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Assessment of Healthcare Worker Protocol Deviations and Self-Contamination During Personal Protective Equipment Donning and Doffing

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OBJECTIVE. To evaluate healthcare worker (HCW) risk of self-contamination when donning and doffing personal protective equipment (PPE) using fluorescence and MS2 bacteriophage.

DESIGN. Prospective pilot study.

SETTING. Tertiary-care hospital.

PARTICIPANTS. A total of 36 HCWs were included in this study: 18 donned/doffed contact precaution (CP) PPE and 18 donned/doffed Ebola virus disease (EVD) PPE.

INTERVENTIONS. HCWs donned PPE according to standard protocols. Fluorescent liquid and MS2 bacteriophage were applied to HCWs. HCWs then doffed their PPE. After doffing, HCWs were scanned for fluorescence and swabbed for MS2. MS2 detection was performed using reverse transcriptase PCR. The donning and doffing processes were videotaped, and protocol deviations were recorded.

RESULTS. Overall, 27% of EVD PPE HCWs and 50% of CP PPE HCWs made ≥1 protocol deviation while donning, and 100% of EVD PPE HCWs and 67% of CP PPE HCWs made ≥1 protocol deviation while doffing (P = .02). The median number of doffing protocol deviations among EVD PPE HCWs was 4, versus 1 among CP PPE HCWs. Also, 15 EVD PPE protocol deviations were committed by doffing assistants and/or trained observers. Fluorescence was detected on 8 EVD PPE HCWs (44%) and 5 CP PPE HCWs (28%), most commonly on hands. MS2 was recovered from 2 EVD PPE HCWs (11%) and 3 CP PPE HCWs (17%).

CONCLUSIONS. Protocol deviations were common during both EVD and CP PPE doffing, and some deviations during EVD PPE doffing were committed by the HCW doffing assistant and/or the trained observer. Self-contamination was common. PPE donning/doffing are complex and deserve additional study.

Infect Control Hosp Epidemiol 2017;38:1077–1083

Personal protective equipment (PPE) is used in healthcare settings to protect healthcare workers (HCWs) from exposure to pathogens and to prevent the spread of pathogens to other patients. Proper use of PPE is crucial when HCWs care for patients with highly pathogenic organisms, such as the Ebola virus. To date, studies on PPE effectiveness are uncommon, small, and potentially out of date, and many evaluate PPE types no longer in use.1 The 2014–2016 Ebola virus disease (EVD) outbreak revealed the need for better empirical data regarding best practices to safely don and doff PPE.1–5

Although EVD is a high-visibility, high-impact disease, HCWS are much more likely to encounter pathogens on a daily basis, including methicillin-resistant *Staphylococcus aureus* (MRSA), vancomycin-resistant *Enterococcus*, *Clostridium difficile*, and carbapenem-resistant *Enterobacteriaceae*. The primary forms of PPE used to protect HCWs and other patients against these important hospital-associated pathogens are associated with contact precautions (CP), which includes gowns and gloves. Few data exist regarding whether HCWs follow guidelines for donning and doffing CP PPE and their risk of self-contamination.6,7

One of the primary challenges when designing studies to evaluate PPE or donning/doffing procedures is determining how to model pathogen transmission. The most commonly used surrogate marker for the presence of pathogens is fluorescence, and it can be delivered in a variety of forms: powder,
Fluorescent markers are inexpensive, easy to use, and because the read out is visual, they can provide immediate feedback to HCWs; however, fluorescence may not be an appropriate surrogate for contamination with infectious viral particles. An alternate marker for viral infection is the MS2 bacteriophage, a single-strand RNA bacteriophage that is a biosafety level 1 agent and nonpathogenic to humans. MS2 has been used previously in several studies of PPE transmission and/or disinfection in a long-term-care facility, a hotel, and an office. Commercial preparations of MS2 are expensive and require significant laboratory expertise to use, but they may provide a more accurate surrogate marker for how pathogens spread in the environment than fluorescence.

The purpose of this study was to evaluate HCW risk of self-contamination when donning and doffing EVD PPE and CP PPE using MS2 bacteriophage and a fluorescent marker as surrogates for pathogen transmission. The frequencies and types of protocol deviations that occurred were documented, and associations between HCW self-contamination after doffing and particular doffing protocol deviations or HCW characteristics were determined.

