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Wipes Coated with a Singlet-Oxygen-Producing Photosensitizer Are Effective against Human Influenza Virus but Not against Norovirus

Katharina Verhaelen,[a,b] Martijn Bouwknegt,[a] Saskia Rutjes,[a] Ana Maria de Roda Husman,[a,b] Erwin Duizer[a]

Centre for Infectious Disease Control, National Institute for Public Health and the Environment (RIVM), Bilthoven, The Netherlands[a]; Faculty of Veterinary Medicine, Institute for Risk Assessment Sciences (IRAS), Utrecht University, Utrecht, The Netherlands[b]

Transmission of enteric and respiratory viruses, including human norovirus (hNoV) and human influenza virus, may involve surfaces. In food preparation and health care settings, surfaces are cleaned with wipes; however, wiping may not efficiently reduce contamination or may even spread viruses, increasing a potential public health risk. The virucidal properties of wipes with a singlet-oxygen-generating immobilized photosensitizer (IPS) coating were compared to those of similar but uncoated wipes (non-IPS) and of commonly used viscose wipes. Wipes were spiked with hNoV GI.4 and GII.4, murine norovirus 1 (MNV-1), human adenovirus type 5 (hAdV-5), and influenza virus H1N1 to study viral persistence. We also determined residual and transferred virus proportions on steel carriers after successively wiping a contaminated and an uncontaminated steel carrier. On IPS wipes only, influenza viruses were promptly inactivated with a 5-$\log_{10}$ reduction. *D* values of infectious MNV-1 and hAdV-5 were 8.7 and 7.0 h on IPS wipes, 11.6 and 9.3 h on non-IPS wipes, and 10.2 and 8.2 h on viscose wipes, respectively. Independently of the type of wipe, dry cleaning removed, or drastically reduced, initial spot contamination of hNoV on surfaces. All wipes transferred hNoV to an uncontaminated carrier; however, the risk of continued transmission by reuse of wipes after 6 and 24 h was limited for all viruses. We conclude that cleaning wet spots with dry wipes efficiently reduced spot contamination on surfaces but that cross-contamination with noroviruses by wiping may result in an increased public health risk at high initial virus loads. For influenza virus, IPS wipes present an efficient one-step procedure for cleaning and disinfecting contaminated surfaces.

Viruses are the most common cause of infectious disease acquired in the indoor environment and have considerable impact on human health (1). Transmission of human norovirus (hNoV) and human influenza virus is assumed to occur mostly directly from person to person, followed by indirect transmission through contaminated fomites, hands, and surfaces (2–4). The public health risk resulting from exposure to contaminated surfaces depends on a number of factors, including the level of shedding of infectious particles, their stability on surfaces, and resistance to decontamination procedures (2, 5).

hNoVs are nonenveloped RNA viruses and recognized as the leading cause of epidemics of gastroenteritis worldwide and an important cause of sporadic gastroenteritis in all age groups (6). The virus is transmitted via feces and vomit of infected individuals, and transmission routes are difficult to separate (1, 7). hNoV shows a great potential for surface transmission. The virus is (i) abundantly shed via vomit and feces (8, 9), (ii) persistent in the environment and resistant to disinfectants (5, 10, 11), (iii) transferable between inanimate and animate surfaces (12–16), (iv) highly infectious (17), and (v) constantly evolving into new antigenic types (18, 19). Contamination of hard (steel, glass, or plastic) surfaces with hNoV in different facilities such as food and health care settings has been described previously (2, 3, 20–23), as has been hNoV transmission via surfaces sustaining a succession of outbreaks in closed settings such as airplanes, cruise ships, and hotels (24–27). In food-borne outbreaks in which surfaces played a role in the transmission, initial contamination occurred mostly when kitchen staff vomited in a sink (26, 28). Influenza viruses are an important cause of respiratory disease, frequently resulting in epidemics and occasionally pandemics (29). It was concluded that the predominant route of influenza virus spreading was by airborne transmission, but transfer of infectious influenza virus via frequently touched nonporous surfaces such as door handles and light switches could make contact transmission a key transmission mode (2, 4). This conclusion was based on the findings that influenza virus could remain infectious for several days on these surfaces and that contamination levels could be quite high due to sneezing and coughing (4, 30).

