MUNI

Annex No. 10 to the MU Directive on Habilitation Procedures and Professor Appointment Procedures

HABILITATION THESIS REVIEWER'S REPORT

Masaryk University

Applicant	Mgr. Jiří Kohoutek, Ph.D.
Habilitation thesis	Role of transcription cyclin-dependent kinases and their cyclins in cellular processes
Reviewer	Doc. MUDr. Josef Srovnal, Ph.D.
Reviewer's home unit, institution	Institute of Molecular and Translational Medicine, Faculty of Medicine and Dentistry, Palacky University Olomouc

Review

1. General Assessment

Dr. Jiří Kohoutek's habilitation thesis presents a coherent and original body of research focused on transcription-associated cyclin-dependent kinases, namely CDK9, CDK12, CDK13, and their cyclins. The work integrates molecular, biochemical, and genetic approaches to investigate their roles in transcriptional control, development, and genome maintenance. **The thesis is based on a strong publication record** in respected peer-reviewed journals, demonstrating both the originality and impact of the research.

2. Scientific Contribution

The central theme of the thesis is the **regulation of RNA polymerase II** elongation and transcriptional responses through CDK-cyclin complexes. Dr. Kohoutek's early work clarified the regulation of P-TEFb via 7SK snRNA and HEXIM1, a now widely cited contribution. He later expanded his focus to CDK12 and CDK13, uncovering their essential roles in **DNA repair gene expression and genomic stability**. These findings have implications for cancer therapy, particularly in the context of synthetic lethality.

Notably, the thesis includes original work on mouse knockout models for CDK12, CDK13, and several cyclins. These in vivo studies reveal the indispensable functions of these factors **in embryonic development and tissue homeostasis**, providing mechanistic insight beyond cell culture models.

Overall, the thesis offers novel insights into transcriptional regulation and convincingly links molecular mechanisms to organismal phenotypes.

3. Methodological Quality

The research employs a **broad spectrum of techniques**, including chromatin immunoprecipitation, RNA-seq, biochemical fractionation, and conditional mouse genetics. The methodologies are well-chosen, appropriately applied, and sufficiently described in the

referenced publications. The combination of in vitro and in vivo approaches strengthens the validity and relevance of the findings.

4. Style, Language, and Format

The thesis is **clearly structured and well-written**. The introduction offers a concise yet comprehensive overview of transcriptional CDKs, and each chapter follows a logical progression. Scientific terminology is used appropriately, and the language is formal, precise, and grammatically correct throughout. Figures and tables are well-integrated into the text and serve as helpful visual summaries of complex concepts. Formatting is consistent, professional, and aids readability. Minor improvements could include further simplification of dense technical paragraphs, but these do not obstruct comprehension.

5. Overall Evaluation

This thesis demonstrates the **applicant's scientific maturity, originality, and leadership in the field of transcriptional regulation.** The research is conceptually sound, methodologically rigorous, and of high relevance to both basic science and clinical research. The thesis also meets all formal and academic standards in terms of presentation, writing, and organization.

Questions for the Applicant:

1. Given the significant roles of CDK12 and CDK13 in transcription elongation and genome maintenance, how do you envision selectively targeting these kinases in cancer therapy without inducing developmental toxicity?
2. Your results point to a functional divergence between cyclin T1 and T2. What mechanisms do you propose underlie their gene-selective regulation, and could this be exploited therapeutically?
3. Could you elaborate on how transcriptional pausing and release by P-TEFb integrates with chromatin state and epigenetic regulation in early embryonic development?

Conclusion

The habilitation thesis entitled "Role of transcription cyclin-dependent kinases and their cyclins in cellular processes" by Dr. Jiri Kohoutek **fulfils** requirements expected of a habilitation thesis in the field of Molecular Biology and Genetics.

Date: July 27, 2025 Signature: