Adenosine Triphosphate Bioluminescence Test of the Nasal Spray Nozzles Attached to an Ear-nose-throat Treatment Unit

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Original

Adenosine Triphosphate Bioluminescence Test of the Nasal Spray Nozzles Attached to an Ear-nose-throat Treatment Unit

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To report a proper protocol for cleaning and disinfecting the nozzles attached to ear-nose-throat (ENT) treatment unit, contamination of nasal spray nozzle, nasal speculum, and suction tube before and after use was assessed by adenosine triphosphate (ATP) bioluminescence test. The measured ATP was expressed as relative light units (RLU).

There were no statistical differences in RLU scores among the nasal spray nozzles, nasal speculums, and suction tubes before patient use. Nozzle RLU scores increased after patient use. The RLU scores (mean ± SD) of the nasal spray nozzles before use was 12 ± 7.1, increasing to 30 ± 28.7 after patient use. The RLU scores of the nasal speculums increased to 9,722 ± 12,398.9 after patient use. The RLU scores of the suction tubes increased to 88,366 ± 106,839.3 after patient use. Increased RLU scores of spray nozzles were statistically lower than those of nasal speculums and suction tubes.

In conclusion, nasal spray nozzles should be wiped with a mid-level disinfectant after use. Alternatively, the nozzle tips should be changed between patients. These recommendations should be considered when developing protocols for cleaning and disinfection of nasal spray nozzles used in conjunction with ENT treatment units.

Key Words: ENT treatment unit, spray nozzle, contamination, ATP bioluminescence test

Introduction

Nasal spray nozzles attached to ear-nose-throat (ENT) treatment units are not routinely changed between patient examinations because the nozzles do not directly contact patients' skin or mucosa. However, there is a risk of cross-contamination by accidental touching of the spray tip to the mucosa, or by contact with nasal discharge, droplets, or aerosolized material because the spray is used near the nostrils (**Fig. 1**). Past studies have reported the detection of coagulase-negative staphylococcus (CNS), methicillin-sensitive *Staphylococcus aureus* (MSSA), and sometimes methicillin-resistant *S. aureus* (MRSA) on the surface of nasal spray nozzles following their use[1)2)].

Microbiological culturing is a traditional method for hygiene monitoring; however, these methods have some limitations. They require several days to get results because a colony growth incubation period is needed. Moreover, these methods cannot detect nonbacterial contaminants such as blood and mucous.

Recently, adenosine triphosphate (ATP)-based micro-

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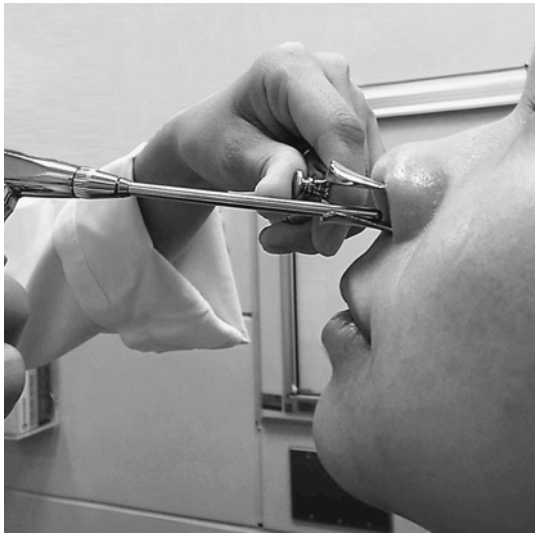

Fig. 1 Using nasal spray near the nostrils of patient

Fig. 2 Nasal spray nozzles attached to ENT treatment unit (arrows: tip)

biologic monitoring methods were developed for hygiene monitoring[3]. These methods measure ATP, the principal carrier of energy in all living organisms including microorganisms. ATP testing can be completed within 10 seconds, and is considerably easier and produces more rapid results compared to traditional swabbing culture methods. We assess the presence of contaminants on the nasal spray nozzle surfaces attached to ENT treatment units via ATP test to report a proper protocol for cleaning and disinfecting the nozzles.

Materials and Methods

The study was conducted in December 2016. We examined six spray nozzles attached to three ENT treatment units located in the consultation room of an ENT practice. Each unit was used to administer two sprays containing 2% lidocaine and 0.02% adrenaline (**Fig. 2**). As per the clinic's standard operating procedures, the spray nozzles were cleaned using hot (90°C) water once per week prior to being reattached to the ENT treatment units.

