

HABILITATION THESIS REVIEWER'S REPORT

Masaryk University

Applicant

PharmDr. Jakub Tremel, Ph.D.

Habilitation thesis

Natural phenolics in alleviation of symptoms and complications of type 2 diabetes mellitus

Reviewer

Doc. PharmDr. Iva Boušová, Ph.D.

Reviewer's home unit, institution

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The habilitation thesis of PharmDr. Jakub Tremel, Ph.D., titled "*Natural Phenolics in Alleviation of Symptoms and Complications of Type 2 Diabetes Mellitus*," provides a thorough summary of the key findings from his scientific and research career to date. His work centers on identifying natural phenolic compounds with antioxidant, anti-inflammatory, and antidiabetic properties that may support the management of hyperglycemia in individuals with type 2 diabetes and help reduce long-term complications of the disease.

The habilitation thesis spanning 217 pages is structured as a commented collection of scientific papers published in peer-reviewed journals indexed in the Web of Science database, with the candidate's specific contributions to each work clearly noted. The thesis is presented in a 37-page annotated overview, followed by 12 of the candidate's publications (10 scientific papers and 2 reviews) published between 2010 and 2025. Dr. Tremel was the first and/or corresponding author on 8 scientific papers, 5 of which were published in journals with Q1.

The thesis is divided into two main parts. The first part briefly discusses the pathophysiological mechanisms underlying the development of chronic complications of type 2 diabetes mellitus (T2DM) and their treatment, emphasizing oxidative stress as a key factor in the progression of these complications. The second part examines the antioxidant, anti-inflammatory, and anti-diabetic properties of natural phenolic compounds (primarily prenylated flavonoids) with particular attention to the geranylated flavanone diplacone, which combines all three of the aforementioned effects. In discussing antioxidant effects, the candidate highlights the discrepancies between results obtained from chemical-based and cell-based in vitro assays. The final section examines the encapsulation of natural products (including curcumin, trans-resveratrol, and diplacone) into glucan particles and various nanoparticle delivery systems to overcome limitations such as low water solubility, poor bioavailability, and limited metabolic stability.

In my opinion, the habilitation thesis is of high quality - well written and logically structured. Nevertheless, a more extensive comparison of the findings with the existing literature would further enhance the overall quality of the thesis. Dr. Tremel demonstrates a broad scientific scope and substantial experience in the study of the biological activity of natural phenolic compounds. His scientific excellence is further supported by long-standing collaborations with both Czech and international research institutions.

Reviewer's questions for the habilitation thesis defense

1. On page 14, the thesis outlines key factors involved in the progression of chronic complications of T2DM. Patients suffering from T2DM are typically obese and show elevated circulating levels of free fatty acids. Could you discuss whether these free fatty acids may contribute to the development of chronic complications in T2DM, and through which mechanisms such effects might be mediated?
2. In the paragraph discussing the absorption of phenolic compounds in the small intestine (page 19), you state that "*Other compounds are actively transported via P-glycoprotein or sodium-glucose cotransporters, such as quercetin glycosides*" which appears to contradict the established understanding that P-gp functions as an efflux transporter rather than an influx mechanism. Could you clarify how P-gp interacts with plant phenolics in the small intestine, and discuss its overall physiological significance in regulating their absorption, metabolism, and systemic availability?
3. In the article 5, you state that diplacone significantly increases the protein expression and enzymatic activity of catalase, while mRNA expression remains unchanged. At the same time, you demonstrate that this flavonoid does not activate the key Nrf2-ARE signaling pathway, which is considered the main regulatory mechanism for catalase expression in response to oxidative stress. Can you please explain by which alternative molecular mechanisms could the induction of catalase expression/activity occur?
4. In article 6 the authors report that compound 10 (5,7,3',4'-tetrahydroxy-3-methoxy-8,5'-diprenylflavone) produced the greatest antioxidant effect in the CAA assay and significantly inhibited NF- κ B/AP-1 activation, yet it had no effect on IL-1 and TNF- α expression - an observation the authors attribute to a primary effect on AP-1. Could you propose an experimental strategy that would unambiguously distinguish whether compound 10 acts via inhibition of the NF- κ B pathway, the AP-1 pathway, or both?
5. What are the prospects and limitations for the use of natural phenolic substances alleviation of symptoms and complications of T2DM?

Conclusion

The habilitation thesis entitled "*Natural Phenolics in Alleviation of Symptoms and Complications of Type 2 Diabetes Mellitus*" by PharmDr. Jakub Trembl, Ph.D., **fulfils** requirements expected of a habilitation thesis in the field of Molecular Biology and Genetics.

Date: 19 November 2025

Signature: