A Research Note

Herpes Simplex Virus Type 1 Applied Experimentally to Gloves Used for Food Preparation

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ABSTRACT

Droplets of saliva containing herpes simplex virus type 1 were placed on latex disposable gloves. The temperature at the surface of the gloved hand was 34°C. There was no loss of infectious virus before 15 min. Between 15 and 30 min there was a 2-log-cycle drop in titer, and infectious virus could still be recovered after 1 h, the longest period tested. The drop in titer was due to drying of the saliva, which occurred at approximately 21 min. Infectious virus was transferred by touch to lettuce and ham at 0 min when the virus-containing droplets were in a liquid condition, and after 30 and 60 min when the droplets were dry.

Key words: Herpesvirus, saliva, latex gloves, lettuce, ham

People often wear gloves when preparing or serving food to be eaten by others. This is particularly true in public eating places. Indeed, wearing disposable gloves is recommended if food must be manipulated by hand (5). If gloves are used, they should be intact, clean, and in a sanitary condition (3). Gloves should be changed following contamination by microorganisms, or if there is doubt concerning contamination. The foregoing notwithstanding, it is not uncommon for gloves to be worn for long periods without being changed, even after contamination may have occurred, for instance, by persons making sandwiches, since they may believe that their gloves do not become contaminated because they are handling food prepared under sanitary conditions. However, as observed by this author, it is not unusual for food handlers to put their gloved hands to their mouths and noses to cover a cough and or to hold a tissue to wipe or blow the nose. After such actions, it is not unusual for them to continue working without changing their gloves. Both the mouth and the nose are habitats for a variety of microorganisms, and are a potential source for contamination of hands that come in contact with those parts of the face or with secretions from the mouth and nose.

Herpes simplex virus type 1 (HSV-1) is the agent of a variety of human diseases ranging from minor to severe illnesses (7). The virus most frequently infects the mouth and face but can cause diseases elsewhere, including the central nervous system.

HSV-1 is widespread in the population at large, causing overt diseases and subclinical infections. The virus can be shed by persons with or without symptoms of disease; thus, healthy carriers can disseminate the virus. Transmission in oral secretions is one mechanism by which HSV-1 is spread from person to person (4). Moreover, the hands have a role in transmission of the virus (8).

In consideration of a report that herpesviruses in body secretions were found to survive for as long as 2 h on human skin (9), and in view of the above discussion, an investigation was undertaken to examine the survival of HSV-1 in saliva on disposable gloves and possible transmission of the virus from gloves to food.

MATERIALS AND METHODS

Virus and cell cultures

HSV-1 (strain F) and an established line of human epithelialoid cells (HEp-2) were purchased from the American Type Culture Collection, Rockville, MD. HEp-2 cells are the cells of choice for laboratory propagation of the F strain of the virus (2). Cells were grown at 37°C in Eagle’s minimum essential medium supplemented with 10% fetal calf serum. The medium contained 100 units of penicillin and 50 μg of streptomycin per ml. HSV-1 was propagated by serial passage in HEp-2 cells. The virus was titrated in tubes containing monolayer cultures of HEp-2 cells. To each tube was added 0.9 ml of growth medium. Serial 10-fold dilutions of virus in growth medium were prepared, and 0.1 ml of each dilution was inoculated into the tubes. Six tubes were inoculated for each dilution. The final reading for cytopathic effect (CPE) was made on day 7 post-inoculation, and the dose that would infect 50% of the tissue cultures (TCID50) was calculated.

Ascertainment of HSV-1 survival on latex disposable gloves

For deposition on gloves (Eagle Latex Disposable Gloves, Home Products, Inc., Westbury, NY), HSV-1 was suspended in unstimulated saliva from a healthy adult person. Freshly collected saliva was used in all experiments. Before the addition of virus, the saliva was subjected to two cycles of centrifugation at...
HERPESVIRUS ON LATEX GLOVES

10,000 \times g for 30 min at 4^\circ C. One part of virus stock suspension was added to nine parts of clarified saliva. The pH of the virus-saliva mixture was 7.0, as was the pH of the saliva before adding virus. There were occasional problems of microfungal contamination when using unclarified saliva, hence the need for clarification. Periodic examination of saliva for antibody to HSV-1 gave negative results by an immunofluorescence test (the procedure for the test has been reported previously [1]).

