A cysteine-scanning mutagenesis study of transmembrane domain 8 of the electrogenic sodium/bicarbonate cotransporter NBCe1.
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We reported under “Experimental Procedures,” paragraph heading “Statistics,” that data sets of transporter function, expression, and sulfhydryl sensitivity were compared using one-way analysis of variance (ANOVA) using a relatively stringent Tukey criterion. However, because each experimental data set was compared with a control data set, the ANOVA analysis was equivalent to a less stringent, two-tailed Student’s t test where a Tukey post-hoc means comparison is irrelevant. This correction has the most impact on the interpretation of the MTSEA and pCMBS sensitivity data presented in Figs. 3 and 4, respectively. As described by Akabas and Karlin (Akabas, M. H., and Karlin, A. (1995) Biochemistry 34, 12496–12500), the stringency of the statistical test can influence conclusions regarding small effects of sulfhydryl reagents in a cysteine-scanning mutagenesis study. The sulfhydryl reagents used in our study did have small effects on a number of the tested NBCe1-A mutants. We have therefore performed a more stringent, combined data set ANOVA analysis using the Dunnett’s post-hoc means comparison ($p < 0.05$). This post-hoc test is appropriate for comparing each experimental data set (i.e. sulfhydryl sensitivity of a mutant NBC) to a single control data set (i.e., sulfhydryl sensitivity of wild-type NBC). As expected, results from the more stringent analysis yielded fewer NBC mutants sensitive to MTSEA or pCMBS. More specifically, the M753C mutant is no longer one of the original seven NBC mutants reported to be inhibited by MTSEA. In addition, the I757C mutant is the only one remaining of the original six NBC mutants reported to be stimulated by MTSEA. Finally, the A740C and Q756C mutants are no longer among the original group of five NBC mutants reported to be inhibited by pCMBS. However, even a more stringent analysis does not impact the following three major conclusions of the study. First, many MTSEA-inhibited positions are clustered from positions 748 to 756. Second, the majority of the pCMBS-inhibited positions lie on one side of transmembrane domain 8. Finally, the accessibility of L750C to pCMBS is influenced by substrate concentration, stilbene binding, and membrane potential.

Acknowledgments—We used SigmaStat V3.5 (Systat Software, Inc.) for the ANOVA analysis with the Dunnett’s post-hoc test. We acknowledge the helpful advice of Drs. Xiangqin Cui and Naomi S. Fineberg of the Department of Biostatistics, University of Alabama at Birmingham.
A novel ARID/Bright-like protein involved in transcriptional activation of cyst wall protein 1 gene in *Giardia lamblia*. VOLUME 282 (2007) PAGES 8905-8914
Chih-Hung Wang, Li-Hsin Su and Chin-Hung Sun

*J. Biol. Chem.* 2007, 282:15940.

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