Cleaning and disinfection procedures of wiping fomites with cloths, with or without disinfectants, are applied commonly in food preparation areas and health care settings to minimize the transmission of human pathogens from surfaces to humans and foods. At present, a wide variety of cleaning products, disinfectants, and cloths are on the market. Chlorine is considered the most efficient disinfectant against norovirus contamination (31, 32); however, the mandated chlorine concentration of 200 ppm chlorine is unlikely to prevent transfer of norovirus on surfaces by wiping (12). For influenza virus, chlorine, but also other cleaning agents such as vinegar and quaternary ammonium, is an effective disinfectant (5, 29, 30). Not only the disinfectant used but also the wiping cloth affects decontamination efficacies and cross-contamination by wiping (33–35). Gibson et al. (34) showed the effect of the type of cloth on the removal of hNoV surrogates from surfaces and their transfer to surfaces, and especially, the transfer to surfaces by wiping with contaminated wipes was greatly affected by the type of cloth, with cotton terry bar towels cross-

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Editor: M. V. Yates
Address correspondence to Katharina Verhaelen, verhaelen@gmail.com, or Erwin Duizer, erwin.duizer@rivm.nl.

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contaminating surfaces to the greatest extent and microfiber cloth to the least. The use of wipes coated with antimicrobials may allow combined cleaning and disinfection of surfaces and prevent cross-contamination via the cleaning cloth if the pathogen is inactivated efficiently in the cloth. Cloths that incorporate antimicrobials such as light-activated polymers or copper-based biocides have been shown to be efficient in inactivating pathogens (36–40).

We studied nonwoven wipes with an immobilized photosensitizer that generates singlet oxygen on activation by light for their virucidal capacities. Additionally, their potential to decontaminate stainless steel surfaces and to minimize virus transfer to clean surfaces was assessed. The commonly found hNoV GI.4 and hNoV GII.4 strains were included as examples of their respective genogroups. Murine norovirus 1 (MNV-1), the only cultivable norovirus, was used as a proxy to study hNoV infectivity. Human influenza A virus (H1N1) was studied as an example of a surface-transmittable enveloped respiratory virus. Human adenovirus type 5 (hAdV-5) was included in the virucidal activity testing of the wipes since it is a nonenveloped DNA virus that can be detected in respiratory secretions and in feces and the European Committee for Standardization suggests testing hAdV-5 to assess the efficacy of disinfectants (41). As far as we know, this is the first study on the use of immobilized photosensitizer fabrics as cleaning wipes for surfaces.

MATERIALS AND METHODS

Viruses and cells. hNoV GI.4 (Hu/NoV/GI.4/10001/2009/Netherlands) and hNoV GII.4 (Hu/NoV/GII.4/10496/2010/Netherlands) were obtained from 10% fecal solutions. In addition, cultivatable MNV-1 (Mu/NoV/GV/MNV1/2002/USA), hAdV-5 (reference strain, Hu/adenovirus/type 5/6270/1988/Ethiopia), and influenza A (H1N1) virus (clinical isolate, Hu/influenza A/266/2008/Netherlands [H1N1] virus) were included in the experiments. Virus stocks were obtained by three freeze-thaw cycles for infected cells and subsequent centrifugation at 1,500 × g for 15 min. Cell lines used to propagate viruses are described in reference 42.

Wipes. Both coated and uncoated Serqet wipes were a gift from Laam-Science, Inc. (Morrisville, NC, USA). The wipes were nonwoven and consisted of a blend of mostly cotton, rayon, and bamboo fibers with less than 8% polypropylene fibers. The fibers of coated wipes were coated with the immobilized photosensitizer rose bengal (43, 44) and are referred to as immobilized photosensitizer (IPS) wipes. Rose bengal-styrene monomers were mixed with acrylic acid for polymerization, and the polymers were subsequently covalently bound to the fibers by multiple amide links at a final dye level of 0.04% to 0.08% (wt/wt). The immobilized rose bengal produced singlet oxygen ($^1O_2$) from molecular oxygen (O_2) during exposure to visible light (i.e., photosensitized generation of singlet oxygen). Besides coated IPS and uncoated non-IPS wipes, commonly used viscose wipes (nonwoven cotton) that can be purchased in any household supply store were studied. Viscose wipes were included to compare IPS wipes to commonly used cleaning wipes. Non-IPS wipes were included to study the effect of the IPS on virus inactivation.