To determine the survival of HSV-1, five 0.01-ml droplets of virus suspension were placed for various periods of time on the palm of a latex disposable glove worn by the investigator. The gloves received no treatment, such as surface sterilization with UV light, before experimental use. They were used in the condition a food handler would use them. Each droplet covered a circular area approximately 0.5 cm in diameter. To recover HSV-1 for quantification, the area of the glove to which virus had been applied was overlaid with 0.1 ml of tissue culture growth medium, which was left in place for 2 min with gentle agitation. The medium was then aspirated, and the virus contained therein titrated as described above. It had been determined that recovery of virus was no greater if the medium was left in place for more than 2 min. Placing the virus-containing droplets on the palms of gloves resulted in a more accurate recovery of virus than placing droplets on the fingers, since there was a tendency for the recovery medium to run down the sides of the fingers.

To act as controls for virus on a glove, 0.01 ml droplets of virus suspension were placed on a sterile glass surface and incubated at 34^\circ C, the temperature at the surface of the palm of the gloved hand. Virus was recovered from glass in the same manner as from a glove. The droplets of virus suspension were completely dry approximately 21 min after being placed on glass and gloves.

For studies requiring prevention of the virus suspension from drying, droplets of virus were put on a glove that was then placed in a humid chamber with a temperature of 34^\circ C.

Transfer of HSV-1 from gloved hand to food

To examine the transfer of virus from a glove to food, 0.01 ml droplets of virus in saliva were placed on the distal end of the three largest fingers of a gloved hand, and at 0, 30, and 60 min a lettuce leaf or piece of ham was firmly touched for 5 s with a finger tip. The lettuce leaf had previously been washed in municipal tap water and then shaken to remove as much water as possible. Sliced ham was used as removed from the sealed package into which it had been placed by the processing company. Thus the lettuce and ham were subjected to the treatment they get when used as ingredients of sandwiches. To determine if virus had been transferred, the touched area of the foodstuffs was subjected to the method described above for recovery of virus from the surface of a gloved hand, after which the aspirated medium was inoculated directly into tubes containing monolayers of HEp-2 cells. The cells were examined over a 7-day period for virus-induced CPE.

RESULTS

Survival of virus on gloves

The results in Table 1 show survival of HSV-1 in saliva on latex disposable gloves for periods of up to 1 h. There was no loss of virus infectivity at 15 min compared with 0 min. There was a marked drop in the titer of infectious virus at 30 min, but thereafter no further reduction occurred. No differences in titers were observed between virus placed on gloves and control virus placed on glass. The droplets of virus in saliva became dry about 21 min after being placed on the gloved hand and also on glass maintained at 34^\circ C. The results were consistent, since similar results were seen in five trials using different virus-saliva suspensions and different subcultures of HEp-2 cells for each trial.

The results in Table 2 show no loss of HSV-1 infectivity for any of the times examined if the virus-containing droplets remained in a liquid state. With controls, there was a drop in titer of infectious HSV-1 with drying of the virus-containing saliva.

Transmission of virus from gloved hand to food

After the surface of a lettuce leaf was touched with the tip of a finger on which HSV-1 had been placed, virus was isolated in five out of five trials at 0 min, when the droplets of virus suspension were in a liquid state. Virus was also isolated in five out of five trials after lettuce was touched at 30 min and 60 min, when the droplets of virus were dry. Similar results were observed after sliced ham was touched.

