1. Basic reporting

In the MS entitled “Anxiety-like behavior and whole-body cortisol responses to components of energy drinks in zebrafish (*Danio rerio*)” the English language is clear, and the tables and figures are easy to understand. The topic approached in the MS is relevant because the consumption of taurine and caffeine association in energy drinks is very popular, and the synergic effects of these psychoactive are still unknown.

The MS addresses current and relevant references on the subject, however, the results found in the study do not support the main idea discussed along the MS (see next evaluation topics).

Basically, some points should be rethought:

- The introduction is extensive and approaches the isolated effects of caffeine and taurine, however, little is discussed about the effects of these drugs in association. What is known about this association? This topic should be deepened.

- Which is the prevalence of the energy drinks consumption? How these energy drinks are commonly consumed (association with alcohol)? It can be related to which social problems?

- The hypotheses described in the end of introduction section are unnecessary.

2. Experimental design

The MS is original and is within the scope of the PeerJ. The research question is relevant and well defined, and the methodology used is referenced. However, the methodology applied to evaluate the effects of taurine and caffeine on anxiety-like behaviors can be mistaken or with poor reproducibility. It can be a problem to replicate this protocol in the future.

In relation to experimental design, some points should be considered:

- Was this study approved by an ethical committee? If yes, which is the approval protocol number?

- Mezzomo et al. (2016) showed that TAU exposure did not alter anxiety responses using the novel tank test. Anxiety-like responses trigger by taurine were observed only when the light dark paradigm was used. Why the authors choose to use the novel tank test to evaluate the synergic effects of taurine and caffeine?

- I suggest that the authors perform an additional experiment using the light-dark test in the same context, in a manner to increase the robustness of the findings.

3. Validity of the findings

The findings of the MS are inconclusive and without statistical significance to support the conclusions argued along all the MS.

Some points should be minutely evaluated:

- The main conclusion of the MS is that taurine exposure act mitigates the anxiety-producing effects of caffeine. However, the MS data not support this affirmation because do not exist statistical significance in the parameters discussed. No parameter evaluated in the novel tank was statistically altered by the association of taurine and caffeine. The MS only argue about trends, so, the results are not robust. I suggest that the experimental design be rethought and the light-dark test be done to confirm the data presented so far.

- The statistical analyse applied in the MS is incorrect. Two factors influence the results: taurine and caffeine, so the statistical analyse correct to be use is the two-way ANOVA.

- Why is the number of animals used in the experiments so dissimilar? In the novel tank test the number of fish per group was 10-38, and in cortisol analyse 19-39 fish per group. Please, explain why this number is so varied.

- Rosa et al. (2018) showed that acute caffeine exposure in the same concentration used in the present study (100 mg/L) increased the cortisol levels in zebrafish. However, the levels of cortisol were not altered in this study. How is possible justify this finding?

- Mezzomo et al. (2016) exposed the fish to 400 mg/L of taurine for 1 hour before evaluate anxiety-like behaviors. Why the authors perform taurine exposure for 15 minutes? There are previous studies that justify the use of this time of taurine exposure to evaluate anxiety behavior?

- In the study, taurine decreased the distance travelled when compared to other groups? However, this result is not approached in the discussion section. How this result can be explained?

- In the discussion section, many results that are not statistically significant are discussed. Mechanisms about the results are hypothesized and some points justified based on trends. Additional experiments are needed to support this discussion.

4. General comments

The topic of the study is original and relevant in neuroscience. The study has a clear objective, however, some methodological aspects need to be improved for the conclusions to be supported.

5. Confidential notes to the editor

Many aspects of the manuscript should be improved. The methodology is inadequate and the results are not statistically significant to support the MS discussion. Therefore, I suggest that MS be accepted only if the authors perform additional experiments and justify all the required queries.

Best regards,

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