Reviewer's report

**Title:** Effect of topical fluoroquinolones on the expression of matrix metalloproteinases in the cornea.

**Authors:**

Dr Victor E Reviglio (victorwilmer@aol.com)
Melinda A. Hakim (mhakim@jhmi.edu)
Jae K. Song (navymed@hanmail.net)
Terrence P. OBrien (tobrien@jhmi.edu)

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**Reviewer:** Alex Gray

**Level of interest:** A paper of considerable general medical or scientific interest

**Advice on publication:** Unable to decide on acceptance or rejection until the authors have responded to the compulsory revisions

The only concern I have regarding the manuscript is the statistical analysis method. The authors have used the T-test for analysis of the imaged data.

As I understand the design they are taking several measurements from the same image and then applying multiple T-tests to the 4 treatment groups. This is fraught with the multiple comparisons issue and not adjusting your p value for these together with not dealing with the fact that we are using multiple values from the same animal. One technique that would be easy to apply with out reverting to a complex mixed model would be a a split plot approach, which would be much better. I have included an example of some SAS code that could be used to obtain the appropriate analysis. I would recommend the authors to redo the analysis with this approach to ensure their results are the same prior to publication.

Example SAS code:

```sas
options ls=72;
data new;
infile 'file name';
input rat id factor$;
logid = log(id);
label factor = 'treatment group';

proc means n mean stderr maxdec=4;
var id;
by notsorted factor;
run;

proc glm;
```

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var id;
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run;

proc glm;
```
class rat factor;
model id logid = factor rat(factor);
test h = factor e = rat(factor);
means factor / dunnett ('control') e=rat(factor);
run;

Section 1 Example of how to get data into SAS.
Variable are RAT, which is the rat number
ID is the response variable for the imaging value
Factor is the treatment group

Section 2 This prints the title on each page

Section 3 The proc means will provide summary stats on the response variable ID, number, mean and standard error.

Section 4 Proc GLM is the procedure that will conduct the 1 way analysis of variance. It is very much like a split plot approach where we nest the treatment within the rat. We then have to define the correct error term, that is e=rat(factor).

The means statement will then compare your three treatment groups to your control and that would need to be specified after dunnett (" ").

Competing interests:

None declared.