Preparation and spiking of wipes. The wipes were cut into 1-cm^2 pieces under sterile conditions in a safety cabinet with the light off and placed in petri dishes wrapped in aluminum foil for storage until use. Wipes were spiked with 50 μl of a mixture of hNoV GII.4 (~10^5 PCR-detectable units [PCRU]), hNoV GI.4 (~10^6 PCRU), and MNV-1 (~3×10^4 infectious particles and 10^6 PCRU) or with 50 μl of influenza A (H1N1) virus (~10^5 infectious particles and 10^6 PCRU) or with 50 μl hAdV-5 virus stock (~10^6 infectious particles and 10^7 PCRU). The spiked virus mix was completely absorbed by the wipes and wetted the previously dry wipes.

Viral persistence on wipes. Experiments were performed in a safety cabinet. The temperature in the cabinet was monitored with a probe every half hour and varied between 25.5°C and 27.5°C. The light intensity inside the cabinet was 1,300 lx. Viruses were spiked on the three types of wipes; exposed to light for 0 min, 5 min, 1 h, 6 h, and 24 h; and afterwards were eluted from the fabrics. Studying viral persistence for up to 24 h was of interest regarding wipe reuse before discarding or washing. To study the effect of singlet oxygen release on virus persistence, IPS wipes were stored in the dark (wrapped in aluminum foil in a petri dish) for 1 h, 6 h, and 24 h to prevent the production of singlet oxygen. Each experimental condition was performed in triplicate. A negative control was included (no virus spiked on the wipes) to control for possible contamination of chemicals and equipment used and to monitor a potentially toxic effect of the eluent on the cells used to determine viral infectivity.

Reduction and transfer of viruses due to wiping. To study virus spread through wiping, a steel carrier, A (2 by 2 cm), was inoculated with 20 μl of the previously used virus suspension containing MNV-1, hNoV GI.4, hNoV GII.4, and influenza A (H1N1) virus. hAdV-5 was not studied in this experiment. Carrier A was then wiped with a dry 1-cm by 1-cm viscose, non-IPS, and IPS wipe in a circular fashion for 10 s with gloved fingertips. Afterwards, a second, uncontaminated steel carrier, B, was wiped in the same manner with the now-damp wipes that were used to clean carrier A. Viruses were eluted from both carriers A and B and from the wipe. As a reference point, viruses were spiked on a steel carrier and subsequently eluted from the carrier without any wiping. Experiments were performed in duplicate.

Virus elution. Viruses were eluted from the wipes in 1.5 ml of Tris base-glycine-beef extract (TGBE) buffer, pH 9.5. Tubes were covered with aluminum foil to prevent further viral inactivation by the treated wipes. Tubes were rotated for 1 h at 4°C. The elution buffer was transferred to a new tube, the pH was adjusted to 7, and the sample was split into two portions: one was used to determine viral infectivity and the other was used for molecular detection. Samples were stored at −80°C before usage. To elute viruses from the steel carriers, the carriers were transferred to 6-well plates and covered with 3 ml of TGBE, pH 9.5; they were then shaken at 100 rpm at 4°C for 1 h. The liquid was recovered from the wells, the pH was adjusted to 7, and the samples were stored at −80°C in aliquots.

Virus enumeration. Viruses were enumerated by endpoint titration in 96-well plates (42, 45). Cells were seeded in a concentration of 2×10^5 cells/ml, and 10-fold serial dilutions in fetal calf serum (FCS)-free medium were prepared (eight dilutions, 10 replicates with each), followed by inoculation in a 96-well plate. hAdV-5 virus dilutions were added to 100 μl of freshly trypsinized HEp-2 cells in medium containing 10% FCS. MNV-1 was replicated on RAW 264.7 cells that were previously incubated for at least 3 h in culture medium containing 2% FCS. We titrated influenza A (H1N1) virus on 3-day-old MDCK-1 cell monolayers after washing twice with phosphate buffer solution prior to infection. The influenza A (H1N1) virus suspensions were prepared in Dulbecco's modified Eagle's medium (DMEM) with 2.5 μg/ml TPCK [1-(tosylamido-2-phenyl)ethyl chloromethyl ketone]-treated trypsin. A cytopathic effect (CPE) on the different cells was observed after 6 to 7 days of incubation at 37°C in 5% carbon dioxide, whereupon HEp-2 cells and MDCK-1 cells were stained with crystal violet to confirm CPE.

