risk and be more motivated to adhere to recommendations than nonpregnant women.

INTRODUCTION

Approximately 40% of infants born of mothers who acquire a primary cytomegalovirus (CMV) infection during pregnancy

are infected *in utero* and born with a congenital CMV infection.1 Congenital CMV infection can result in mental retardation, deafness and other neurologic problems and up to 8000 infants are born annually in the US with CMV-related neurologic handicaps.2 An important risk factor for maternal acquisition of CMV infections is having young children younger than 3 years of age with CMV infections acquired in group day care.3-5 Seronegative mothers with infected children acquire CMV infections at rates 20 to 25 times higher than other women, and at least one-half of seronegative mothers will become infected within 1 year after their child becomes infected.3, 6 Children younger than 3 years who acquire CMV postnatally excrete CMV in urine and saliva for 6 to 42 months (a mean of 18 months).7 In the US approximately 50% of the mothers of children in day care are seronegative and one-half of all young children are in group day care. Between 15 and 70% of all children in group day care acquire a CMV infection.8 These facts indicate that prevention of maternal infection during pregnancy is important for reducing the frequency of congenital infection.

Because studies of vaccines against CMV are only in their nascent stages, 6 it is important now to determine whether modifying potentially risky maternal behavior reduces the rate at which CMV-seronegative women acquire CMV from their children. We previously developed an intervention protocol that was feasible and acceptable to mothers and that could potentially change protective and risky behaviors for mothers of young children in day care. 9 This protocol was therefore evaluated in a trial comprised of both pregnant and nonpregnant seronegative women with children excreting CMV.

METHODS

Subjects. Subjects were seronegative mothers whose child was shedding CMV and was enrolled in 1 of 15 day-care centers in Richmond, VA, during 3 years (1992 through 1994). All children were younger than 36 months of age at the time of maternal enrollment.

Protocol. Each day-care center was randomly assigned to one of three groups, A, E or C, described below. To achieve approximately equal sample sizes in each group, annually the assignment of each center was randomized again but the randomization was weighted based on enrollment in the previous year. Families enrolled in one group did not cross over but remained in their group until termination. Randomization was done with SAS. 10

Education (E). At an orientation session mothers were given information about CMV infection and its potential complications. Women in Group E were given detailed written and oral instructions for frequent hand washing, wearing gloves for diaper changing and avoiding a variety of types of intimate contact with their child. At biweekly home visits adherence was assessed. Mothers estimated the percentage of opportunities that they performed protective or risky behaviors on a scale of 0 to 100%. After establishing a baseline we discussed and answered questions about the procedures with each mother at the day-care center for approximately 15 min.

Education and adherence (A). Group A was treated the same as Group E except that at the orientation session hand washing and glove changing techniques were demonstrated and then performed by the mothers, and at biweekly meetings the research nurse problem solved and provided positive reinforcement to enhance adherence.

Controls (C). Group C women received basic information about CMV and the reasons for the study but no intervention. Included also for comparison were two groups of previously published historical controls, each comprised of seronegative mothers with children younger than 3 years of age excreting CMV. 3, 6

Pregnant (P). For ethical reasons seronegative pregnant women were not randomly assigned. Women who were pregnant received an intervention equivalent to that for Group E except visits to the home were not made to confirm glove and soap usage.

Participation in the study and monitoring continued for 9 months or until a women requested to terminate, became pregnant or began to attempt to become pregnant. Four seronegative women became pregnant or attempted to become pregnant and were reassigned to Group P.

Protective behaviors. Protective behaviors included frequent hand washing (i.e. after exposure to a child's bodily fluids, after diaper changes, handling dirty laundry, touching the child's toys and other objects and bathing the child) and wearing protective gloves (i.e. during diaper changes). Mothers were given boxes of latex gloves, bottles of liquid hand soap and disposable diapers.

Risky behaviors. Risky behavior included intimate contact with the infant child such as kissing on the mouth, sleeping

together, sharing towels and washcloths and eating or drinking from the child's utensils.

Measures. Measures included the self-reported percentages of opportunities that they performed protective or risky behaviors on a scale of 0 to 100%. At a biweekly home visit a research nurse provided a new box of gloves, a new bottle of hand soap and disposable diapers. For objective measures of adherence to protective behaviors, the number of remaining gloves was counted, the amount of remaining soap was weighed and the number of diapers used were counted.

