Regarding "Clostridium Difficile Ribotype Does Not Predict Severe Infection"

To the Editor—We read with interest the recent article from Walk et al, "*Clostridium difficile* Ribotype Does Not Predict Severe Infection" [1]. However, their interpretation makes 2 epidemiologic errors, rendering their conclusions unreliable.

First, they treat "no evidence of difference" as "evidence of no difference": These are equivalent if, and only if, a study is adequately powered to exclude the possibility of a modest effect, for example, based on a 95% confidence interval (CI) (as in noninferiority trials). The comparison of 027/078 vs other ribotypes for severe disease had an unadjusted odds ratio (OR) of 2.33 (95% CI, 1.03–5.02; $P = .035$). Adjusting for age, Charlson comorbidity index, hematocrit, or platelets reduced the OR only slightly (still remaining >2.1) with P values of .06–.07 and 95% CIs of .92–4.78 (ie, essentially unchanged). The .05 P value cutoff is an arbitrary threshold from days when significance tables had to be calculated laboriously by hand [2]. Statistically, there is little difference in the strength of evidence provided from findings with P values of .035 and .07, as evidenced by the CIs in Table 3 [1], which all extend above 4, well above any plausible margin that might be considered noninferior. The lowest OR is for tcdC deletion (1.26 [95% CI, .38–4.94]); however, the meaning of this estimate is unclear as this deletion is ubiquitous among ribotype 027 isolates, and hence is completely confounded with the majority of the

"hypervirulent" strains included in the dataset. Unfortunately, replicating "no evidence of difference" in a separate validation cohort [1] does not transform it into "evidence of no difference"—it simply means that 2 underpowered studies have been conducted.

Another problem is adjusting for factors (biomarkers at diagnosis) on the causal pathway between an exposure (*C. difficile* ribotype) and an outcome (disease severity), sometimes denoted mediation or overadjustment bias [3]. Consider a pathogen in which the entire mechanism for causing disease is through raising white blood cell (WBC) count. Suppose there are only 2 strains, 1 of which causes twice the WBC count rise. By definition, all the differential mortality between these 2 strains will be due to their differential WBC count rise—adjusting for this will reduce the WBC count–adjusted effect of a strain to zero. Essentially, if there is any strain effect on a biomarker (not analyzed by Walk et al [1]), then the biomarker-adjusted OR for a strain represents an effect that is *not* mediated through biomarkers. It cannot be interpreted as the causal effect of strain, because it overadjusts for strain-related biomarker differences.

This study shows that WBC count and albumin are strong predictors of *C. difficile* severity and that there is an unadjusted association between strain and severity. However, unfortunately it does not address the key question as to the causal impact of strain including biomarker-mediated and biomarker-independent effects. Our larger study of 1893 enzyme immunoassay–positive, culture-positive, multilocus sequence–typed strains shows definitively that strains vary in their overall impact on mortality (adjusted for multiple potential confounders) and in their overall impact on biomarkers (predominantly those associated with inflammatory pathways), and that residual variation in mortality risk remains even after adjusting for biomarker-mediated effects [4].

Notes

Financial support. This work was supported by the National Institute for Health Research (NIHR) under its Oxford Biomedical Research Centre Infection Theme and the UK Clinical Research Collaboration (UKCRC) Modernising Medical Microbiology Consortium, the latter funded under the UKCRC Translational Infection Research Initiative supported by the Medical Research Council, the Biotechnology and Biological Sciences Research Council, and the National Institute for Health Research on behalf of the Department of Health (grant G0800778) and the Wellcome Trust (grant 087646/Z/08/Z). D. W. C. and T. E. A. P. are NIHR Senior Investigators. D. W. E. is a NIHR Doctoral Research Fellow.

Disclaimer. The views expressed in this publication are those of the author(s) and not necessarily those of the National Health Service, the NIHR, or the Department of Health.

Potential conflicts of interest. The institution of D. W. C. and T. E. A. P. received per-case funding from Optimer Pharmaceuticals to support fidaxomicin trial patient expenses. D. W. C. and T. E. A. P. also received honoraria from Optimer Pharmaceuticals for participation in additional meetings related to investigative planning for fidaxomicin. M. H. W. has received honoraria for consultancy work, financial support to attend meetings, and research funding from bioMérieux, Optimer, Novacta, Pfizer, Summit, The Medicines Company, and Viropharma. All other authors report no potential conflicts.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

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References

1. Walk ST, Micic D, Jain R, et al. *Clostridium difficile* ribotype does not predict severe infection. Clin Infect Dis **2012**; 55: 1661–8.
2. Sterne JA, Davey Smith G. Sifting the evidence—what's wrong with significance tests? BMJ **2001**; 322:226–31.
3. Schisterman EF, Cole SR, Platt RW. Overadjustment bias and unnecessary adjustment in epidemiologic studies. Epidemiology **2009**; 20:488–95.
4. Walker AS, Eyre DW, Wyllie DH, et al. Relationship between bacterial strain type, host biomarkers and mortality in *Clostridium difficile* infection. Clin Infect Dis **2013**; in press.

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Clinical Infectious Diseases 2013;56(12):1845–6

DOI: 10.1093/cid/cit098