A negative control (inoculation of wells with only dilution medium and no virus) was included on each plate. Furthermore, we tested for toxic effects of the negative sample matrix (eluent of each of the three different wipes that were spiked with water) on the cells to be able to distinguish between CPE and cell death by sample toxicity. We also analyzed whether the negative sample matrix affected the detection of virus infectivity, for example, by modifying the virus cell receptor, which would not allow replication of infectious virus particles but would not necessarily affect the cell monolayer. To accomplish this, we inoculated the cells with 100 μl of the negative sample matrix and, after an incubation of 2 h, added 100 μl of

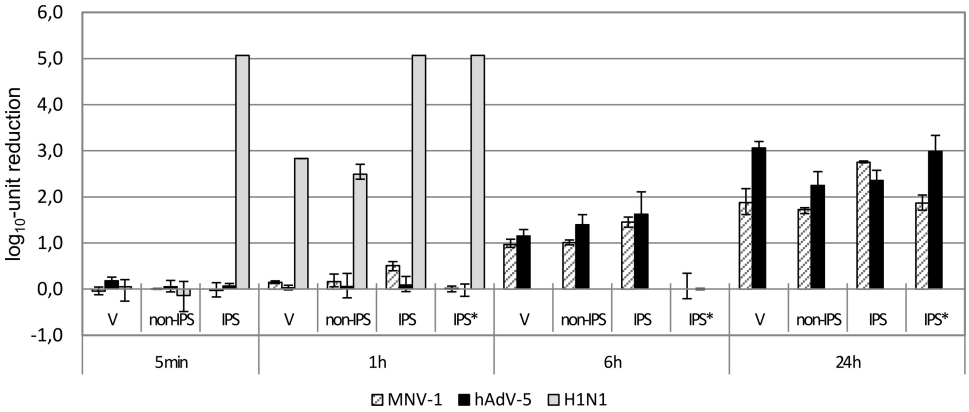

FIG 1 Average reductions of infectious MNV-1, hAdV-5, and H1N1 virus particles on viscose wipes (V), non-IPS wipes (non-IPS), IPS wipes (IPS), and IPS wipes stored in the dark (IPS*). The error bars represent the minimum and maximum $\log_{10}$ reduction values of the tested condition. Because rapid and complete decay of infectious H1N1 viruses was observed on IPS wipes, persistence of this virus was not determined after 6 and 24 h. The letter "a" indicates that the maximum inactivation of infectious influenza virus is represented.

stock virus dilution to the cells; we then compared the infectious virus titer of those cells to that of cells incubated solely with dilution medium.

Molecular detection. Nucleic acids were extracted with the NucliSens miniMag magnetic extraction kit (bioMérieux, Zaltbommel, The Netherlands), according to the manufacturer's instructions with minor modifications (46). To monitor the extraction efficacy, samples were spiked with mengovirus (vMC_0) before extraction. In each extraction cycle, a negative control without a target was included. hAdV-5 and MNV-1 were detected as described by Verhaelen et al. (45), and hNoV GI, hNoV GII, and influenza virus were detected as described by Tuladhar et al. (47) and Svraka et al. (48). Inhibition controls were included in all PCRs to monitor PCR inhibition (42, 45, 48). The fragment length analyzed was short and varied from 95 to 149 nucleotides. PCRU of the virus stocks were determined, by the slope of standard curves, with the highest dilution resulting in a positive PCR signal being equal to a value of 1 PCRU (47).