1. ATP bioluminescence test

Prior to testing, the spray nozzles were wiped with cotton soaked in isopropanol. The 3M™ Clean-Trace™ Surface ATP test system was used, consisting of a test swab, cuvette, and 3M™ Clean-Trace™ NG Luminometer. The ATP testing protocol was performed according to the manufacturer's instructions[4]. The tips of the nozzles were rubbed with a test swab. The swabs were then inserted into a cuvette containing a luciferin, luciferase and Mg^{2+}, causing the following enzymatic reaction:

ATP + luciferin/luciferase→adenosine monophosphate (AMP) and phosphoric residues+ light (wave of 562 nm)

The intensity of this light is proportional to the amount of ATP on the object surface, which corresponds to the amount of contamination by organic residues and microorganisms. Measurement of light intensity requires a Luminometer. This device contains a measuring chamber isolated from external light sources, and a detector that processes the optic signal to the electrical signal, which is expressed in relative light units (RLU)[4]. Baseline RLU scores were recorded as "before use" scores.

The spray nozzles were then used on three patients, and ATP testing was repeated, with the resultant RLU scores recorded as "after use" scores. This cycle of nozzle cleaning, baseline ATP testing, nozzle use with three patients, and repeat ATP testing was performed 4-6 times per each spray nozzle, for a total of 28 times. For comparison, we also obtained RLU scores from the outer surfaces of nasal speculums, which were in direct contact with the nasal cavity, and suction tubes, which were inserted into the nasal cavity.

All statistical analyses were performed using GraphPad Prism version 6.0 for Windows. Paired t-tests were used to compare spray nozzle RLU scores obtained before patient use (after cleaning) and after patient use. A one way analysis of variance and Tukey-Kramer HSD

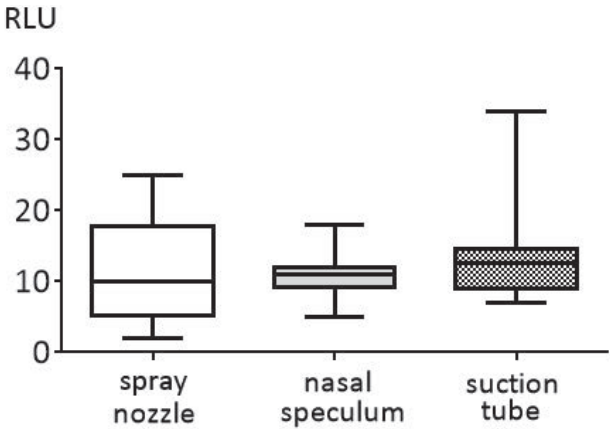

Fig. 3 Comparison of RLU scores between nasal spray nozzles, nasal speculums and suction tubes before use
There were no statistical differences in RLU scores among the nasal spray nozzles, nasal speculums, and suction tubes before patient use.

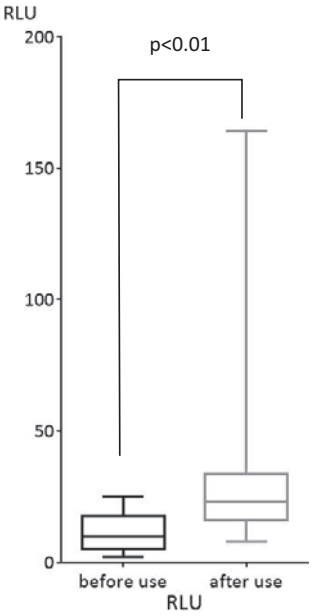

Fig. 4 RLU scores of nasal spray nozzles before and after use
Nozzle RLU scores increased after patient use. The RLU scores (mean ± SD) of the nasal spray nozzles before use was 12 ± 7.1 (highest RLU = 25), increasing to 30 ± 28.7 (highest RLU = 164) after patient use.

tests were used to compare the logarithm of RLU scores among the spray nozzles, nasal speculums, and suction tubes. Statistical significance was set at a p-value of <0.05.

2. Culturing methods

We obtained 10 samples from the surfaces of nasal

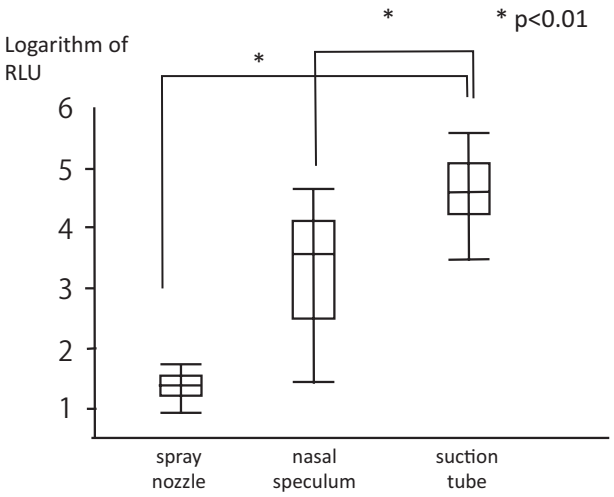

Fig. 5 Comparison of logarithm of RLU scores between nasal spray nozzles, nasal speculums and suction tubes after use
The RLU scores (mean ± SD) after patient use is:
nasal spray nozzles, 30 ± 28.7;
nasal speculums, 9,722 ± 12,398.9;
suction tubes, 88,366 ± 106,839.3.
Increases in RLU scores of spray nozzles were statistically smaller than the increases observed with the nasal speculums and suction tubes.

