Gender, Cytidine Deaminase, and 5-Aza/Decitabine—Response

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We thank Ciccoline and colleagues for their attention and comments regarding "Increased CDA expression/activity in males contributes to decreased cytidine analog half-life and likely contributes to worse outcomes with 5-azacytidine or decitabine therapy" (1). Ciccoline and colleagues have not found a sex difference in CDA activity using high-performance liquid chromatography (HPLC)–based measurements of CDA enzyme activity in red blood cell (RBC) lysates (2). In addition to use of gemcitabine as a substrate, which has a more than 10-fold higher $K_m$ for CDA than cytidine (3), this methodology is limited by its analysis of RBC lysates, which may not be representative of CDA activity in pharmacologically more relevant tissues, for example, the liver. Also, pharmacologically relevant are the cancer tissues that are the therapeutic targets of therapy. In contrast to the use of a single methodology (2), we evaluated sex differences in CDA using HPLC-based enzyme activity analysis of heparinized plasma, quantitative real-time PCR (qRT-PCR) quantification of CDA expression in murine liver, mass spectrometric measurement of decitabine levels (a CDA–drug substrate) in murine plasma, and Western blot evaluation of decitabine pharmacodynamic effect. We had consistent results across these model systems and methods, and furthermore, observed sex differences in CDA expression in various target cancer tissues analyzed by microarray. Here, we provide additional verification: CDA was expressed at significantly higher levels in noncancerous human liver from human males compared with females (Fig. 1A). That expression levels of CDA can be more important than CDA-genotype in determining CDA enzyme activity (1) is also illustrated by substantially greater CDA enzyme activity in U937 cells, that express higher levels of the enzymatically less active “CC” CDA variant, compared with cell lines that contain more active “AA” and “AC” CDA variants expressed at lower levels (Fig. 1B and C). We agree that our evaluation of clinical impact was limited by its retrospective nature (1), and sex-based dosage adjustment would be premature. Even so, the clear difference in MDS treatment outcomes despite similar disease biology (cytogenetics and myeloblasts) in the 2 sexes was highly provocative and consistent with epigenetic

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therapeutic effects that by being S-phase dependent must interact with brief drug half-lives that are further abbreviated by higher CDA enzyme activity in males. Furthermore, clinical trial results and mathematical modeling indicate that the negative impact of lower drug exposure time is exacerbated in low S-phase fraction malignant disease (e.g., MDS; ref. 1). Given that marginal differences in exposure time can determine treatment success or failure with 5-azacytidine or decitabine (4–5), pharmacodynamic or other dosage guidance is clearly a worthy goal.

Disclosure of Potential Conflicts of Interest
No potential conflicts of interest were disclosed.

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References