



SDI Review Form 1.6

Journal Name:	International Journal of Biochemistry Research & Review
Manuscript Number:	2014_IJBcRR_8940
Title of the Manuscript:	Tissue Standards for Aging Research
Type of the Article	Method Article

General guideline for Peer Review process:

This journal's peer review policy states that **NO** manuscript should be rejected only on the basis of '**lack of Novelty**', provided the manuscript is scientifically robust and technically sound.

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PART 1: Review Comments

	Reviewer's comment	Author's comment (if agreed with reviewer, correct the manuscript and highlight that part in the manuscript. It is mandatory that authors should write his/her feedback here)
Compulsory REVISION comments	The article evaluates the linearity and reproducibility of the stain free method as a loading control compared to direct blue staining and western blot for different housekeeping proteins. The authors found that for aging study in mice the total protein staining as loading control were superior to housekeeping proteins, and among the total protein detection methods the stain free (i.e. UV detection) is superior to direct blue. Figure 1. could benefit by having a more descriptive parts. Showing the total protein stain or stain free and let the reader reveal its linearity is impractical, the figure in its current form is very general, no statistical analysis to show the differences between the different methods and different tissues. It would be valuable if the authors add graphs to describe the differences and the linearity of the different methods. Using the pure CoV and R2 in the table is not descriptive at all. This reviewer would like to see that these data are presented as graphs or imbedded in the graph of figure 1, describing the quantitative and significant linearity of the methods. I must question the total scientific value of this type of a manuscript to the scientific community? The idea of total protein staining has been described elsewhere, stain free method for aging research could be beneficial, however, the description of the results and the discussion are very generalistic and its significance is not clear. The authors must discuss the data comprehensively and indicate the advantage and the disadvantage of their methods. UV total protein detection needs specific instrumentation which increase the costs, why the authors didn't indicate that. the main question is does this manuscript add a significant contribution to the scientific community?	1-We understand the reviewer's comments. We ran nine gels for the young and old tissue comparison, while a further nine gels were run for the linearity analysis; we thought this was too many gels to include in one figure. Each of the gels used in the linearity analysis contained triplicate samples, resulting in nine separate regression curves each with three data points and standard deviations for each data point. Again we felt this would be incomprehensible as a single graph. Hence we choose to show a demonstration gel and a statistical summary. We have included P values in the updated paper to help describe the important differences in the various methods. We thank the reviewer for their comments, and we have expanded the description of the results and the discussion section to indicate the unique additions to the field this paper will make. We have also included a more comprehensive discussion of the benefits and drawbacks to the stain free method.
Minor REVISION comments		
Optional/General comments		