The present study was designed to demonstrate the recovery of viable hRSV.

Virus stock was hRSV ATCC VR-1540 grown on MRC-5 cells. For each specimen held at either ambient or refrigerator temperatures, the tube was titrated in cultures of MRC-5 cells. Cells were examined daily for the presence of cytopathic effect (CPE). All Time zero swabs showed over 75% CPE by 6 days, while the 96 hours swabs all showed over 75% CPE after 10 days for both temperatures. The study has shown that Sigma Virocult transport swabs will recover hRSV as clinically significant concentrations.

Methods

The basis of the study was M40-A, CLSI's standard for microbiology transport devices, which requires virus transport devices for hRSV to be able to recover and maintain a defined titre of infective virus for 96 hours at either room temperature or refrigerated temperatures.

1. A stock was prepared of hRSV (ATCCVR-1540) in MRC-5 cells, with a titre of $10^4.7$ TCID$_{50}$ per 50µl. The required titre for inoculation is $10^4.7$ TCID$_{50}$, so an inoculum of 88µl was used.

2. 88µl of virus suspension was added to the required number of wells in a sterile tissue culture plate.

3. Swabs were inoculated by inserting the tip into the well and moving around until all of the virus suspension had been absorbed. The swab was then placed in its tube of transport medium, the shaft snapped at its breakpoint, and the cap screwed onto the tube.

4. Each tube was gently shaken, then incubated at either 4°C or 20-25°C for 96 hours.

5. A reference Time Zero swab was also inoculated.

6. Swabs were used for each incubation condition.

7. Cells were examined for the presence of cytopathic effect (CPE) on each day following inoculation for a maximum of 2 weeks.

8. The degree of CPE was recorded as follows:

<table>
<thead>
<tr>
<th>No CPE</th>
<th>&lt;25% CPE</th>
<th>26-50% CPE</th>
<th>&gt;51-75% CPE</th>
<th>&gt;75% CPE</th>
</tr>
</thead>
<tbody>
<tr>
<td>-</td>
<td>+</td>
<td>++</td>
<td>++++</td>
<td>++++</td>
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</tbody>
</table>

Results

The study was performed in accordance with CLSI M40-A.

Conclusion

The study showed acceptable recovery of hRSV at both holding temperatures.

Discussion

Although nasopharyngeal aspirates (NPA) are considered the best type of specimen for the detection of Human Respiratory Syncytial Virus (hRSV), it often impractical for specimens may have to be collected in the community without access to vacuum required for NPA. The procedure for collecting NPA can be painful and distressing for the patient, often an infant. A nasal swab is more acceptable to the patient, but have often been considered unsatisfactory because of lower yields of detectable virus. It is thought that the very small virus particles are more likely to be entrapped in the fibre matrix of the swab bud.

In recent years new materials have been used in the manufacture of swab buds. These include flocked polyester fibres such as the Filifigament polyester fibres in PurFlock® and HydraFlock® swabs from MWE, and the open cell polyurethane foam used with MWE Sigma Swabs®. In the present laboratory study, the ability of Sigma Virocult® to recover hRSV was investigated. Sigma Virocult® combines a novel swab (Sigma Swab® with plastic shaft and soft polyurethane foam bud) with Virocult®, a long established virus transport medium from MWE. The soft foam bud is particularly suited for infant patients, and the cellular structure allows free movement and optimum release of the small hRSV virus particles.

In the study Sigma Virocult® was shown to give excellent recovery of hRSV in swabs held for up to 4 days at either room temperature, or refrigerated temperature, and so would be capable of maintaining the viability and infectivity of hRSV at the limits of the feasible transport periods. The study was performed in accordance with CLSI M40-A.

References

4. CLSI M40-A, Quality Control of Microbiological Transport Systems, Approved Standard 2003