never smoked (3). In 2014, we provided estimates of lung cancer risk according to both average respirable elemental carbon (REC) exposure and cumulative REC exposure (lagged 15 years), adjusting for smoking using 3 approaches: smoking status/duration of smoking, smoking status/pack-years of smoking, and status/duration and packs per day (6). All 3 approaches for smoking adjustment yielded similar estimates of risk—highly significant trends for cumulative REC and borderline-significant trends for average REC intensity. Dr. Möhner suggests that adjustment for smoking should be based on the crude dichotomy “ever smoker/never smoker” (1). Although ignoring smoking intensity seems undesirable given the large differences in lung cancer risk by number of packs smoked per day, ever/never adjustment leads to estimates of risk similar to those in the original publication (3). For example, odds ratios for cumulative REC exposure (lagged 15 years) adjusted for ever/never smoking, by quartile, were 1.00 (referent), 0.79 (95% confidence interval (CI): 0.44, 1.41), 1.50 (95% CI: 0.75, 2.98), and 2.58 (95% CI: 1.22, 5.48), respectively (P for trend = 0.002).

Second, it is critical to include worker location in the models used to estimate the diesel exhaust exposure-response relationship. Because the effect of smoking is attenuated in underground workers, adjustment for smoking without including worker location in the model will lead to erroneous estimates of risk (7). Worker location is, however, highly correlated with diesel exposure but not an independent risk factor for lung cancer (after adjustment for diesel exposure and smoking). Consequently, inclusion of worker location in the models may also have resulted in underestimation of lung cancer risk associated with diesel exposure in DEMS.

Dr. Möhner also asserts that a permanent change in worker location depends on miners’ health status (1). However, health status is one of several factors (such as preferences in type of work and work environment) that can result in a change in worker location. In the nested case-control study, we examined movement between surface and underground jobs and found cases and controls to be similar with respect to movement patterns. Thus, there is direct evidence against a healthy worker effect by worker location, as we previously noted (7).

Lastly, Dr. Möhner calls for additional reanalyses (1). DEMS data have undergone extensive reanalyses by several independent groups. The value of additional reanalyses is unclear, since epidemiologic, experimental, and mechanistic findings, taken together, provide convincing evidence that diesel exhaust causes lung cancer in humans (8).

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RE: “SOCIOECONOMIC POSITION AND DNA METHYLATION AGE ACCELERATION ACROSS THE LIFE COURSE”

We were excited to read the paper by Hughes et al. (1) examining DNA methylation (DNAm) age acceleration and measures of socioeconomic position using a life-course approach. They found that, compared with participants whose parents worked in professional or managerial occupations, individuals with unemployed parents or parents in unskilled positions in childhood had elevated epigenetic age acceleration

later in life. These findings are interesting and may implicate early-life circumstances with alterations in mortality-associated biomarkers.

Metrics of biological age, including DNAm age, are an exciting new area of research (2–4)—they might capture age-related biological variability which, in turn, can be used to improve risk predictions. However, with escalating attention, there is a growing

opportunity for misuse. In this study, Hughes et al. calculated the difference between DNAm age and chronological age to represent epigenetic age acceleration (Δ age). Unfortunately, as noted by the authors, this method creates a situation where the chronologically younger individuals had only positive age acceleration and the chronologically older individuals had only negative age acceleration. The introduction of this systematic bias is unnecessary and could result in inaccurate association estimates. Notably, the negative correlation between Δ age and chronological age has previously been reported (5), with the proposed solution of regressing DNAm age on chronological age and using the residuals to represent age acceleration. By construction, the residual differences are independent of chronological age, thereby producing age-acceleration metrics that are immune to age-associated bias. The authors should consider reexamining their data using this approach.

Biological age estimators are increasingly under investigation in epidemiologic studies. We hope to raise attention to this critical issue given that miscalculation of novel biomarkers might slow our understanding of otherwise useful risk-assessment instruments.

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THE AUTHORS REPLY

We thank Kresovich and Taylor for their interest (1) in our work (2). As the authors note, other researchers have previously reported the issue of age-related confounding where DNA methylation (DNAm) age is the outcome. However, as social scientists increasingly begin to use these measures with exposures especially vulnerable to age-related confounding, we believe it is crucial to stress just how important it is to take age-related confounding into account, either by use of residuals or by adjustment. The case of the spurious relationship with unemployment was given to illustrate this point.

Age-related confounding of DNAm age acceleration can be addressed in different ways. One approach is to regress DNAm age on chronological age and use the residuals; another, the approach taken in our paper (2), is to adjust in analyses for chronological age (and a squared term, to allow for possible nonlinearity in age-related confounding). Adjusting for a confounder in one model and using the residuals as an outcome in a second model is, statistically speaking, extremely similar to adjusting for the confounder in the second model.

It is therefore unsurprising that we obtain very similar results when using the residuals method suggested by Kresovich and Taylor (1). In new analyses, Horvath and Hannum age were regressed on chronological age, age squared, and the white blood cell measures following Chen et al. (3). The residuals were then used as outcomes in models with socioeconomic position variables as the exposure, adjusting for the remaining confounders. Using this method, the elevations in Horvath age for parental

social class groups (compared with professional/managerial) were: for skilled nonmanual, 1.32 years (95% confidence interval (CI): 0.12, 2.51); for skilled manual, 0.53 years (95% CI: −0.23, 1.29); for semiskilled/unskilled, 0.78 years (95% CI: −0.11, 1.67); and for neither parent in work or both deceased, 2.20 years (95% CI: 0.52, 3.88). For the Hannum clock, equivalent elevations were: 0.22 years (95% CI: −0.55, 0.99); 0.75 years (95% CI: 0.15, 1.36); 0.94 years (95% CI: 0.11, 1.77), and 1.67 years (95% CI: 0.48, 2.86). As with our original results, no significant elevations were seen for the other measures of socioeconomic position. Thus, our conclusions are not altered by the method used to adjust for age-related confounding.

We also stress that, holding other aspects of the model constant, identical coefficients for exposures other than age are obtained whether the outcome is DNAm age or DNAm Δ age. The proof is given below.

Consider the following 2 regression models, of DNAm age and DNAm Δ age, respectively, on an exposure of interest and covariates, adjusting for chronological age and a squared term:

$$\text{DNAm age} = \beta_0 + \beta_{\text{age}} \times \text{age} + \beta_{\text{age}^2} \times \text{age}^2 + \beta_x \mathbf{X} + \varepsilon_1 \quad (1)$$

$$\begin{aligned} \text{DNAm } \Delta \text{age} = & \alpha_0 + \alpha_{\text{age}} \times \text{age} + \alpha_{\text{age}^2} \times \text{age}^2 \\ & + \alpha_x \mathbf{X} + \varepsilon_2 \end{aligned} \quad (2)$$