Reviewer Evaluation of Bachelor Thesis

Author Name: Viktor Valentini

Thesis Title: Effect of disulfiram metabolites on NK and T cell cytotoxic activity against tumor resistant cell lines

Reviewer Name: Viswanath Das, MSc, PhD

Criteria n.	Evaluation Criteria	Points (0-5)
1	**Completeness and timeliness of the literature search**	5
2	**Quality of the introductory part (number of used original articles, the suitability of literature selection)**	5
3	**Fulfilment of thesis goals**	5
4	**Logics of literature search or experimental work approaches**	5
5	**Completeness of methods and protocols description**	5
6	**Level of results presentation (suitable choice of graphs and tables etc.)**	4
7	**Adequate interpretation of obtained results and their discussion**	4
8	**Conciseness of thesis summary in Czech and English languages**	5
9	**Graphical editing of text and images**	4
10	**Language and stylistic level, respect of valid nomenclature**	5
11	**Correctness and completeness of figure and table legends (intelligibility without regard to the main text, correct explanation of used symbols and units)**	5
12	**Correct usage of citation references (presence of non-cited data, compliance with a unified citation style, utilization of official journal names abbreviations)**	5
	Total points	**57**

max 60

Specific comments and questions to the thesis (*please use additional sheets if necessary*)**:**

The thesis evaluates the effect of CuEt, a metabolite of disulfiram, on antigen presentation of tumor cells. The study uses isolated mononuclear cells from humans and human colorectal tumor cell lines HCT116 and HT29. The experiments are well-designed, the literature the up-to-date, and the results are analysed critically.

On page 5, the candidate describes colorectal cancer (CRC) statistics relative worldwide. I would have liked some statistics on CRC locally, given that CRC is one of the major cancer types in the Czech Republic.

Can the candidate explain why only one concentration of CuEt was chosen? Where other lower concentrations tested?

On page 16 (line 2), it is mentioned that "The cytotoxic effect was more significant when immune cells were treated as opposed to the tumor cells being treated" – this means CuEt Lympho vs CuEt Tumor cell line in Graph 1, but it is not correct, or I did not understand. Please explain.

For a better understanding of the results, it would be important to present the complete statistical details. What is being compared to what in the graphs?

NFkB is mentioned as crucial for the anti-cancer effect of disulfiram if applied at 1 nM. Do you have any direct evidence?

It is mentioned that CuEt promotes the expression of antigens in the cancer cell line – since these were not directly measured, can you speculate which antigens?

Errors required to be corrected:

Minor errors (These are sugegstions and should not affect the outcome of the thesis result):

Pages 1, and Figures 2, 3, 4: HCT116 has been mistyped (text and figures) as HC116. Please correct them.

Page 11: In the section on medium composition, it is mentioned that 50 mL of antibiotics were added to 500 mL of RPMI or McCoy. I think it is 5 ml. 50 mL will be 10% antibiotics. Similarly, it should be 50 ml of FBS (100%) not 50 ml of 10% FBS.

Page 13: Tumor cells were fixed with what percentage of ethanol?

Page 14: Sulfamethoxazole was used as a negative control.

Page 17: Graph 2- I suggest both the same scales in both (right and left) y-axes.

All graphs: Please indicate data are mean +/- SEM or SD.

Page 19 (line 2): 3.5.4 should be 3.5.5

Conclusion: I recommend this thesis to defence.

In Olomouc on: 16.6.2021 Signature

Thesis evaluation according to total points:

- A- 56-60
- B- 51-55
- C- 46-50
- D- 41-45
- E- 36 -40
- F- 35 and less