

Opponent's report on the habilitation thesis of **Ing. Antonín Kouba, Ph.D.**
entitled **Marbled crayfish, an emerging invasive species**

The habilitation thesis of **Ing. Antonín Kouba, Ph.D.** deals with different aspect of biology of the marbled crayfish *Procambarus virginalis*, but widely comments on the actual state and perspectives of distribution of non-indigenous crayfishes in Europe and their invasive potential. The thesis is composed of general introduction (10 pp.), summary of publications (3 pp.), future perspectives (1 p.), and references cited in the general introduction (10 pp.) covering among many others also 23 studies by A. Kouba (2 as the first author, and 9 with A. Kouba in the senior position). Six selected publications representing the main milestones of the research leading from crayfish invasions, to the original reseraches on survival and breeding of invasive crayfish species are attached as Studies I - VI.

The habilitation thesis proves that A. Kouba is a well-established and experienced researcher showing his ability to both personally realize and publish research projects, as well as to coordinate team research and lead students.

The Introduction of the habilitation thesis starts from the phenomenon of current biological invasions as the major threats to global biodiversity and ecosystem functioning; in freshwater ecosystems, the crayfish plague associated (in Europe) with non-indigenous crayfish species is among the most important factors negatively affecting natural ecosystems. The major volume of the introduction is thus devoted to a review of non-native crayfish species, their biology, distribution, and invasive potential, while the final section is devoted to the target species of the habilitation thesis, the marbled crayfish, pointing to its unique reproduction by apomictic parthenogenesis, the history of its introduction in Europe, and a review of its invasive potential.

The thesis is well written and presents a good insight to the problematics for readers. I found only several minor errors or typos. Generally, I do not understand why the author did not follow the standard chronological order of multiple citations in the text (instead of alphabetical order used); for me, such style of citations obscures their historical value. But all those are just minor remarks not diminishing the value of the thesis.

The summary of publications provides short insights to the six studies presented in the habilitation thesis. These studies themselves are all papers published in journals covered by the Web of Science, thus underwent proper peer-review process proving respective quality of those studies. The originality of the presented studies is highlighted mainly in experimentally confirmed results on the marbled crayfish ability to overwinter in the temperate zone of Europe, and on reproduction capacities of *Procambarus clarkii* under conditions of severe drought.

Based on all mentioned above, I consider the submitted habilitation thesis of dr. Antonín Kouba for clear proof that the applicant is a prominent person in Czech astacology, who undoubtedly deserves the scientific-pedagogical title of associate professor.

In Ostrava, 10 Jan. 2021

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Questions, or remarks:

- In the study V (Introduction, p.1, Results, p.2), there is a statement, that "The [European – rem. opan.] native species and *Pacifastacus leniusculus* exhibited no ability to construct vertical burrows". This seems to be the truth. But, the study deals only with the fact that invasive [in Europe] species can survive severe droughts by means of creating vertical burrows with high humidity inside. It does not discuss another possible phenomenon applicable to at least one European native species, *Astacus astacus*, which may survive such drought by vertical burying itself into gravel bottom (personal observation). Thus, not by constructing defined vertical hollows in the bottom with upper opening, but by digging itself deeply into wet gravel, even in multiple specimens. Both strategies, burrow construction and burying, might provide comparable protection against body desiccation, if not even more effective in the second case. **Did you meet such case in this, or other native, or non-native, crayfish species?**
- Sophisticated study requires sophisticated methods. But not always... I found a slightly funny the methodological approaches dealing with burrow „biometry“. I very agree with fabrication of solid casts of the burrow, but I do not think that there was so strong necessity to measure obtained gypsum casts „by a digital caliper to the nearest mm (p. 5)“ ... measurement by simple scale might provide the same result....But why not, if such caliper is available. ... But, further is more interesting (same page): "Casts were further scanned by an Artec Spider™ hand-held 3D laser scanner (Artec Group, Luxembourg) located at the Department of Cybernetics, Faculty of Applied Sciences, University of West Bohemia in Pilsen with a stated resolution of 0.1 mm and accuracy up to 0.03 mm. The scanner ... provides a 3D mesh of the object ... The resulting STL (STereoLithography) mesh was imported to the Artec Studio, version 10 (Artec Group, Luxembourg) where the volume of the 3D mesh **was calculated**"...- **I understand that such sexy methodology might help in peer review process. My question is - would not it be much easier to measure the cast volume in scaled glass cylinder with a water? Are you sure that such high progressive methodology was applied in gypsum casts depth and volume?**
- In the study VI you limited your experiment (egg incubation in high air humidity) to the period of 7 days. **Was the length of the period tested by any preliminary experiments?**