Monitoring for CMV. For each woman enrolled, saliva, serum and urine were collected every 3 months for 9 months. Fathers and other children in the home had urine and saliva obtained every 3 months. All samples were cultured for CMV. 11 Seroconversion was measured by an enzyme immunoassay. 12 Neither the women nor the research nurse were aware of the results of virologic or serologic testing until the end of the study. Genome analysis of all isolates was completed with restriction enzymes as described previously. 13

Statistical analysis. Infection rates in the groups were compared by computing Kaplan-Meier survival estimates and tested for group differences using a log rank chi square test. Relationships between survival time (time to seroconversion) and child and maternal age were tested by a proportional hazards model. The measures of adherence in Groups A and E were compared by repeated measures analysis of variance. Average daily adherence measures were also calculated for each subject to illustrate the magnitude of differences. All statistical tests were with using JMP software.10

Informed consent was obtained from all subjects. The human experimentation guidelines of the US Department of Health and Human Services and those of Virginia Commonwealth University/Medical College of Virginia were followed in the conduct of the clinical research.

RESULTS

Seroconversion by group. During the 3 years of the study 116 seronegative women with a child excreting CMV in urine or saliva were identified. Twelve women refused to participate. Thirty-nine others were ineligible because they were likely to be unavailable for 9 months, or they had participated in a previous study or they had already been instructed about possible ways to avoid acquiring CMV from an infected child. The children of 12 women (5 in Group A, 3 in Group E, 3 in Group C and 1 in Group P) spopped shedding after enrollment, and these 12 women were excluded from the analysis of the association of seroconversion rates and group assignment (Tables 1 and 2). The 4 groups studied were similar for maternal age and age of the children at enrollment (Table 1).

Creare	Number	Mean Age at Entry		
Group		Mother (yr)	Child (mo)	
A	11	$33.4 \pm 3.4^{*}$	24.2 ± 6.0	
E	11	32.9 ± 4.4	16.8 ± 5.1	
С	17	31.9 ± 3.6	19.4 ± 6.4	
Р	14	32.4 ± 3.9	22.6 ± 7.8	
Historical Group 1"	36	32.4 ± 3.6	20.7 ± 7.2	
Historical Group 2 ⁶	38	33.8 ± 4.9	18.1 ± 6.6	

TABLE 1. Comparison of groups by maternal and child's age

Group	No. of Subjects		Total Days Observed	
	Enrolled	Infected	Mean	Range
A	11	2 (18)*	199	66-281
Е	11	4 (36)	180	68-372
С	17	8 (47)	135	60 - 273
Р	14	0(0)	252	92-503
Historical Group 1 ³	36	19 (53)	377	27-930
Historical Group 26	38	17 (45)	516	30 - 756

TABLE 2. Infection rates by group

Table 2 lists the infection rates and observation times by group. Among Groups A, E and C, Group A had the lowest infection rate (18%); however, according to survival analysis the time to seroconversion among these three groups were not significantly different (chi square 2.5, df = 2, P < 0.29). None of 14 pregnant women acquired CMV during pregnancy. The time to seroconversion for this group was significantly lower than observed for Groups A, E and C (chi square 6.7, df = 2, P < 0.29). Group assignment was not associated with the interval between enrollment and infection (Fig. 1).



FIG. 1. Survival estimates for 53 mothers of infected children. Mothers were assigned to one of four groups: adherence (A),

education (E), control (C) or pregnant (P). Pregnant women (P) differed significantly from those of the other groups (P < 0.01).

Two groups of historical controls, one with 36 subjects and the other with 38, were used to augment the size of Group C in this study (Tables 1 and 2). Group C and the 2 historical control groups did not have significantly different distributions of the intervals between enrollment and seroconversion (chi square 1.69, df = 2, P < 0.4286). When these 3 control groups are collapsed and compared with Group A, Group A still did not experience a significantly lower time to seroconversion (chi square 1.129, df = 1, P < 0.5687).

Without considering the historical controls, survival analysis was also performed to determine whether the group differences were associated with maternal age or age of the child at enrollment. Maternal age was unrelated to time to seroconversion (chi square 0.72, df = 1, P < 0.4). Having a child <20 months of age at enrollment was an independent predictor of infection (chi square 6.7, df = 1, P < 0.01); however, after consideration of group assignment having a child <20 months of age is not significant (chi square 3.2, df = 1, P < 0.07)

Viral shedding. Of the 14 mothers who seroconverted, 7 shed CMV. Six of the 7 mothers shed strains identical by genome analysis with those shed by the their child. One mother in Group C shed an isolate that differed by genome analysis from that shed by her child suggesting maternal acquisition form a source other than her child. Two fathers shed CMV. Both of their wives seroconverted, one in Group A and one in Group C, but the father in Group C shed an isolate that differed by genome analysis from that shed by his wife and child.

Measures of adherence. Women in Groups A and E were monitored for behavioral adherence (Table 3).