Data analysis. Infectivity counts of MNV-1, hAdV-5, and influenza H1N1 virus were estimated by maximum likelihood from the presence-absence profile of the endpoint dilutions, assuming homogeneous (Poisson) mixing (45). The $\log_{10}$-unit reductions of infectious virus particles were calculated on the basis of the virus count at time t relative to the count at $t = 0$ (viruses eluted from wipes immediately after spiking). The reductions of viral genomes were calculated on the basis of the difference in quantification cycles (C_q values) and not on absolute genome numbers. Therefore, C_q values were determined at time t and $t = 0$, and a difference in C_q value of log(10)/log(2) (equivalent to a difference of 3.3 C_q values) was assumed equal to a 1-$\log_{10}$-unit reduction of viral genomes. The statistical significance of differences in reduction between treated and untreated IPS wipes and among viruses was determined with the likelihood ratio test using a general linear model with infectious virus counts or C_q values as response variable and treatment and virus as explanatory variables ($\alpha = 0.05$).

The residual viral contamination after wiping A carriers was expressed as the proportion of viruses recovered from the wiped carriers and the viruses recovered from the reference carrier (spiked unwiped carrier). The transferred viral contamination was expressed as the proportion of viruses recovered from the B carriers and the viruses recovered from the reference carrier (spiked unwiped carrier), assuming similar recoveries. To express the proportions of residual viral genomes on A carriers and transferred viral genomes on B carriers, the $\log_{10}$-unit differences between wiped and reference carriers were determined in the same way and subsequently translated into percentages.

RESULTS

We analyzed the persistence of hNoV GI.4 and GII.4, MNV-1, hAdV-5, and H1N1 virus on viscose, non-IPS, and IPS wipes and determined furthermore the residual and transferred virus proportions on and to steel carriers after successive wiping.

Controls. The efficiency of viral genome extraction from wipes was monitored using vMC_0. The extraction efficiency was found to be constant for all wipes, with a maximum difference of 1 C_q value between the samples. The yields of infectious viruses varied from 10 to 100% for MNV-1, influenza H1N1 virus, and hAdV-5. No toxic effect of the sample matrix of the wipe eluates on the cells was detected, and the virus titer was not affected by the sample matrix (data not shown). The internal and external amplification controls showed that the sample matrix did not inhibit the amplification of the PCRs.

Viral persistence on viscose wipes, non-IPS wipes, and IPS wipes. Both the type of wipe ($P \leq 0.002$) and the studied virus ($P < 0.001$) influenced the persistence of the infectious viruses significantly. Enveloped influenza viruses were least persistent, followed by hAdV-5 and MNV-1 (Fig. 1). The IPS coating resulted in a rapid reduction of influenza virus infectivity. After immediate elution of influenza virus from the IPS wipes ($t = 0$), no infectious viruses could be recovered, which relates to a $\geq$4-$\log_{10}$-unit reduction of infectious influenza virus for a recovery of $\geq$10% (estimated recovery of influenza viruses from non-IPS wipes) and a 5-$\log_{10}$-unit reduction assuming complete virus inactivation. Consequent to the immediate influenza virus decay on IPS wipes, after 1 h no infectious influenza viruses could be recovered from IPS wipes stored under dark conditions (Fig. 1). On viscose and non-IPS wipes, no rapid inactivation of influenza virus infectivity was observed, but a reduction of over 2 $\log_{10}$ units was found after 1 h (Fig. 1).

Inactivation of infectious nonenveloped viruses was not rapid on any wipe, with mean D values of 7 to 9 h and of 9 to 10 h for infectious MNV-1 and hAdV-5 particles, respectively (Table 1). Infectious MNV-1 and hAdV-5 were marginally but significantly less persistent on IPS wipes than on non-IPS wipes ($P < 0.001$), and the difference in viral persistence on viscose and non-IPS wipes was also small but significant ($P = 0.002$) (Fig. 1; Table 1). After storage for 6 h, wipes of the dark control were still wet, whereas the wipes stored in light were completely dried.

Concomitantly, MNV-1 and hAdV-5 were more persistent on IPS wipes under dark conditions than under light conditions, with

TABLE 1 Mean *D* values of infectious influenza virus, MNV-1, and hAdV-5 particles on the three tested wipes and their 95% confidence intervals

Virus	*D* value,[b] h (95% confidence interval), on wipe:		
	Viscose	Non-IPS	IPS
Influenza H1N1 virus	0.6 (0.55–0.67)	0.71 (0.6–0.87)	≤0.02[a]
MNV-1	10.2 (8.7–12.2)	11.6 (9.0–15.1)	8.7 (6.8–11.2)
hAdV-5	8.2 (7.6–8.8)	9.3 (7.9–10.9)	7.0 (6.0–8.1)

[a] Because no infectious influenza viruses could be detected at time zero, an exact *D* value could not be calculated.
[b] The *D* values (time needed for the first 1-$\log_{10}$-unit reduction) were calculated on the basis of the linear regression model.

a difference of about 2 $\log_{10}$ units (Fig. 1). After 24 h of storage, wipes stored under light and dark conditions were dried.