spray nozzles used with the three patients for culturing using the Transwab™ ENT Amies charcoal MWE. Routine laboratory procedures were used to identify bacterial species on the surface of the nozzles. All growths were considered microbiologically positive, regardless of species or number of colonies forming the unit.

Results

1. ATP bioluminescence test

There were no statistical differences in RLU scores among the nasal spray nozzles, nasal speculums, and suction tubes before patient use (**Fig. 3**). Nozzle RLU scores increased after patient use. The average RLU score (mean ± SD) of the nasal spray nozzles before use was 12 ± 7.1, increasing to 30 ± 28.7 after patient use (**Fig. 4**). The average RLU score of the nasal speculums increased from 11 ± 2.8 to 9,722 ± 12,398.9 after patient use. The average RLU score of the suction tubes increased from 13 ± 5.7 to 88,366 ± 106,839.3 after patient use. Increases in RLU scores of spray nozzles were statistically smaller than the increases observed with the nasal speculums and suction tubes (**Fig. 5**).

2. Culturing methods

Microbiological cultures of all 10 samples were negative for bacterial growth.

Discussion

According to Spaulding[5], medical devices are classified into three categories based on the risk of infection associated with use: critical, semi-critical, and noncritical. Critical devices contact sterile tissue or vasculature and should therefore be sterilized. Semi-critical devices contact intact mucous membranes, but do not penetrate sterile tissue, and therefore should be cleaned with a high-level disinfectant or undergo hot water disinfection. Noncritical devices touch only intact skin or do not touch the patient at all. These devices should be cleaned using a low-level disinfectant.

Nasal spray nozzles are classified as noncritical devices because, in typical practice, they do not touch the skin or mucosa. However, our study demonstrated increased RLU scores following patient use, suggesting contamination originating from the aerosol or accidental touching with patient's mucus, or nasal discharge. RLU scores obtained from the spray nozzles were low compared with those obtained from the nasal speculums and suction tubes, which directly contacted patient skin or mucosa.

In the present study, all samples obtained from the spray nozzles were negative for conventional (swab-based) tests of bacterial growth. However, previous studies reported normal bacterial flora obtained from spray nozzle tips, including CNS, MSSA, and MRSA. Organisms that routinely cause respiratory tract infections such as *Haemophilus influenzae*, *Streptococcus pneumoniae*, or *Pseudomonas aeruginosa* were not detected[1)2)6)].

Nasal spray nozzles should be wiped using a mid-level disinfectant such as ethanol or isopropanol following patient use. Nasal sprays that use disposable tips are also useful.

ATP tests for hygiene monitoring are well-established in food production facilities, with additional applications for monitoring clinical devices and environments[3]. A study assessing hospital kitchen surfaces showed a statistically significant relationship between ATP testing and traditional cultures obtained via microbial swabbing[7]. ATP testing can be completed easily and rapidly compared with traditional swabbing culture methods. However, ATP tests are not a substitute for culturing methods because the ATP test is unable to determine the identities of specific bacterial strains. Furthermore, ATP testing sensitivity varies among substrates. Turner et al.[8] reported that pure bacteria are weakly detected with an limit of detection of 10^4 for representative gram-negative and 10^2 for representative gram-positive bacteria. ATP testing is unable to detect the gram-negative bacteria because of incomplete cell lysis.

ATP tests can determine the general form of surface contamination, including cultivable and non-cultivable microbial and organic contamination. In the present study, ATP testing detected mild contamination on the surface of the nasal spray nozzles. These results would not have been obtained using traditional swabbing cultural methods.

Conclusion

We measured contamination of the surfaces of nasal spray nozzles using ATP bioluminescence testing. After use with three patients, the nasal spray nozzles showed increased RLU scores, indicative of contamination, although RLU scores obtained from nasal spray nozzles were statistically lower than those obtained from nasal specula and suction tubes, both of which directly contact the nasal cavity or nasal discharge. Nasal spray nozzles should be wiped with a mid-level disinfectant after use. Alternatively, the nozzle tips should be changed between patients. These recommendations should be considered when developing protocols for cleaning and disinfection of nasal spray nozzles used in conjunction with ENT treatment units.

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Conflicts of Interest: The authors have no conflicts of in-

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