METHODS

This prospective pilot study was performed at Barnes-Jewish Hospital (BJH), a 1,250-bed, tertiary-care hospital in St Louis, Missouri. The study was approved by the Washington University Human Research Protection Office, and all participants provided written, informed consent. During the study period, EVD PPE consisted of inner and outer gloves (Esteem XP, Cardinal Health, Dublin OH), boot covers (Convertors FullGuard High Top Shoe Covers, Cardinal Health), impermeable gown with Velcro on the back of the neck (Convertors SmartSleeve, Cardinal Health), a powered air-purifying respirator (PAPR) and hood with face shield (Versaflx, 3M, Maplewood, MN), and an outer apron (Tyvek apron, Uline, Pleasant Prairie, WI). CP PPE consisted of gloves and a gown (Cardinal Health).

HCW Characteristics

Two sets of HCWs were enrolled in this study. EVD PPE HCWs were enrolled during EVD PPE practice sessions and included respiratory therapists, nurses, infection control preventionists, and critical care physicians. CP PPE HCWs were recruited from BJH hospital wards during their normal shifts and included nurses, patient care technicians, and physicians. HCWs were interviewed regarding demographics and years of service, previous PPE training; HCW height and weight were measured and body mass index (BMI) was calculated.

PPE Donning and Doffing and Contamination Procedures

During EVD PPE training sessions, the donning and doffing processes were aided by a donning/doffing assistant and a trained observer who instructed HCWs step-by-step as per CDC guidelines. HCWs using CP PPE were not given donning or doffing instructions; they were encouraged to proceed according to their usual practices.

After consent, participants were scanned for baseline fluorescence using an UV-A light. Any areas of fluorescence detected were cleaned and noted. Next, HCWs were instructed to don the PPE. Upon completion, HCWs were instructed to close their eyes, and the MS2 bacteriophage and fluorescent marker were applied to HCW palms, abdomen, and ankles (for EVD PPE HCWs) or palms and abdomens (for CP PPE HCWs). Dummy applications of molecular grade water were applied to HCW shoulders. After donning, EVD PPE HCWs practiced various EVD patient care activities before doffing. CP PPE HCWs proceeded directly to doffing. The order and technique used to don and doff the PPE were videotaped and recorded. Immediately after doffing, the participant was scanned for fluorescence. Any areas of fluorescence detected were photographed and sampled utilizing a flocked swab in universal transport medium (Quidel, San Diego, CA). HCW hands (1 swab for both hands), coat sleeves or wrist, and peri-orbital/nasal/oral areas were swabbed regardless of fluorescence.

Doffing and doffing videos were reviewed and protocol deviations were recorded. A second reviewer randomly reviewed selected videos to ensure accuracy. Protocol deviations were grouped into categories based on site and the donning/doffing procedural step during which they occurred (ie, glove removal and hand hygiene; PAPR and hood removal). Proper CP PPE and EVD PPE removal sequence were based on recommendations from the CDC and written protocols used by the BJH infection prevention team.

A commercially available preparation of MS2 (Zeptometrix, Buffalo, NY), supplied as a stock solution of $1.0 \times 10^9$ PFU/mL, was utilized as a surrogate for viral transmission. This substance was diluted to a 1:10 solution in viral transport medium for a working solution of $1.0 \times 10^8$ PFU/mL. GloGerm Mist liquid was selected as the fluorescent marker (GloGerm, Moab, UT). A mixture of 100 µL GloGerm Mist liquid with 0.5 mL working solution MS2 was applied to each contamination site. This combination was tested, and there was no negative effect on MS2 recovery and detection. The mixture of GloGerm liquid and MS2 was drawn into a 3-mL syringe with a needleless, Luer-lock tip (Becton Dickinson, Franklin Lakes, NJ). The syringe was attached to a pediatric intranasal laryngeal mask airway mucosal atomization device (LMA MAD Nasal, Teleflex, Westmeath, Ireland). Syringes were not reused.

