Reviewer's report

Title: In vivo Assessment of Arterial Stiffness in the Isoflurane Anesthetized Spontaneously Hypertensive Rat

Version: 1
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Reviewer: Francesco Stea

Reviewer's report:

Major Compulsory Revisions

- It is wrong to report data as "mean +/- SEM". Please express variability with standard deviation all throughout the manuscript and the tables.

Page 7, Methods, Doppler US..., last paragraph: variability in Doppler-derived PWV is mainly due to path length measurement and probe placing and angling. Measuring the time intervals on a frame, i.e. counting the pixels, is a much smaller source of error (as your data do confirm). Therefore, measuring variability of time intervals is far from "assessing intra- and inter-observer variability of PWV" and this section should be restated.

Did the authors perform a real repeatability test? If not, this should be stated as a limitation.

Page 9, Results, Intra and Inter..., and figure 2: units of measurements in the text are wrong since the error is expressed either as a percentage or as a time, not m/s.

Linear regression can't be performed since here we are dealing with two distinct and different populations and this violates the basic assumptions of the test. Please remove scatter plots from Figure 2 and rely on the Bland-Altman plot. Pearson's coefficients (with 2 decimals) could just be incidentally cited in the text, bearing in mind that it's not a formally correct statistics.

Page 11, Discussion: it is not surprising at all that the correlation between readings of time intervals is "statistically significant". Actually, the opposite would have been worrying. Please remove this sentence.

Table 1: there should be something wrong. What's in columns "Carotid", "Iliac" and "PTTci"? It seems milliseconds (ms) rather than m/s. Anyway, "Carotid" and "Iliac" (presumably the time intervals between the R wave and the Doppler waves) are not defined in a table caption, which is actually missing.

Figure 1: why there's a table imbedded in the figure (with wrong units of measurement and column titles not defined)? Why there's a 160 mm length?

Please remove and replace with the formula used to calculate time interval (R to iliac - R to carotid). In the caption, "the mean pulse wave was subtracted" is meaningless.
Minor Essential Revisions

Page 2, Abstract, Background: the Background is the largest section in the abstract. Please reduce this section and expand the Methods part, where no mention is made to the actual methods used.

Page 2, Abstract, Conclusions: "demonstrates" is a strong term, since the population is small but, above all, involves only two age points. I'd replace with "suggests".

Page 7, Methods, Doppler US...: probe placing and angling is a major source of error in Doppler-derived PWV. This should be stated as a limitation later on.

Why did the authors choose to use a tape meter, greatly decreasing precision in length measurement as compared to measuring with a caliper? This should also be stated as a limitation.

Page 9, Results, PWV, and later on all throughout the manuscript: please do not repeat "12 weeks old" and "32 weeks old" every time the groups are cited, as "younger" and "older" have already been defined.

Please do not repeat "The mean X was Y and values ranged from Z to W" for every measure, but shorten to ... "and Y for X (range Z-W)" or similar after the first occurrence, to improve readability and avoid repetitions.

For non-significant P's, 2 decimals are enough (i.e. p=0.62 rather than p=0.6153). Please check here and the rest of the manuscript.

Page 10, Results, PWV and EaI: also for Pearson's coefficient 2 decimals are enough (0.53, not 0.5286)

Page 11, Discussion, last 4 lines: for the reasons already stated, the conclusions should be softened: no data is provided to say that PWV values are "highly reproducible" or "accurate", (and the correlation between PWV and EaI is not strong enough to say they measure the same thing).

Page 13, Conclusion: see above, "precise", "accurate" and "repeatable" are terms not supported by the data shown in the paper. Please soften.

Tables 1 and 2: replace SEM with SD.

Discretionary Revisions

All throughout the manuscript, PWV, being a velocity, should be defined "high" or "higher", rather than "fast" or "faster". Please change.

Level of interest: An article of importance in its field

Quality of written English: Acceptable
Statistical review: No, the manuscript does not need to be seen by a statistician.

Declaration of competing interests: I declare that I have no competing interests