Comments on “Experimental determination of net protein charge, [A]_{tot}, and $K_a$ of nonvolatile buffers in bird plasma”

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TO THE EDITOR: I am writing to express some concerns about the article by Stämpfli et al. (7), “Experimental determination of net protein charge, [A]_{tot}, and $K_a$ of nonvolatile buffers in bird plasma.”

First, it appears that the BCG dye binding technique was used to determine albumin concentration. This technique has not been validated for avian albumin and does not correlate with electrophoretic results (4). Electrophoretic determination of albumin concentration would have been a better choice of measurement. I think the authors need to qualify their statements about indexing [A]_{tot} to albumin concentration, as their measurements may have been in error.

Second, the authors use an incorrect term, “total solids,” to describe the readings obtained from a Leica Vet 360 refractometer. They were actually measuring total protein by a total solids technique. The term total solids refers to all solutes in plasma and is $\sim 15–20 \text{ g/l}$ higher than the protein concentration for most mammalian samples. Refractometers measure protein by the angle of light produced by refraction caused by the total solids in the sample and then subtracting the expected amount of refraction from non-protein solids in the sample. The Leica Vet 360 includes that calculation and has protein (g/dl) listed clearly on the scales in the internal reticle (2).

Refractometers have been reported to give higher results than the biuret technique for avian samples for various species (4–6). That discrepancy may be due to relatively higher glucose or other non-protein solids in the plasma of many avian species (4). Pigeons have higher glucose concentrations in their plasma than most mammals, perhaps contributing to higher protein results by refractometry reported by the authors (1).

Lastly, I believe Ref. 12 is incorrect (3). That article does not contain any information about comparative accuracy of refractometry in various species. Perhaps they meant to cite a review article written by me containing that information (2).

REFERENCES