MS2 RNA was extracted utilizing a QIAamp viral RNA mini kit (Qiagen, Valencia, CA). MS2 detection was performed using reverse-transcriptase polymerase chain reaction (PCR) using previously described primers and the Cepheid Smart Cycler with QuantiTect Probe RT-PCR Kit (Qiagen, Valencia, CA). A positive control with MS2 RNA, and a negative control of PCR water was included in each run. The cycle threshold for all positive results was recorded.
Statistical Analyses

The primary outcomes of interest were the presence and frequency of MS2 and/or fluorescent contamination on the HCW after removal of PPE. The secondary endpoints were the correlations of the presence of contamination with the number of lapses in PPE donning techniques, years of experience, type of PPE, and BMI. Univariate analyses were performed, and $P \leq .05$ was considered significant. We used $\chi^2$ or univariate logistic regression for categorical variables, and we used the Mann-Whitney test for continuous variables. Analyses were performed with SPSS version 24 (IBM, Armonk, NY).

RESULTS

In total, 36 HCWs were enrolled in the study: 18 with EVD PPE and 18 with CP PPE. Most HCWs were nurses: 78% of EVD PPE HCWs and 61% of CP PPE HCWs (Table 1). EVD PPE HCWs were significantly older than CP PPE HCWs (median age, 38 vs 28.5 years; $P = .02$), and there was a trend toward greater years of service among the EVD PPE HCWs (median years of service, 8.5 vs 5.25; $P = .10$).

Donning and Doffing Protocol Deviations

Donning videos were available for review for 15 EVD PPE HCWs (Table 2). Donning videos for the remaining 3 HCWs were unavailable because the HCW was donning simultaneously while another HCW was being recorded. Overall, 27% of EVD PPE HCWs made at least 1 donning protocol deviation, compared with 50% of CP PPE HCWs ($P = .28$). Protocol deviations occurred most often in the gloves and hand-hygiene steps (20% of EVD PPE HCWs and 33% of CP PPE HCWs). All EVD PPE HCWs had at least 1 doffing protocol deviation, versus 67% of CP PPE HCWs ($P = .02$) (Table 2).

The median number of doffing protocol deviations was greater among EVD PPE HCWs (median, 4 vs 1 among CP PPE HCWs). Moreover, 15 protocol deviations during EVD PPE doffing were committed by the doffing assistant or trained observer: 6 during gown or apron removal, 2 involving hand hygiene, 2 during hood removal, 2 during boot-cover removal, 1 during PAPR removal, and 2 miscellaneous deviations. Among EVD PPE HCWs, the unique doffing step with the greatest number of protocol deviations was boot-cover removal: 78% of HCWs made at least 1 protocol deviation during EVD PPE doffing boot covers. The doffing step category with the greatest number of HCWs that committed at least 1 protocol deviation (in both PPE types) was gowned/apron removal (83% of EVD PPE HCWs; 50% of CP PPE HCWs), followed by glove removal/hand hygiene (67% of EVD PPE HCWs; 39% of CP PPE HCWs).

MS2 and Fluorescence

Overall, fluorescence was detected on 8 EVD PPE HCWs (44%) and 5 CP PPE HCWs (28%) ($P = .49$), and 21 unique HCW sites fluoresced. The most common site of fluorescence was HCW hands: 6 among EVD PPE HCWs and 5 among CP PPE HCWs (Table 3). Of the 125 samples tested for MS2, 5 were positive (4%). MS2 was recovered from 2 EVD PPE HCWs (11%) and 3 CP PPE HCWs (17%). The 2 EVD PPE HCW sites from which MS2 was recovered were from an alcohol foam pump in the doffing area and an HCW’s hands (Table 3). The 3 CP PPE HCW sites from which MS2 was recovered were from the face of 1 HCW and from the sleeves/wrist of 2 HCWs. Among the 5 sites positive for MS2, 2 (40%) also fluoresced. The association between fluorescence and doffing protocol deviations is given in Table 4. There were no significant differences in detection of any fluorescence by protocol deviation type, although there was a trend toward...
TABLE 2. Donning and Doffing Protocol Deviations by Personal Protective Equipment (PPE) Type