For MNV-1, decay of nucleic acids and that of infectious particles were comparable (Fig. 2). For influenza virus and hAdV-5, decay of viral nucleic acids was less pronounced but followed trends similar to those of infectious particles (Fig. 2). As was the case for infectivity, genome persistence of the tested viruses was significantly influenced by the type of wipe ($P = 0.002$) and virus ($P < 0.0001$). After 1 h, the reduction of influenza virus genomes on IPS wipes relative to IPS wipes stored in the dark was 1 $\log_{10}$ unit higher. After 6 and 24 h, the hAdV-5 DNA genomes were consistently more persistent than were the RNA NoV genomes. The effect of any wipe on viral genome persistence was not statistically different among MNV-1, hNoV GI.4, and hNoV GII.4 ($P = 0.32$).

Residual and transferred contamination on steel carriers after wiping. No residual contamination of infectious MNV-1 and influenza virus particles could be recovered from the spiked steel carriers after wiping. Residual contamination was determined only for norovirus genomes, with similar residual virus proportions of 0.2 to 0.6% for the three wipes and both genotypes (Fig. 3). No transferred infectious influenza virus particles could be recovered from the subsequently wiped B steel carriers. Transferred contamination proportions of infectious MNV-1 particles and hNoV genomes to B steel carriers were similar and ranged, like the residual virus proportions, between 0.2% and 0.6%,

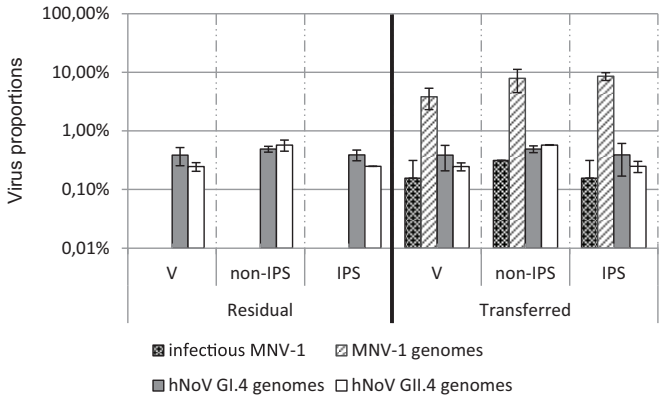

FIG 3 Residual and transferred virus proportions of hNoV genomes and infectious MNV-1 particles and genomes after wiping sequentially with viscose wipes (V), non-IPS wipes (non-IPS), and IPS wipes (IPS). The error bars represent the minimum and maximum residual and transferred virus proportions.

whereas transferred MNV-1 genome proportions were higher (Fig. 3). The three different wipes had little impact on the transfer of hNoV GI.4 and GII.4 genomes to steel carriers. The presence or absence of viruses was also determined on the used wipes. Infectious MNV-1 and hNoV GI and GII genomes were detected on all wipes, whereas infectious influenza virus was detected only on non-IPS wipes and viscose wipes, not on IPS wipes.

DISCUSSION

Efficient cleaning and disinfection procedures are needed to reduce the infectious load of pathogenic viruses on surfaces and interrupt virus transmission. The actual efficiency of the cleaning and disinfection procedures in practice depends on several factors, including the cleaning method, disinfectant, and cloth used. Wiping with or without disinfectant has been shown to reduce the residual contamination of hard surfaces significantly but not always to a level low enough to exclude continued transmission (5, 12). Additionally, wiping may result in a more extensive spread of viruses on surfaces by transferring pathogens from a highly contaminated spot to a larger surface or a secondary surface. This is

