



SDI FINAL EVALUATION FORM 1.1

PART 1:

Journal Name:	International Journal of TROPICAL DISEASE & Health
Manuscript Number:	Ms_IJTDH_36186
Title of the Manuscript:	SDS-PAGE and Densitometric analysis of Myofibrillary Proteins : Actin and Myosin in Skeletal Muscle of Fluoride intoxicated rats
Type of Article:	Original Research Article

PART 2:

FINAL EVALUATOR'S comments on revised paper (if any)	Authors' response to final evaluator's comments
With all due respect I have to insist that the fundamental problem of this paper lies in the lack of specificity. Since the gel was stained with Coomassie brilliant blue, and the dye interacts electrostatically by noncovalently with all proteins with amino and carboxyl groups; this is the reason of lack specificity. In other words, there is no evidence to reliably conclude that the bands quantified correspond to actin and myosin. I have insisted that at least the high-purity standards of each of the 2 proteins of interest would have to run on the same electrophoresis. Even if they had done so, the bands seen in samples with the same molecular weight do not necessarily correspond to actin and myosin. It is essential to confirm its identity with an additional analysis; this could be done for example, using a specific antibody for these proteins. In the latter case, the procedure known as Western Blot (WB) is required. WB is a method widely used for many years and is currently being reviewed in relation to its reliability to quantitatively determine a protein of interest. Below I am posting a link where this current topic can be reviewed in further fashion. http://www.bioradiations.com/defining-the-new-normal-in-quantitative-western-blot-data/ Finally, I also want to reiterate, that it is imperative to apply the same amount of total protein (μg) to the wells for electrophoresis. The 30 μL	Present study was undertaken to evaluate the SDS-PAGE and Densitometric analysis of Myofibrillary Proteins : Actin and Myosin in Skeletal Muscle of Fluoride intoxicated rats . Our study aimed at mainly the study of two important contractile proteins - Actin and Myosin by using the technique SDS-PAGE. • The skeletal muscle tissue samples were processed for Electrophoresis, samples were loaded and were analysed . the molecular weight ladder was also used and marked as 'M' • Further the protein bands were confirmed by determining the Molecular weight in gel documentation system. • The molecular weight were thus compared with the literature available, thereby confirming the samples to be as Myosin and Actin. • The samples were analysed using Image J software and the densitometric analysis was further done.



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volume as the authors point out, may contain very different amounts in the total protein mass. It could be that in 30 μL of the samples from rats treated with 300 ppm of NaF, 300 μg of the total protein mass are contained, and that in 30 μL of the samples from rats treated with 600 ppm of NaF, are contained 100 μg of protein total mass. With this situation, equivalent results to those intended in this paper would be obtained.

We aimed at the pathological effects of Sodium fluoride on these two proteins by comparing the treatment groups with control,