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Ebola PPE HCW (n = 18)</th>
<th>Contact precautions PPE HCW (n = 18)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Donning</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Any</td>
<td>4 (27)</td>
<td>9 (50)</td>
</tr>
<tr>
<td>Gloves/Hand hygiene</td>
<td>3 (20)</td>
<td>6 (33)</td>
</tr>
<tr>
<td>Gown/Apron</td>
<td>3 (20)</td>
<td>4 (22)</td>
</tr>
<tr>
<td>PAPR/Hood</td>
<td>2 (13)</td>
<td>N/A</td>
</tr>
<tr>
<td>Other</td>
<td>1 (7)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>No. of protocol deviations, median (range)</td>
<td>0 (0–4)</td>
<td>0.5 (0–2)</td>
</tr>
<tr>
<td>Doffing</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Any</td>
<td>18 (100)</td>
<td>12 (67)</td>
</tr>
<tr>
<td>Boot-cover removal</td>
<td>14 (78)</td>
<td>N/A</td>
</tr>
<tr>
<td>Gloves/Hand hygiene</td>
<td>12 (67)</td>
<td>7 (39)</td>
</tr>
<tr>
<td>Gown/Apron</td>
<td>15 (83)</td>
<td>9 (50)</td>
</tr>
<tr>
<td>PAPR/Hood</td>
<td>7 (39)</td>
<td>N/A</td>
</tr>
<tr>
<td>Shoe disinfection</td>
<td>8 (44)</td>
<td>N/A</td>
</tr>
<tr>
<td>Other</td>
<td>2 (11)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>No. of protocol deviations, median (range)</td>
<td>4 (2–8)</td>
<td>1 (0–2)</td>
</tr>
</tbody>
</table>

NOTE. HCW, healthcare worker; N/A, not applicable; PAPR, powered air-purifying respirator.

a Unless otherwise specified.

b n = 18 and n = 15 for donning; donning videos were available for 15 of 18 EVD PPE HCWs.

c18 among EVD PPE HCWs.

P = .28.

dP = .02.

TABLE 3. Sites of Fluorescence and/or MS2 (N = 24) by PPE Type

<table>
<thead>
<tr>
<th>Site of Fluorescence and/or MS2</th>
<th>EVD PPE No. of Detections</th>
<th>Contact Precautions PPE No. of Detections</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hands</td>
<td>7b</td>
<td>5</td>
</tr>
<tr>
<td>Alcohol foam pump</td>
<td>2c</td>
<td>0</td>
</tr>
<tr>
<td>Chest</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>Forearm</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Knee</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Sleeves/Wrist</td>
<td>1</td>
<td>2d</td>
</tr>
<tr>
<td>Thigh</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>Face</td>
<td>0</td>
<td>1</td>
</tr>
</tbody>
</table>

NOTE. PPE, personal protective equipment; EVD, Ebola virus disease; HCW, healthcare worker.

a6 HCWs had >1 site of fluorescence (none had >1 site of MS2).
b6 were fluorescent; 1 was MS2 positive.
c1 fluorescent only; 1 fluorescent and MS2 positive.
d4MS2 positive only.
eFluorescent and MS2 positive.

significance with boot-cover removal (100% of EVD PPE HCWs with fluorescence detected had a boot cover protocol deviation, versus 60% of EVD PPE HCWs without fluorescence; P = .09).

TABLE 4. Fluorescence and Doffing Protocol Deviations

<table>
<thead>
<tr>
<th>Doffing Protocol Deviation</th>
<th>Any Fluorescence Detected</th>
<th>No Fluorescence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Any</td>
<td>12 (92)</td>
<td>18 (78)</td>
</tr>
<tr>
<td>Boot-cover removal</td>
<td>8 (100)</td>
<td>6 (60)</td>
</tr>
<tr>
<td>Gloves/Hand hygiene</td>
<td>8 (62)</td>
<td>11 (48)</td>
</tr>
<tr>
<td>Gown/Apron</td>
<td>10 (77)</td>
<td>14 (61)</td>
</tr>
<tr>
<td>PAPR/Hood</td>
<td>3 (38)</td>
<td>4 (40)</td>
</tr>
<tr>
<td>Shoe disinfection</td>
<td>5 (63)</td>
<td>3 (30)</td>
</tr>
<tr>
<td>Other</td>
<td>0 (0)</td>
<td>2 (9)</td>
</tr>
</tbody>
</table>

a n = 13; n = 8 among EVD PPE HCWs.
b n = 25; n = 10 among EVD PPE HCWs.
c Among EVD PPE HCWs only (n = 18).