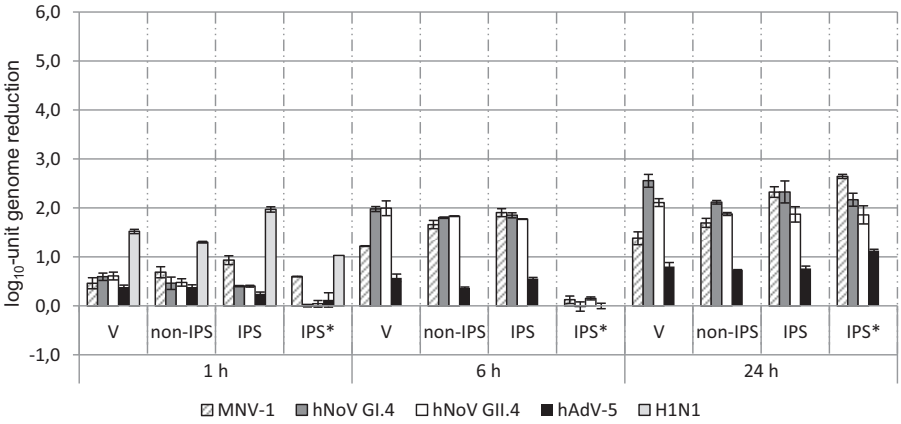

FIG 2 Average genomic copy reductions of MNV-1, hNoV GI.4, hNoV GII.4, hAdV-5, and influenza H1N1 virus on viscose wipes (V), non-IPS wipes (non-IPS), IPS wipes (IPS), and IPS wipes stored in the dark (IPS*). The error bars represent the minimum and maximum $\log_{10}$ unit reduction values of the tested condition. Influenza virus genomes were analyzed for only up to an hour because of the observed rapid decay of infectious influenza virus on IPS wipes. No decay of viral genomes was observed after 5 min.

especially problematic for highly persistent and highly infectious pathogens such as hNoV. Both the efficiency of the removal of infectious viruses by wiping of contaminated surfaces and the rapid viral inactivation on the wipes, and/or irreversible binding of viruses to the wipes, are relevant for the overall efficiency of cleaning procedures to prevent spread of viruses via surfaces.

Wiping is a short-duration activity, and to have a significant effect in decontamination or transfer prevention, the virucidal activity should be very fast, i.e., within seconds. Neither the viscose wipes nor the IPS wipes caused rapid inactivation of infectious MNV-1 or rapid reduction of hNoV GI.4 and GII.4 genomes. IPS wipes are, therefore, not likely to prevent the spread of noroviruses better than viscose wipes. Nevertheless, residual norovirus contamination on spiked steel carriers after wiping with any of the tested wipes was little, and no infectious MNV-1 and only a small fraction of human norovirus genomes were recovered. In food preparation and health care settings, it is recommended to clean surfaces subjected to liquid norovirus soiling by, for example, vomit or fecal contamination with dry absorbent material such as paper towels or wipes (49, 50). Initial virus removal from surfaces by cleaning with dry wipes after spot contamination with 10^4 noroviruses was found to be efficient.

We did show that infectious MNV-1 particles and hNoV genomes were transferred from one steel carrier to another by reuse of wipes, indicating that viruses are not irreversibly bound to the wipes. The transferred norovirus genome proportions are therefore likely to represent infectious viruses, given the short time interval of wiping and the data on persistence of infectious MNV-1 on wipes. Noroviruses are thus possibly spread to surfaces by continued use or reuse of wipes. Cross-contamination may be prevented by discarding or by laundering (depending on the initial virus load) (51, 52) wipes directly after contamination. The use of chlorine in preventing cross-contamination of hNoV on surfaces by wiping is not well investigated, and a study by Barker and coworkers (12) showed that even the use of high chlorine concentrations was not always able to prevent transfer of hNoV RNA to secondary surfaces by wiping, especially where there was fecal soiling. However, research showed that detectability of RNA (fragments) is not necessarily a good measure of infectivity (47, 53).

