Přírodovědecká fakulta
Faculty of Science

Jihočeská univerzita v Českých Budějovicích
University of South Bohemia in České Budějovice

STATEMENT OF THE BACHELOR/~~DIPLOMA~~* THESIS REVIEWER

Name of the student: Anna Hehenberger

Thesis title: Derivatization study of selected steroid compounds
Supervisor: prof. Ing. Jan Tříska, CSc.

Reviewer: Ing. David Kahoun, Ph.D.
Reviewer`s affiliation: University of South Bohemia in České Budějovice
Faculty of Science, Institute of Chemistry and Biochemistry

	Point scale[1]	Points
(1) FORMAL REQUIREMENTS		
Extent of the thesis (for bachelor theses min. 18 pages, for masters theses min. 25 pages), **balanced length of the thesis parts** (recommended length of the theoretical part is max. 1/3 of the total length), **logical structure of the thesis**	0-3	3
Quality of the theoretical part (review) (number and relevancy of the references, recency of the references)	0-3	3
Accuracy in citing of the references (presence of uncited sources, uniform style of the references, use of correct journal titles and abbreviations)	0-3	2
Graphic layout of the text and of the figures/tables	0-3	3
Quality of the annotation	0-3	3
Language and stylistics, complying with the valid terminology	0-3	3
Accuracy and completeness of figures/tables legends (clarity without reading the rest of the text, explanation of the symbols and labeling, indication of the units)	0-3	2
Formal requirements – points in total		19
(2) PRACTICAL REQUIREMENTS		
Clarity and fulfillment of the aims	0-3	3
Ability to understand the results, their interpretation, and clarity of the results, discussion, and conclusions	0-3	2
Discussion quality – interpretation of results and their discussion with the literature (absence of discussion with the literature is not acceptable)	0-3	3
Logic in the course of the experimental work	0-3	3
Completeness of the description of the used techniques	0-3	2
Experimental difficulty of the thesis, independence in experimental work	0-3	3

* Choose one

[1] Mark as: 0-unsatisfactory, 1-satisfactory, 2-average, 3-excellent.

Quality of experimental data presentation	0-3	2
The use of up-to-date techniques	0-3	3
Contribution of the thesis to the knowledge in the field and possibility to publish the results (after eventual supplementary experiments)	0-3	2
Formal requirements – points in total		23
POINTS IN TOTAL (MAX/AWARDED)	48	(0-42)[2]

Suggestions and questions, to which the student has to answer during the defense:

1) Chapter 5.4.2: *"In contrast to experiment No. 1, however, 100 µL of sample and subsequently 100 µL of n-hexane for re-dissolution were used (instead on only 70 µL).".* The same changes were made in Chapter 5.4.3, but no explanation is given. Why did the author make this change?

2) Chapter 6.1: *„The chromatographic separation of the individual analytes and the internal standard was successful.".* This assessment is not made objectively. Neither objective results nor acceptance criteria are given. Please express efficiency of chromatographic separation using the parameter Chromatographic Resolution (R_s) and compare these results with general acceptance criteria of this parameter.

3) Chapter 6.3: Linearity assessment of calibration curves is not done objectively. It is not evident if these calibration curves are applicable for quantitative analyses in the full concentration range. Linearity assessment could be done using e.g. the quality coefficient (QC) in combination with the correlation coefficient (r) or using other appropriate test. It is evident that e.g. calibration curve shown in Image 10 is not applicable at least in the concentration range 0.5 – 2 µg/L. How did the author do linearity assessment and is the author sure that all calibration curves fulfil criteria for linearity in the full concentration range?

4) Chapter 6.3: All slopes of calibration curves obtained from experiment No. 3 are approx. 25x lower than slopes of calibration curves obtained from experiments No. 2 and No. 3. These results point out to the fact that response of analytes from experiment No. 3 are significantly lower (on condition that internal standard was added in the same amount). Please clarify this fact.

5) Chapter 6.3.1: There is not given which reference values are used for calculation of individual yields. Please give an example how are individual yields calculated and which reference values are used for calculation.

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[2] Enter the number of points awarded.

6) Chapter 6.3.3: The author guesses that some divergences (low response or response below the limit of detection) occurred within one set of measurement may be caused by volatilization of analyte derivatives. From my point of view this explanation is not correct because the internal standard is used. If the internal standard is chosen properly, differences in analyte derivatives amount should be compensated by differences in the internal standard amount. There is also no information about response of the internal standard in these problematic samples. Does the author have another explanation of these divergences?

Eventual mistakes, which the students should avoid in the future:

This part does not have to be read during the bachelor's thesis defence. These comments are made especially for the author.

1) Use a capital letter for a reference in the text to a table (e.g. page 4 – "*In tables 1 and 2 the results of studies...*").

2) Table captions are not left aligned (e.g. page 4, Table 1).

3) Font size of table captions are not the same as that used for the text in the rest of the thesis (e.g. page 4, Table 1).

4) Do not let any empty cell in a table (e.g. page 4, Table 1), but appropriate expression (e.g. Not Available, Not Detected etc.) or abbreviation (e.g. N/A, ND etc.) must be used.

5) Insert one space between a numerical value and its unit (e.g. page 5 – "... *about 90% of all estrogens...*").

6) Use a capital letter for a reference in the text to a figure (e.g. page 5 – "... *are shown in figure 1...*").

7) Do not leave a one-letter word at the end of a line (e.g. page 5).

8) Do not start a sentence with a number (e.g. page 7).

9) In-text citation in figure captions are not stated consistently (e.g. Figure 1 vs. Figure 2).

   When numeric referencing style with square brackets is used a caption of a figure from a website should be stated as follows:

   Figure X. Descriptive phrase that serves as title and description [reference number].

10) Chapter 5.3 Sample preparation – it is recommended (required in the regulatory sphere) to describe these operations completely because of their potential significant effect on

accuracy and precision of measurements (weight of a standard compound, volume of a volumetric flask, subsequent dilutions – volume of a standard solution vs. volume of a volumetric flask) and/or stability of a solution (storage conditions – temperature, lifetime period, material of a storage vessel etc.).

11) Chapter 5.5: Splitless period is not stated.

12) Chapter 5.5: The transfer line temperature should be higher because cholesterol is eluted at approx. 287 °C under that chromatographic conditions. The temperature should be set at least at 290 °C or optimally at 305 °C (final temperature of the GC temperature program).

13) No expression of uncertainties used in the thesis is done (e.g. Chapter 6.3.1, Figure 11). The parameter used for expression of uncertainty (e.g. standard deviation, 2x standard deviation, standard error of the mean, expanded uncertainty etc.) must be always unequivocally specified.

14) Numbers on the x-axis and/or on the y-axis have to many digits in some cases (e.g. Image 1 – 9).

15) The y-axis does not contain any name of the axis (e.g. Relative Abundance) it does not contain any unit (e.g. %).

Eventual additional comments of the reviewer on the student and the thesis:

The thesis is well-written and based on scientific publications, which demonstrates familiarity with the studied theme. The background theory has a logical connection to the research task, as well as to methodological solutions. The results are presented clearly but some assessments are not done objectively. The structure is clear, consistent and discussion is anchored to the main results. I am convinced that the thesis achieves its goals and meets all requirements for this type of work.

Conclusion:

In conclusion, I

recommend / ~~do not recommend~~*

the thesis for the defense and I suggest the grade 1 .[3]

In **České Budějovice** date **02 September 2016**.

Kahoun

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Signature

[3] You can suggest a grade, which can be modified during the defense based on the presentation. However, if the reviewer is not present at the defense, the grade will not be counted.