**Rebuttal letter, “Gamma-H2AX biodosimetry for use in large scale radiation incidents: comparison of a rapid lyse/fix protocol with a routine method" (#2013:12:1121:0:0:REVIEW)**

Dear Søren, Helen and anonymous reviewers,

We would like to thank you all for the positive comments and helpful suggestions. We have now included more details on the methods and the data analysis, as requested. See below for the specific changes made to the manuscript to address the points raised in your comments.

Reviewer 1:

Methods section: 17 donors were used for this study. The repetition regarding control samples has been removed. ID numbers for antibody, lyse/fix buffer and deep-well plates have been added.

Results/Discussion section: The opening sentence now notes that Figure 1 shows the individual protocol steps.

Comments for the author: Line 78 of the text and the title of Figure 5 have now been amended as requested.

Reviewer 3:

Small blood volume: Thank you for raising this important point. The following paragraph has been added to the end of the Results/Discussion section: “Given the growing interest in point of care diagnostics, finger prick sampling of capillary blood has been widely tested and confirmed in numerous studies as a suitable alternative to venipuncture. Examples of such studies include a comparison of seroepidemiology of hepatitis B (Bond et al, 1978), CD4 cell counting (MacLennan et al, 2007) or differential leukocyte counting (Hollis et al, 2012). We are not currently aware of any specific comparison of gamma-H2AX induction following irradiation but expect that results for capillary and venipuncture samples should be similar for this endpoint. One exception might be a scenario where blood is taken within seconds (or perhaps up to a few minutes) following a partial body exposure. In such a situation, blood lymphocytes in capillaries may not have had sufficient time to mix completely. However, this could only be of concern in studies where samples are taken immediately following a planned, very short and localised exposure, such as a CT scan (Rothkamm et al, 2007). Naturally, the lyse/fix method presented here would also work with venipuncture samples where it would save blood, money and time that could then be used for other tests.”

Methodology: Additional methodological details regarding the radiation exposure setup / dosimetry and statistical analysis have been added.

Lyse/Fix method: We have now added “96 well” in the title, abstract and main text to highlight this feature.

Specific comments:

Page 1, Abstract, line 9: changed as suggested.

Page 1, Abstract: “Accuracy” findings are now also given as linear regression coefficients and associated uncertainties for estimated vs. true doses, both in the abstract and towards the end of the Results/Discussion section.

Page 1, Abstract: Information about the doses used to test accuracy has been added.

Page 1, Abstract: “mean absolute differences” have been replaced by linear regression coefficients and associated errors.

Page 3, Materials and Methods: radiation exposure setup and dosimetry details are now provided.

Page 4, Materials and Methods: We have added the clarification that blood was layered onto Histopaque in 15 ml tubes.

Page 4, Materials and Methods: A paragraph has been added that describes the statistical methods used.

Page 5: Scoring times are now presented in Table 1.

Page 6: The Student’s t-test was performed to test for significant differences in foci counts and results are now included.

Page 6: Mean absolute differences have been replaced by linear regression as suggested.

Page 8, Conclusion: False negatives in automatic scoring are now mentioned.

Figure 1 legend: Information about scaling estimates has been included.

Editorial comment: The correction has been made.