Available online at www.sciencedirect.com
SCIENCE DIRECT
ELSEVIER
Scientia Horticulturae 105 (2005) 197–211
SCIENTIA HORTICULTURAE
www.elsevier.com/locate/scihorti

Net CO_2 exchange rate of in vitro plum cultures during growth evolution at different photosynthetic photon flux density

S. Morini *, M. Melai

Dipartimento di Coltivazione e Difesa delle Specie Legnose "G. Scaramuzzi", Sezione di Coltivazioni Arboree, University of Pisa, Via del Borghetto 80, 56124 Pisa, Italy

Received 25 September 2003; received in revised form 29 September 2004; accepted 6 January 2005

Abstract

CO_2 concentration was monitored during three 15-day subculturing cycles in vessels containing actively proliferating plum cultures of *Prunus cerasifera*, clone Mr.S. 2/5. The effects of two photosynthetic photon flux density regimes: 50 ± 5 μmol m^{-2} s^{-1} and 210 ± 5 μmol m^{-2} s^{-1} were compared. Three distinct phases in the CO_2 trend were distinguished during each culturing cycle of both light treatments. In the first, occurring at the beginning of the culture cycle, the amount of CO_2 emitted by the cultures during dark periods was greater than that assimilated during the light periods. In the second phase, the opposite trend was detected, while in the third, the range of CO_2 day–night fluctuations increased or remained stable according to the number of explants per vessel. The treatment with 210 ± 5 μmol m^{-2} s^{-1} did not modify the CO_2 phase trend but induced more pronounced fluctuations in day–night CO_2 concentration. Under this light treatment, cultures reached CO_2 compensation point for a period as long as 48% of the total number of light hours, while under 50 ± 5 μmol m^{-2} s^{-1}, it was only 8%. The different range in CO_2 day–night fluctuations monitored throughout a subculturing cycle, appeared to be mainly induced by changes in culture growth dynamics.

Keywords: CO_2 dynamics; CO_2 monitoring equipment; Light intensity; Micropropagation; *Prunus cerasifera*

Abbreviations: BA, 6-benzylaminopurine; GA_3, gibberellic acid; IBA, indole-3-butyric acid; NaFeEDTA, ethylenediaminetetra-acetic acid ferric monosodium salt; PPFD, photosynthetic photon flux density

* Corresponding author. Tel.: +39 050 571551; fax: +39 050 544420.
E-mail address: smorini@agr.unipi.it (S. Morini).

0304-4238/$ – see front matter © 2005 Published by Elsevier B.V.
doi:10.1016/j.scienta.2005.01.016

1. Introduction

Several studies have demonstrated that the photosynthetic capacity of cultures in micropropagation is strongly influenced by the photosynthetic photon flux density (PPFD) and CO_2 availability inside the culture vessel. In commercial laboratories, light is generally applied at roughly 40–50 μmol m^{-2} s^{-1}, often too low to optimize photosynthesis, while CO_2 supplying to the cultures is hampered by the vessel closure type that reduces or prevents gas exchange with the external atmosphere and is the cause of accumulation or depletion of various gaseous compounds mainly produced or utilized by the cultures (Melé et al., 1982; De Proft et al., 1985; Righetti et al., 1987, 1990; Blazkovà et al., 1989; Woltering, 1989; Adkins et al., 1990; Jackson et al., 1991; Buddendorf-Joosten and Woltering, 1994). Increasing PPFDs increase the capacity of the cultures to assimilate CO_2, which in turn may become a limiting factor for photosynthesis (Infante et al., 1989; Kozai, 1991; Pospisilova et al., 1992, 1997; Righetti et al., 1993; Navarro et al., 1994). Increasing both PPFD and CO_2 availability by exogenous enrichment, photosynthesis can be optimized and photoautotrophic growth of the culture induced (Kozai et al., 1990; Predieri et al., 1991; Righetti et al., 1993; Fournioux and Bessis, 1993; Deng and Donelly, 1993; Jin et al., 1993; Hdider et al., 1994; Navarro et al., 1994; Figueira and Janick, 1994; Morini et al., 1999).

In conventional micropropagation procedures, CO_2 concentration inside the culture vessels fluctuates as a result of the photosynthetic and respiratory processes. Thus, the most important factor that determines variations in CO_2 concentration is the succession of light to dark periods. During darkness, CO_2 produced by dark respiration of the culture accumulates inside the vessel, while during light periods if PPFD is sufficiently high CO_2 is promptly assimilated and often depleted in just a few hours (Fujiwara et al., 1987; Infante et al., 1989; Solárová, 1989; Righetti et al., 1993; Pospisilova et al., 1997).

The information available today on CO_2 day–night fluctuation has been acquired in general by daily point determinations of CO_2 concentration over a long period of culture (Infante et al., 1989; Righetti et al., 1990, 1993; Navarro et al., 1994) or by a number of determinations of CO_2 concentration performed during a light–dark cycle only, or repeated at different culture age, or without any specification about the time the determinations were referred to (Fujiwara et al., 1987; Kozai and Sekimoto, 1988; Kozai and Iwanami, 1988; Solárová, 1989; Navarro et al., 1994; Baroja et al., 1995; Fujiwara and Kozai, 1995). No information has been acquired about possible daily variations in photosynthetic and dark respiration activities that could occur as a consequence of culture growth evolution over a subculture period that may be as long as roughly 15–20 days. During this period, the cultures pass throughout different growth processes such as differentiation and outgrowth of neoformed buds, shoot development, leaf enlargement, up to culture ageing processes, which could be responsible for different CO_2 culture metabolisms. Deeper studies on this aspect may be of interest to improve knowledge on the eco-physiological conditions that are established inside hermetically closed vessels like those used in the micropropagation laboratories. The methodological approach for performing experiments in this field must take into account the use of monitoring equipment allowing frequent daily determinations of CO_2 concentration over prolonged time periods to detect even small variations that could be missed by brief point determinations.