CHARLES UNIVERSITY
FACULTY OF PHARMACY IN HRADEC KRALOVE

Department of Pharmaceutical Chemistry and Pharmaceutical Analysis

Study program: Pharmacy

Opinion of the Opponent of the Diploma Thesis

Year of the defense: 2023

Student:	**Miroslav Domanský**
Thesis Tutor:	doc. PharmDr. Jan Zitko, Ph.D.
Consultant:	Mgr. Marek Kerda
Opponent:	prof. PharmDr. Petr Zimčík, Ph.D.
Thesis title:	**SYNTHESIS OF NOVEL INHIBITORS OF HUMAN TOPOISOMERASE BASED ON HIGHLY SUBSTITUTED PHENYL SCAFFOLD**

Scope of work, number of 76 pages, 26 figures, 0 tables, 48 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):	Excellent
d)	The quality of the experimental data obtained:	Excellent
e)	Processing of results (clarity):	Very good
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:	Excellent
k)	Language level (stylistic and grammatical level):	Very good
l)	Formal level of the work (text structure, graphic design):	Very good

I recommend the thesis for recognition as a rigorous thesis ☒

Comments on the evaluation:

The presented work has been done in collaboration with Faculty of Pharmacy, University of Ljubljana within the framework of Erasmus+ program. It is focused on the synthesis of novel inhibitors of human topoisomerase based on the structures investigated in the foreign institution. The thesis is composed in accordance with the recommendations. It starts with theoretical part discussing mostly cancer and involvement of topoisomerases, their type and characterization. I highly appreciate the following description of inhibitors of topoisomerases with their mechanism of action – both used and investigated compounds have been mentioned. The Experimental part describes well the performed reactions with all necessary details for their reproduction. However, it is more common to place the characterization of the compounds directly to the chapter/paragraph describing their synthesis. The data in presented work are placed in a separate chapter. Besides problems with finding the data, it also leads to situation where e.g., HPLC chromatograms are placed in Experimental section or in Characterization section without any system. Additionally, the Experimental section

contains part of the discussion (e.g., on the sideproduct from DMF) that should be rather placed in Discussion. The following Discussion section describes the “story” behind the synthesis and change of the conditions with the brief results of biological testing. Although not very long, it is sufficient to explain the problems and attempts to solve them. The antiplagiarism software detected several similarities that are mostly irrelevant with exception of the thesis of Barbora Koutníková (24 %) where e.g., the whole paragraph describing the method for topoisomerase inhibition is completely the same. This is understandable as this is exact description of the procedure performed in Ljubljana. Another important similarities can be found in the Theoretical part. These are, however, taken from literature and properly cited. The overall style and grammar are on very good level and the work is well readable.

Questions and comments to student:

- In really many cases, the space between number and unit has not been kept.

- Anhydrous MeOH was used during synthesis of IMD-04. Is it necessary? In the next step you add solution of NaOH (I suppose in water?).

- The original aim was to prepare also other derivatives of IMD-04 (amide, hydroxamic acid). It has not been performed but it is not explained why. Can you comment on this? What methods would you use?

- The neutralization of the rection mixture after reduction with Fe was done by ammonia. Have you observed potential aminolysis of the ester bond?

- Please, can you explain the sentence (p.37): "Loss of the product was caused by overpressure and leakage"?

- Tables with reagents in Experimental section always contain the starting amount of the material, despite some additional equivalents have been sometimes added in the course of reaction. This should be somehow reflected in these tables.

- Why have you added additional (nonactivated) acid IBI-06 during synthesis of IMD-24c? The acid is not expected to react.

Evaluation of the thesis: Excellent		**For the defense: Recommend**
In Hradec Králové	8. září 2023	signature of the opponent