PRACTICAL APPLICATIONS OF CHLOROPHYLL FLUORESCENCE IN PLANT BIOLOGY

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PREFACE

The technique of chlorophyll fluorescence has a relatively short history, beginning with the observations by Kautsky (Kautsky and Hirsch, 1931). Since that time there have been several reviews devoted to the subject, with most of them highly theoretical (Bohlàr-Nordenkampf and Öquist, 1993; Dau, 1994; Schreiber et al., 1994). There have also been many books devoted to generalized spectrophotometric and microscopic fluorescence techniques. However, to the best of our knowledge there has not been a book completely devoted to the practical applications and uses of chlorophyll fluorescence in plant biology. As techniques mature, applications multiply and so do their potential advantages. The chlorophyll fluorescence technique is maturing as can be seen in the increasing numbers of publications that are devoted to its use. Therefore, we considered that now was a good time to compile the existing knowledge for the applied use of this technique and provide a single volume to which a novice or experienced user could refer.

Highly trained experts in the field of photobiology have primarily used the chlorophyll fluorescence technique in the past. In that work, understanding the mechanisms and controls of the photosynthetic processes was the main focus of activity and discussion. Much of the equipment used was highly specialized and expensive, or in some cases one-of-a-kind lab designed units. However, the development of several reliable commercially available chlorophyll fluorescence monitoring instruments has changed the potential user base for the technique. There has been a review of chlorophyll fluorescence instrumentation that discusses the features, potential and limitations of many of these instruments (Mohammed et al., 1995). One important feature in most commercial instruments is that they have pre-programmed protocols for taking measurements, making the technique accessible to novices in the field of photobiology. However, taking measurements without a basic understanding of the theoretical aspects of the technique can lead to inappropriate interpretation or poor results. This book has been designed to acquaint the novice user of the chlorophyll fluorescence technique with essential background theory, and some examples of applied uses for the technique, with cautions regarding potential pitfalls.

As this book will demonstrate, there have been numerous developments in the instrumentation and approaches for use of chlorophyll fluorescence as a probe to plant adaptation to an environment or as an indicator of the level of stress. The advantage of chlorophyll fluorescence over many techniques that have been used is that it provides rapid and nondestructive measures. As such, more measurements can be taken and data processing is quite simple. However, this technique, like others, is not a miracle approach. It will be demonstrated in the following chapters that one

must understand some basic theory and must also accept the fact that unless experiments are designed to provide specific response measures, fluorescence cannot provide a simple approach to inferring underlying causes for the physiological status of a plant.

The first two chapters are devoted to provide a clear, understandable explanation of the theoretical basis for chlorophyll fluorescence analysis. The definitions and terminology that are specific to chlorophyll fluorescence analysis are included in this discussion. In addition, the discussion leads to the bridging of chlorophyll fluorescence analysis to plant tissue condition or status. These two chapters should give a reader a solid background as to how and why chlorophyll fluorescence is used. Subsequent chapters focus on the monitoring of stress in the natural terrestrial and aquatic environments, assessing seedling quality in forestry, and postharvest quality in fruits and vegetables. A final chapter is devoted to a newly emerging use for the technique in plant breeding programs. These chapters should provide the reader with good examples for specific approaches in a variety of applied plant science studies. The content of these chapters will also demonstrate the versatility of the technique and will hopefully encourage the development of new uses that are not reported in this book.

It is the hope of the editors and authors that readers who have not used the technique will be encouraged to explore the possibilities in their area of study. For those who have used the technique previously, we hope that this book will offer some new insights, which may encourage development and/or refinement of approaches. Ultimately, we hope that the contents of this work will contribute in some manner to advances in the understanding of plant-environment interactions and hence to improvements in environmental quality, as well as in forestry and agri-food industries.

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CHLOROPHYLL FLUORESCENCE NOMENCLATURE

F	actual fluorescence intensity at any given time
Fo	minimal fluorescence in dark-adapted tissue; fluorescence intensity with all PSII reaction centers open while the photosynthetic membrane is in the non-energized state ($q_P = 1$ and $q_N = 0$); it can also be used for the O level in Kautsky nomenclature
Fi	fluorescence intensity at the I level in Kautsky nomenclature
Fp	fluorescence intensity at the P level in Kautsky nomenclature
Fm	maximal fluorescence in dark-adapted tissue; fluorescence intensity with all PSII reaction centers closed ($q_P = 0$), all non-photochemical quenching processes are at a minimum ($q_N = 0$)
Fv	variable fluorescence in dark-adapted tissue; maximum variable fluorescence in the state when all non-photochemical processes are at a minimum, i.e. Fm-Fo
Ft	fluorescence intensity at the T level in Kautsky nomenclature
Fs	fluorescence in steady state; defined by an author as a period within which the fluorescence intensity does not change while the external circumstances remain constant
Fv/Fm	exciton transfer efficiency in dark-adapted tissue; (Fm-Fo) / Fm
T½	half-time for rise in Fv; time (msec) taken for fluorescence intensity from O level to reach half the fluorescence intensity of the Fv level.
Fo'	minimal fluorescence in light-adapted tissue; fluorescence intensity with all PSII reaction centers open in any light adapted state ($q_P = 1$ and $q_N \geq 0$)
Fm'	maximal fluorescence in light-adapted tissue; fluorescence intensity with all PSII reaction centers closed in any light adapted state ($q_P = 0$ and $q_N \geq 0$)

Fv'	variable fluorescence in light-adapted tissue; maximum variable fluorescence in any light adapted state, i.e. Fm'-Fo'
Fv'/Fm'	exciton transfer efficiency in light-adapted tissue; (Fm'- Fo') / Fm'
q_P	photochemical quenching; (Fm'-F) / (Fm'-Fo')
q_N	non-photochemical quenching; 1- (Fm'-Fo') / (Fm-Fo)
Φ_{PSII}	quantum yield of photochemistry; (Fm'-F) / Fm'