Variable	odean/a	F tout*	P	
* at satisfie	Group A	Group E		
Objective measure	States and the second			
No. of biweekly samples	$18.0 \pm 8.6^{+}$	17.9 ± 6.7		
Total days observed	204.8 ± 85.4	222.4 ± 80.4	<1	
No. of diapers/day	2.5 ± 1.2	2.4 ± 0.7	<1	
g of soap/day	7.1 ± 4.6	6.0 ± 3.0	<1	
No. of gloves/day	4.6 ± 2.0	4.3 ± 1.8	<1	
Self reports				
No. of times biweekly				
Changed child's diaper	27.8 ± 14.0	29.9 ± 9.3	<1	
Gloves worn while changing diapers	23.6 ± 14.2	24.5 ± 10.1	<1	
Washed hands after changing diapers	27.0 ± 13.7	28.7 ± 9.8	<1	
Helped child use potty chair or toilet	3.5 ± 6.3	2.5 ± 6.0	<1	
Washed hands after helping child use potty chair or toilet	3.5 ± 6.3	2.4 ± 6.0	<1	
Washed child's laundry	3.4 ± 1.7	4.0 ± 3.7	<1	
Gloves worn while washing child's laundry	2.8 ± 1.8	3.1 ± 3.8	<1	
Washed hands after washing child's laundry	3.4 ± 1.7	3.7 ± 3.8	<1	
Bathed child	6.0 ± 2.8	6.7 ± 3.8	<1	
Washed hands after bathing child	5.9 ± 2.8	5.8 ± 3.3	<1	
Kissed child on lips	0.6 ± 0.6	0.4 ± 0.9	<1	
Ate/drank after child	0.3 ± 0.8	0.2 ± 0.3	<1	
Slept in the same bed with child	0.9 ± 1.3	1.5 ± 2.6	<1	
Used towel or washcloth after child	0.1 ± 0.2	0.2 ± 0.3	<1	
% of time biweekly				
Washed hands after handling child's objects	81.6 ± 9.3	69.2 ± 21.0	5.33	0.0285
Washed hands after wiping child's nose	89.6 ± 8.6	73.8 ± 24.9	6.63	0.0155
Washed hands after spending a lot of time with child	82.6 ± 10.1	70.3 ± 24.5	4.00	0.0548
peated measures F test with 1.29 df.				

TABLE 3. Monitoring of child care activities by group

Women in Group A self-reported more frequent hand washing than women in Group E, but the average amounts of gloves and soap used by each group were the same (Table 3). There were no other significant differences in either indirect objective measures (monitoring) or subjective measures (self-reporting).

For Groups A and E combined, infected and uninfected women were similarly compared (Table 4). There were no significant differences in either indirect objective measures (monitoring) or subjective measures (self-reporting).

	Mean/Subject			
	Infected $(n = 6)$	Not infected $(n = 16)$	F Test*	Р
Monitoring				
Samples	$15.5 \pm 5.5^{\dagger}$	19.4 ± 8.1		
Sum (days)	197.0 ± 67.0	223.5 ± 78.8	<1	
Diapers/day	2.7 ± 0.6	2.2 ± 1.1	<1	
Soap/day	7.1 ± 4.0	5.6 ± 3.7	<1	
Gloves/day	3.9 ± 2.3	4.5 ± 2.0	<1	
Self reports				
No. of times biweekly				
Changed child's diaper	32.2 ± 14.3	26.5 ± 11.5	<1	
Gloves worn while changing diapers	22.9 ± 15.5	22.4 ± 11.0	<1	
Washed hands after changing diapers	30.4 ± 15.3	25.5 ± 11.1	<1	
Helped child use potty chair or toilet	1.1 ± 2.1	3.6 ± 6.5	1.09	0.3075
Washed hands after helping child use potty chair or toilet	1.1 ± 2.1	3.5 ± 6.4	1.01	0.3261
Washed child's laundry	2.9 ± 1.3	3.9 ± 3.7	<1	
Gloves worn while washing child's laundry	1.7 ± 1.5	3.3 ± 3.7	<1	
Washed hands after washing child's laundry	2.5 ± 1.0	3.8 ± 3.7	<1	
Bathed child	7.3 ± 1.8	5.6 ± 4.0	1.12	0.3024
Washed hands after bathing child	6.8 ± 2.2	5.0 ± 3.4	1.44	0.2434
Kissed child on lips	0.2 ± 0.3	0.7 ± 1.0	1.34	0.2587
Ate/drank after child	0.2 ± 0.3	0.2 ± 0.5	<1	
Slept in the same bed with child	0.2 ± 0.3	1.2 ± 1.9	1.16	0.2404
Used towel or washcloth after child	0.1 ± 0.3	0.0 ± 0.1	2.98	0.0971
% of time biweekly				
Washed hands after handling child's objects	73.4 ± 16.8	78.6 ± 12.7	<1	
Washed hands after wiping child's nose	80.8 ± 12.8	83.4 ± 20.0	<1	
Washed hands after spending a lot of time with child	74.5 ± 19.0	81.0 ± 14.8	<1	
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TABLE 4. Monitoring of child care activities by infection status