HCW Characteristics and Donning/Doffing Protocol Deviations

Among EVD PPE HCWs, there was no significant difference in the median number of donning or doffing protocol deviations by years of service (data not shown). There were no significant differences in fluorescence and/or MS2 detection between BMI categories (ie, normal, overweight, or obese; data not shown). There also were no significant differences in the frequencies of types of donning or doffing protocol deviations by BMI (data not shown).

DISCUSSION

Proper use of PPE is essential to protecting patients and HCWs from infectious diseases. However, our results indicate that protocol deviations were common in both donning and doffing. Notably, we found that 100% of EVD PPE HCWs committed at least 1 protocol deviation during doffing, and 27% while donning. This finding is not surprising, given the complexity of EVD PPE, and it is consistent with previous studies.8,14,22 In a study involving 120 students, Casalino et al22 found that EVD PPE doffing errors occurred even after a 3-phase training program. While protocol deviations while doffing are a major focus for HCW self-contamination, donning deviations, such as an improperly tied gown (a deviation we observed) may increase the future risk of self-contamination while doffing. Furthermore, we demonstrated that not all protocol deviations were committed by the donning HCW. For example, several doffing assistants touched the inside of HCW gowns when undoing the neck Velcro, and trained observers occasionally failed to instruct HCWs to perform hand hygiene. While previous studies have evaluated HCW protocol deviations while doffing PPE, few have evaluated the role of other HCWs in the doffing process. This is an important area for future investigation.

Boot-cover removal was particularly problematic. HCWs received varied instructions on the specifics of the boot-cover removal process. HCWs struggled to balance their legs in the air or rest their legs on their scrubs without contaminating themselves, and left-handed HCWs struggled to use right-handed scissors. All 3 left-handed EVD PPE HCWs made ≥1
protocol deviation during boot-cover removal. Many HCWs touched their scrubs with their shoes or gown, both potentially contaminated, during boot-cover removal and shoe disinfection. Herlihey et al.\textsuperscript{23} also reported difficulties with shoe-cover removal. HCWs caring for EVD patients may be exposed to large amounts of environmental contamination;\textsuperscript{13} thus, this component of EVD PPE removal may benefit from process improvement. Notably, the recommendations for boot-cover removal have changed since this study was performed. At BJH, the revised process doffs gowns before boot covers, eliminates the use of scissors, and allows HCWs to keep their feet on the ground. We hypothesize that these changes will decrease protocol deviations, but more studies are needed to confirm this.

Hand hygiene and glove removal protocol deviations were common during doffing of both EVD and CP PPE (67% and 39% of HCWs made ≥1 error, respectively). During EVD PPE doffing, common protocol deviations included touching outer gloves with inner gloved hands and touching the outside of gloves with bare hands. Herlihey et al.\textsuperscript{23} reported similar challenges doffing multiple pairs of gloves. Casanova et al.\textsuperscript{15} compared HCW self-contamination after doffing PPE with single gloves versus double gloves, using MS2 as a marker, and found that although double gloves reduced viral transfer, MS2 was still recovered from the hands of 23% of HCWs after doffing. These results may not be directly comparable to our study because it is unclear whether those HCWs performed hand hygiene after doffing. Regardless, hand hygiene and glove removal are high-risk opportunities for HCW self-contamination. For both the EVD and CP groups, we found fluorescence on HCW hands more often than any other site. HCWs may benefit from targeted training in the correct method for glove removal during EVD PPE doffing, and training should reinforce the fact that gloves are not a substitute for proper hand hygiene.

Measures to reduce HCW self-contamination rates include training and maintenance of training. Several previous studies have suggested that, regardless of PPE type, increased training and access to published doffing/dofﬁng guidelines improves HCWs’ ability to don and doff PPE without protocol deviation.\textsuperscript{7,12,22,24,25} All EVD PPE HCWs in our study previously had received formal EVD PPE training. By contrast, although 72% of CP PPE HCWs reported having previous training in PPE donning/dofﬁng, this training was often informal, "on the job" training from other HCWs. Similarly, Turnberg et al.\textsuperscript{26} reported that 15%–40% of HCWs had not received PPE training during the previous 12 months, and John et al.\textsuperscript{27} found that "on the job" training was the most common method of PPE training for HCWs. Despite the comparative simplicity of the CP PPE doffing process, only half of the HCWs were able to don PPE without protocol deviation, and only approximately one-third were able to doff PPE without protocol deviation. Common CP PPE doffing protocol deviations included touching the front of the gown with bare hands or allowing the contaminated gown to brush against scrubs while disposing. Possibly, HCWs may be unaware that specific guidelines exist for doffing/dofﬁng CP precautions.\textsuperscript{6} Beam et al.\textsuperscript{28} demonstrated that simple exposure to a poster showing the correct doffing/dofﬁng sequence may not be enough to improve HCW practices. Tomas et al.\textsuperscript{2} found that a training session on CP PPE doffing techniques led to a significant decrease in HCW self-contamination. Formal, targeted interventions or education programs may be needed to improve CP PPE doffing/dofﬁng practices.