DISCUSSION

The wearing of gloves by employees is now quite prevalent in public eating places in New York City and northeastern New Jersey, and this geographical location most likely reflects a similar practice in other parts of the country. Food served to the public should be protected

TABLE 1. Survival of herpes simplex virus type 1 on the surface of a gloved hand

<table>
<thead>
<tr>
<th>Survival period (min)</th>
<th>Virus titer (TCID$_{50}$/0.1 ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Control virus on sterile glass$^b$</td>
</tr>
<tr>
<td>0</td>
<td>$10^{5.3}$</td>
</tr>
<tr>
<td>15</td>
<td>$10^{5.6}$</td>
</tr>
<tr>
<td>30</td>
<td>$10^{5.2}$</td>
</tr>
<tr>
<td>45</td>
<td>$10^{5.3}$</td>
</tr>
<tr>
<td>60</td>
<td>$10^{5.5}$</td>
</tr>
</tbody>
</table>

$^a$ Latex disposable gloves were used.
$^b$ Control virus was incubated at 34°C, the temperature at the surface of the gloved hand.
$^c$ By 21 min the droplets of virus-containing saliva were dry on glass and gloves.

TABLE 2. A comparison of the survival of herpes simplex virus type 1 at 34°C in saliva maintained in a liquid state and in saliva that dried with passing time

<table>
<thead>
<tr>
<th>Survival period (min)</th>
<th>Virus titer (TCID$_{50}$/0.1 ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Saliva kept liquid</td>
</tr>
<tr>
<td>0</td>
<td>$10^{5.4}$</td>
</tr>
<tr>
<td>15</td>
<td>$10^{5.6}$</td>
</tr>
<tr>
<td>30</td>
<td>$10^{5.3}$</td>
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<tr>
<td>45</td>
<td>$10^{5.4}$</td>
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<tr>
<td>60</td>
<td>$10^{5.5}$</td>
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</tbody>
</table>

$^d$ The droplets of virus-containing saliva were placed on latex disposable gloves and either incubated in a humid atmosphere or allowed to dry in room atmosphere at 34°C.

$^e$ The saliva dried in approximately 21 min.
from contamination by microorganisms, hence the reason for the use of gloves to help safeguard food. However, habits can lead to gloves becoming contaminated with saliva and other bodily secretions that are vehicles for transmission of microorganisms.

As shown herein, HSV-1 in saliva contaminating latex gloves can survive for at least 1 h, the longest period tested. There was a drop in titer of the virus as time elapsed. This was not due to some kind of antiviral activity associated with the gloves, since a similar reduction in titer was observed with HSV-1 placed on sterile glass. The drop in titer appears to be a consequence of drying, as it occurred at the time the virus-containing droplets became dry. Additional evidence for this can be seen from the study in which droplets on gloves were kept in a liquid state. Under those conditions there was no drop in HSV-1 titer. The results for virus-containing saliva kept in a liquid state also show that the saliva itself had no antihSV-1 activity, as there was no drop in titer of the virus at any of the times tested.

The presence of HSV-1 on latex gloves led to handled food becoming contaminated with the virus. The transfer of virus to food by touch when the virus-containing droplets were in a liquid state was to be expected, since liquid on the hand is readily transferred to touched objects. That might not be expected with virus in a dry state adhering to the finger tips. However, virus in dried saliva was also transferred by touch to food. Washed lettuce and sliced ham are usually moist when used for making sandwiches or eating in other ways. It is likely that the moist surface of the foodstuffs facilitated transfer of the virus, since there would be a tendency for the dried virus-containing saliva to liquefy when brought into contact with the moisture associated with lettuce and ham.

Concerning foodborne viral diseases, poor personal hygiene was the cause, or a contributing factor, in 92% of studied cases (6). Most studies of virus contamination of food have been directed to enteric viruses (6). This reflects the fact that many microorganisms cause diseases of the intestinal tract. Be that as it may, viruses infecting other parts of the body, for example, viruses infecting the mouth and respiratory tract, are likely candidates for contamination of food: on the basis of some unhygienic actions of food handlers noted herein.

ACKNOWLEDGMENTS

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REFERENCES