Whether the viral loads on surfaces transferred by cross-contamination lead to an enhanced health risk through continued transmission of the virus depends on (i) the initial virus load on the surface, (ii) the virus proportion transferred to secondary surfaces, and (iii) the proportion of viral transfer from the secondary surface to a human host. We found that about 0.2% of the initial norovirus contamination was transferred to a clean spot through cross-contamination by wiping. Assuming a target level of residual contamination of 50 noroviruses per spot, a level resulting in minimal risk of continuous virus transmission or infection (5), we can deduce that the contamination of the initial spot should be above 2.5×10^4 infectious viruses per spot to pose a risk. Considering that spots of about 1 mg of fecal contamination are just barely visible and are thus realistic fecal contaminations, shedding of over 2.5×10^7 hNoV particles per gram of feces is required. Projectile vomiting in the acute disease phase may add as well to the spread of the virus (54), with a release of 10^4 genomic norovirus copies/ml and a total excreted volume of up to 1.7 liters (55). This indicates that cross-contaminated surfaces may pose a risk when the initial contamination occurred during a period of about 10 days around the peak of viral shedding (9, 56) or after an episode of vomiting. We may thus conclude that cross-contamination through wiping of surfaces could pose a public health risk and should be considered in implementing transmission intervention measures in food production and health care facilities.

In contrast to noroviruses, infectious influenza viruses were found to be inactivated promptly by 4 to 5 $\log_{10}$ units by IPS wipes but not by regular viscose wipes and non-IPS wipes. Unlike noroviruses and adenoviruses, influenza viruses have a lipoprotein envelope, which makes them less tolerant to disinfectants than nonenveloped viruses, as has been shown for several disinfectants (5, 30, 57). In a study on the virucidal activity of rose bengal on influenza viruses, it was proposed that singlet oxygen had a direct effect on the fusion protein of the virus after enrichment of the photosensitizer rose bengal in the viral envelope (58). In our study, the rose bengal was immobilized by covalent binding to the fibers of the wipes, thus not allowing for enrichment of the photosensitizer in the viral envelope. The prompt inactivation of the influenza viruses on IPS wipes suggests the relevance of singlet oxygen in the pathogen inactivation, as was shown before for other enveloped viruses (58, 59) and for bacteria such as *Escherichia coli* (60). Yet, we could not show that the efficient inactivation of influenza virus was due to released singlet oxygen or to other mechanisms (e.g., antiviral effect of the coating), as complete viral decay on IPS wipes was immediate ($t = 0$) and therefore also no infectious influenza viruses could be detected on wipes stored in the dark. However, the design of our study was aimed not at elucidating the exact mechanism of the very efficient influenza virus inactivation but at determining the overall effect on (infectious) virus reduction on wipes and surfaces, including the transfer of viruses by different cloths. On the basis of our study, wiping with IPS wipes is likely to be an efficient intervention method for surface-related influenza virus transmission, because infectious viruses are efficiently removed and inactivated, thus preventing transmission and cross-contamination. Similar effects of IPS wipes may be expected for other enveloped viruses (58).

Wipes for cleaning kitchen or bathroom surfaces and in food preparation settings such as restaurants, but also in private households, may not be instantly discarded as is prescribed for health care settings, and thus, they may contribute to viral spread over an extended period of time. Therefore, we analyzed the persistence of viruses on wipes after 6 h and 24 h and observed substantial reductions of between 2 and 3.5 $\log_{10}$ units of infectious MNV-1 and hAdV-5 viruses on all wipes. IPS wipes showed a 1-$\log_{10}$-unit-higher reduction of infectious MNV-1 particles after 24 h than did viscose wipes, indicating an increased potential of these wipes to contain norovirus spread by reused wipes compared to that of viscose wipes. Instead of discarding the wipe, drying IPS wipes for a day may thus prevent potential viral cross-contamination by both enveloped and nonenveloped viruses, which is especially of interest in countries with restricted resources. An important factor of viral decay after prolonged times on wipes may be desiccation.

In summary, we conclude that cleaning wet spots with dry wipes efficiently reduced spot contamination of the viruses on surfaces. Yet, IPS wipes, as well as the other wipes that we used, were not able to prevent surface cross-contamination of the nonenveloped viruses by wiping, which may result in an increased public health risk at high initial virus loads. However, the IPS wipes tested present a one-step procedure for cleaning and disinfecting influenza virus-contaminated surfaces. In addition, we

show that the risk for continued virus transmission by reuse of viscose or IPS wipes is limited if wipes are allowed to dry before reuse.

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