We were unable to demonstrate clear superiority of either surrogate marker. Fluorescence was detected more frequently than MS2. MS2 was not detected from most sites with fluorescence, and MS2 was detected from 3 sites without fluorescence. Commercial preparations of MS2 are expensive; thus, fluorescent markers, which are inexpensive, may be preferable. Conversely, Casanova et al.\textsuperscript{15} found considerable MS2 transfer to HCW hands and scrubs in the absence of fluorescence; thus, fluorescence may not accurately mimic transmission of viral particles. MS2 is not visible to the naked eye, and it is possible in our study that additional areas of MS2 contamination were not detected because, outside of HCW hands, face, and arms, we sampled only those areas that fluoresced. Additional data are needed on the relative benefits and limitations of these surrogate markers.

This study has several limitations. It was a relatively small pilot study and as such was underpowered. The small number of HCWs may not be reflective of HCW populations at large. The methods need replication in larger studies, and our methods and results may be useful in designing these. The end of the 2014–2015 EVD outbreak may remove the impetus for healthcare facilities to continue EVD PPE training programs, potentially making future studies of HCWs using EVD PPE more challenging. Some EVD PPE doffing recommendations have been revised since this study was performed. CP PPE, however, are routinely used in healthcare facilities, and larger studies may be possible. We used PCR for MS2 detection; therefore, MS2 detection may not be reflective of viable MS2.

In conclusion, PPE are critical for protecting both HCWs and patients from pathogens, regardless of whether the pathogen in question is high impact like EVD or commonly encountered like \textit{C. difficile}. Previously published data on doffing and doffing EVD PPE are limited, both by the number of studies available and the types of data and analyses.\textsuperscript{1,4} There are even fewer data on doffing and doffing CP PPE. Our study highlights some potential areas for future research, including an improved boot-cover removal process, improved HCW education in the correct processes for glove removal, and an overall need for better training in the use of CP PPE. Both fluorescent markers and MS2 can be used safely as surrogates for pathogen transmission, although the relative strengths of each need further evaluation. Overall, improved processes for doffing/dofﬁng PPE and improved methods for evaluated these processes will help to protect both HCWs and patients from exposure to pathogens.

\textbf{Acknowledgments}

The authors would like to thank the Barnes-Jewish Hospital Infection Prevention and Interventional Epidemiology Team for their support and assistance with this study.
Financial support: This study was supported by the Centers for Disease Control and Prevention (CDC) Prevention Epi-Center (grant no. 3U54CK000162-05S1). J.H.K. was supported by the Washington University Institute of Clinical and Translational Sciences and a grant (grant no. UL1TR000448, subaward KL2TR000450) from the National Center for Advancing Translational Sciences of the National Institutes of Health (NIH). The content is solely the responsibility of the authors and does not necessarily represent the official view of the NIH. S.Y.L. was supported by the KM1 Comparative Effectiveness Research Career Development Award (grant no. KM1CA156708-01), the Clinical and Translational Science Award program (grant no. UL1RR024992) of the National Center for Advancing Translational Sciences, and the Barnes-Jewish Patient Safety and Quality Career Development Program, which is funded by the Foundation for Barnes-Jewish Hospital.

Potential conflicts of interest: S.Y.L. serves as a subinvestigator for institutional research studies supported by Cepheid. V.J.F. reports that her spouse is Senior Vice President and Chief Medical Officer for Express Scripts; she has current funding from the NIH, the CDC, the Doris Duke Charitable Foundation, and the Foundation for Barnes-Jewish Hospital. She has past funding from the NIH, the CDC, and the AHRQ. All other authors report no conflicts of interest relevant to this article.

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