

**CHARLES UNIVERSITY  
FACULTY OF PHARMACY IN HRADEC KRALOVE**

Department of Biochemical Sciences

Study program: Pharmacy

**Opinion of the Opponent of the Diploma Thesis**

Year of the defense: 2024

Student: **Melodie Haddad**  
Thesis Tutor: prof. Ing. Vladimír Wsól, Ph.D.  
Consultant: Dr. Anselm Morell Garcia  
Opponent: RNDr. Miloslav Macháček, Ph.D.  
Thesis title: **The potential of aldo-keto reductase 1C3 inhibition as an off-target effect of isocitrate dehydrogenase 2 inhibitor enasidenib in leukemia**

Scope of work, number of 80 pages, 19 figures, 2 tables, 93 citations

**Evaluation of the work:**

- |   |           |
|---|-----------|
| a) Processing of the theoretical part:                                  | Excellent |
| b) The complexity of the methods used:                                  | Excellent |
| c) Preparation of the methodological part (clarity, comprehensibility): | Very good |
| d) The quality of the experimental data obtained:                       | Excellent |
| e) Processing of results (clarity):                                     | Excellent |
| f) Evaluation of results, including statistical analysis:               | Excellent |
| g) Discussion of results:   | Excellent |
| h) Clarity, conciseness, and adequacy of conclusions:                   | Excellent |
| i) Meeting the objectives of the work:                                  | Excellent |
| j) Quantity and up to date of references:                               | Very good |
| k) Language level (stylistic and grammatical level):                    | Excellent |
| l) Formal level of the work (text structure, graphic design):           | Excellent |

I recommend the thesis for recognition as a rigorous thesis

**Comments on the evaluation:**

Melodie Haddad's work is very well written and easy to read. I would like to point out that although the student is not a native speaker, the style and grammar of the text is great. Text has minimum of errors – however, it has occasional minor flaws of a rather typographical nature: D/L isomerism should be written in small caps not as “d” or “l” (e.g. page 16 - NMDA); or maybe done by autocorrect function of an editor (4HNE is 4-hydroxynonenal or 4-hydroxy-2-nonenal, not hydroxy-2-nominal). E.g. on page 24, citations are repeated in the two subsequent paragraphs – it should be one citation after those two paragraphs. I believe that chapter 3.2.3 on page 43 should be numbered 4.2.3? However, minor drawbacks mentioned above have minimal effect on the overall quality of the whole diploma thesis. Overall, I evaluate the thesis as being of very high quality.

Questions and comments to student:

Comments:

A) On page 43 – chapter „Daun quantification by HPLC“ (in Methods) is missing the methodology. It's a description of HPLC, but description of the specific method used to quantify Daun by HPLC is missing.

Questions:

1) In your thesis, you describe the targeted therapy as a potential candidate for combination therapy with anthracyclines (e.g. Daun). Is there different approach how to decrease toxicity of anthracyclines while maintain the same level of activity against malignant cells (e.g. by changing pharmacokinetics and change tissue distribution)?

2) In the determination of Daun accumulation in the cells (page 44) you used 1  $\mu\text{M}$  concentration – what is a clinically achievable concentration of Daun in the blood/plasma?

3) One hour incubation is used in the determination of accumulation (page 44), while 24 h incubation is used in the determination of Daun metabolism in KG1 $\alpha$  cells (page 45) and 30 min and 24 h in the determination of synergism. Why not the same incubation time, or why not use more, but in all experiments (0,5 + 1 + 24 h)?

**Evaluation of the thesis: Excellent**

**For the  
defense:**

**Recommend**

In Hradec Králové

31. května  
2024

signature of the opponent