DISCUSSION

CMV-seronegative women who have frequent contact with young children in group day care have a 5- to 25-fold increased risk for acquiring CMV from these children than do seronegative women not in contact with young children. 3-5, 14-16 Although harmless for the nonpregnant women, the consequences of CMV acquisition during pregnancy can be severe for the developing fetus. 2 Whether CMV transmission occurs via contact with contaminated environmental surfaces (fomites), saliva, urine or respiratory droplets is unknown. Transmission via respiratory droplets is unlikely but CMV survives on fomites, including diapers, toys and the hands of day care personnel. 17-19 After experimental inoculation of diapers with virus in urine, CMV can be recovered for up to 48 h after inoculation, with viral titers remaining between 10² to 10³ plaqueforming units/ml for the first 12 h 18 and CMV was recovered from objects and hands for up to 30 min after salivary contamination from infected children. 19 CMV, a lipid-enveloped virus, is readily inactivated by most detergents, soaps and alcohols. Improved hygiene and more frequent hand washing should reduce the frequency of child-to-mother transmission. We therefore attempted to determine whether behavioral intervention would reduce the risk for CMV infection among pregnant and nonpregnant seronegative women. We found that for nonpregnant women behavioral intervention was not successful.

Our study, however, was limited by an insufficient sample size to detect potentially statistically significant differences between women in the control group and those in the intervention groups. For the nonpregnant women intervention may have reduced the risk of acquiring CMV by as much as 2-fold. However, the significant difference in infection rates between the nonpregnant women and the pregnant women suggests that pregnant women are much more highly motivated than nonpregnant women. No CMV infections occurred among the pregnant women who received a limited intervention. Pregnant women were instructed on how to reduce risks but their success was not monitored nor did they receive reinforcement during the study. Because of the low rate of infection among the pregnant women and because the target group for intervention is pregnant women, we terminated enrollment of nonpregnant women after a significant difference was observed between pregnant and nonpregnant women. Additional pregnant women will have to be monitored more precisely to determine the reduction in relative risk afforded by intervention.

Fifty percent of the mothers who seroconverted shed CMV. This rate is identical with the 50% rate of viral shedding that we observed in previous studies using the same schedule of sample collection. 3, 6 We also previously observed that approximately 95% of mothers who seroconverted and shed CMV shed an isolate with the same genome pattern as excreted by their child, indicating that the young children were the source of infection for the majority of these women. 3, 6 For this reason and because one-half of the mothers did not excrete CMV, the end point for the study was seroconversion. Therefore the mother in Group C who shed a different strain than her child was included in the data analysis.

Behavioral invention as practiced by the mothers in this study was feasible. Reports from all mothers indicated that protective behaviors were easily performed, although several did not like using latex gloves. None of the mothers were concerned about reducing the specific types of intimate contact with their children. Nearly all of the mothers were able to

make changes consistent with the recommendations to increase protective behavior and decrease risky behaviors and to sustain these changes for an average of 200 days/subject. We did not, however, observe an association between either the subjective measures (selfreporting) or the objective measures (soap, glove and diaper use) and infection rates. Self-reporting and indirect objective measures may also be unrelated to the actual behavior change; therefore the validity of targeting these behaviors was not established. We did not rigorously monitor the pregnant women for fear of enhancing anxiety during pregnancy.

In spite of the encouraging results of this study, how to advise mothers with young children in day care who are anticipating additional pregnancies remains difficult. Serologic testing of all pregnant women may not be practical, cost effective or necessary. However, testing mothers with young children in day care may help because prevention of CMV infection during pregnancy appears possible. Mothers with children in day care who are pregnant or anticipating pregnancy could be given the option for serologic testing of themselves and their child. Serologic testing is simple, rapid, very accurate, commercially available and relatively inexpensive and can be performed on the serum obtained by a finger prick. Seropositive children between 6 months and 2 years of age should be considered potentially infectious. 11 Seronegative mothers of seropositive children could be counseled to avoid intimate contact with their child during pregnancy. Counseling would advise frequent hand washing, the use of gloves, especially when handling diapers or respiratory secretions, and avoidance of mouth to mouth contact. Our results suggest that counseling and intervention for pregnant women will be effective because pregnant women will perceive a higher risk and be more motivated to adhere to recommendations than nonpregnant women.

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Photo Insert shows the appendage that penetrates the skin to suck blood. It is apparent why it is difficult to remove the tick and the appendage. The scaly flaps work like an open umbrella when an attempt is made to remove the tick.

Why the Tick Sticks

Photograph by Helieh Oz, D.V.M., Ph.D. Memphis, TN

Key words: Cytomegalovirus; child day care; pregnancy

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