A consensual Diving-PAM protocol to monitor *Posidonia oceanica* photosynthesis

Gobert Sylvie¹, Lepoint Gilles¹, Silva João², Santos Rui², Lejeune Pierre³, du Jardin Patrick⁴, Delvaux Bruno⁵, Cornelis Jean-Thomas⁶, Richir Jonathan^{1,3*}

¹Laboratory of Oceanology, MARE Centre, University of Liege, Sart Tilman, Belgium

² Marine Plant Ecology Research Group, Center of Marine Sciences (CCMar), University of Algarve, Faro, Portugal

³STARESO SAS, Pointe Revellata, Calvi, France

⁴ Unit of Plant Biology, Gembloux Agro-Bio Tech, University of Liege, Gembloux, Belgium

⁵ Earth and Life Institute, Environmental Sciences, Catholic University of Louvain-la-Neuve, Louvain-la-Neuve, Belgium

⁶ Biosystem Engineering Department, Gembloux Agro-Bio Tech, University of Liege, Gembloux, Belgium

* jonathan.richir@alumni.ulg.ac.be

The seagrass *Posidonia oceanica* is widely recognized as an effective bioindicator of the health status of Mediterranean coastal waters. Chlorophyll fluorescence measurements, in particular through the Pulse Amplitude Modulated (PAM) fluorometry method, are performed to study aquatic plant ecology and vitality and to assess their responses to diverse stressful factors. However, the current understanding of *P. oceanica* photosynthetic responses to environmental stresses does only allow scientists to use the PAM-method as a complementary tool to other more-robust monitoring techniques. Consequently, a more in-depth knowledge of the natural causes of variability of *P. oceanica* photosynthetic responses are a prerequisite to any surveys relying on that time and cost-effective method. In the framework of the STARECAPMED project, this work aimed to determine the influence of several environmental (depth, daytime, season) and plant-specific characteristics (leaf age, leaf part analyzed, epiphytic coverage) on the photosynthetic responses (Y, ETR, RLC) of P. oceanica. Water temperature, irradiance and several biochemical parameters of the seagrass (chl.a, chl.b, C, N, P, micronutrients such as Fe, Cu) were measured as well. The field survey was performed in a pristine meadow in the Calvi Bay, Corsica. Environmental and plantphysiological characteristics deeply influenced P. oceanica photosynthetic responses. As an example, ETR decreased with depth, contrary to Y that mostly increased. ETR was lower in the basal part of leaf blade, and the epiphytic coverage of leaf tips slightly increased their ETR compared to leaf tips cleaned of epiphytes. Depth and leaf part-related variations in RLC were also observed. Because of this natural variability, it appears essential to develop a consensual protocol of chlorophyll fluorescence measurements to publish reliable and comparable results between studies. We therefore notably suggest to perform measurements close to

midday, when photosynthetic responses are the highest; at 10-15 m depth in order to avoid, among others, low depth light irradiance variability; on the middle part of the 3rd-4th external leaf, well developed, highly photosynthetic, and little epiphyted. Finally, because *P. oceanica* fluorescence was correlated with N, P and chl.*b* leaf contents, the PAM-method could afterwards be used as bioindicator technique, according to the consensual protocol proposed.