

DUAL-DPM

Photomultiplier Detector

User's Instruction

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Heinz Walz GmbH • Eichenring 6 • 91090 Effeltrich • Germany
Phone +49-(0)9133/7765-0 • Telefax ++49-(0)9133/5395
E-mail info@walz.com • Internet www.walz.com

DUAL-DPM

Photomultiplier Detector Module

DUAL-PAM-100 accessory for high sensitivity
fluorescence measurements



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1 General Description of DUAL-DPM

The photomultiplier detector module DUAL-DPM has been designed for applications which require highest sensitivity in fluorescence signal detection.

For chlorophyll fluorescence detection, the signal cable of the DUAL-DPM (labeled "DETECTOR1") is plugged into the detector 1 socket of the DUAL-C Power-and-Control-Unit. Modulated fluorescence excitation and actinic light is provided by DUAL-DB or DUAL-DR measuring heads.

Typically, the DUAL-DPM is mounted at right angle with respect to fluorescence excitation by the DUAL-DB or DUAL-DR (Fig. 1A). The right-angled set-up reduces the amount of measuring light reaching the photomultiplier and, thus, the level of artifactual background signals.

Also, the right-angled orientation permits simultaneous P700 absorption measurements which require that infrared measuring light emitter (DUAL-E) and infrared light detector (DUAL-DB or DUAL-DR) are oriented in line (compare Fig. 2B).

For concomitant recording of fluorescence by the DUAL-DPM and P700 absorption changes, the signal cable of the DUAL-DB or DUAL-DR head (labeled "DETECTOR") is connected to the detector 2 socket (Fig. 1B).

Frequently, however, the low chlorophyll concentrations, which require fluorescence detection by the DUAL-DPM, are too small for high-quality P700 measurements: Chl contents of 20 mg Chl/l are required for P700 measurements of reasonable signal/noise ratios,

For measurements with suspensions, the DUAL-DPM housing is connected to a special port of the optical unit ED-101US/MD

Fig. 1A Measuring Set-up Using the DUAL-DPM

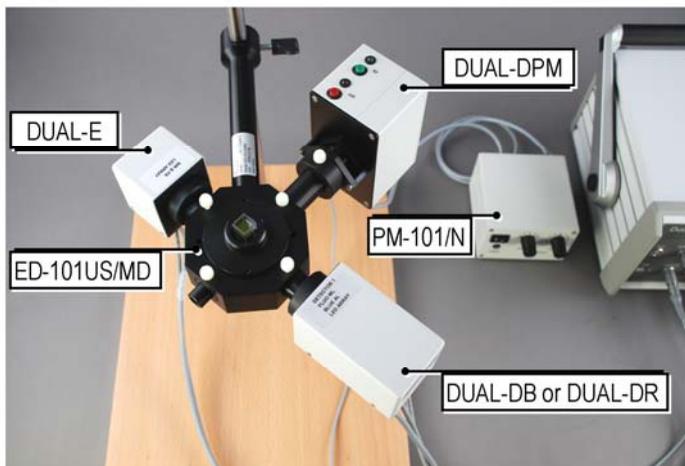
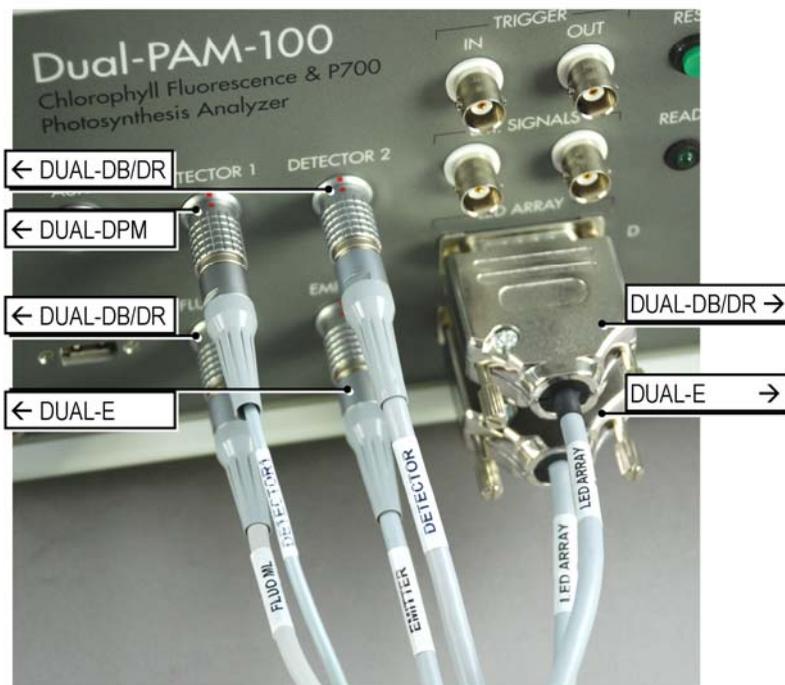


Fig. 1B Connections



which contains a Perspex rod. The Perspex rod serves to carry the fluorescence from the 10 x 10 mm cuvette to the detector.

It is important that ED-101US/MD ports which are not used are closed to prevent external light from reaching the interior of the optical unit.

1.1 Description of Parts

1.1.1 DUAL-DPM Housing

The DUAL-DPM housing features a cable connecting to the DETECTOR 1 input of the DUAL-C Power-and-Control-Unit, and a socket to plug in the power cable.

A filter slot is situated in front of the housing such that filters of variable size and shape can be inserted between the light-guiding rod and the PM window (Fig. 2).



Figure 2. DUAL-DPM. Filter slot with filter in place.

To prevent damage to the photomultiplier by excessive light exposure, the photomultiplier is automatically switched off at too high light intensities and the red LED on top of the housing lights up. The photomultiplier is switched on again by pushing the green button: the green LED lights up. The photomultiplier can also be manually turned off by the red push-button.

The circuit board of the DUAL-DPM features a special preamplifier for the pulse-modulated fluorescence signal. The actual photodetector consist of an 8 mm diameter side-on photomultiplier tube with a high voltage power supply assembled in a compact aluminum housing (Hamamatsu H6779-20 Photo-sensor Module). This photosensor is outstanding because of its high sensitivity, excellent output linearity, and compact size.

Table 1. DUAL-DPM Power Supply: Settings and Signal

The table shows data obtained with a typical prototype. All data are relative to the signal amplitude at settings COARSE=1 and FINE=0.

| Setting COARSE | Setting FINE | Signal amplitude | Setting COARSE | Setting FINE | Signal amplitude |
|-------------------|-----------------|---------------------|-------------------|-----------------|---------------------|
| 1 | 0 | 1 | 3 | 0 | 57 |
| 2 | 0 | 12 | 3 | 1 | 63 |
| 3 | 0 | 57 | 3 | 2 | 69 |
| 4 | 0 | 123 | 3 | 3 | 75 |
| 5 | 0 | 168 | 3 | 4 | 80 |
| 6 | 0 | 202 | 3 | 5 | 87 |
| 6 | 11 | 227 | 3 | 6 | 93 |
| | | | 3 | 7 | 99 |
| | | | 3 | 8 | 105 |
| | | | 3 | 9 | 111 |
| | | | 3 | 10 | 117 |
| | | | 3 | 11 | 124 |

1.1.2 DUAL-DPM Power Supply

The power supply has a cable connecting to the DUAL-DPM housing. By adjusting the COARSE and FINE switches in front of the power supply housing, the photomultiplier sensitivity is adjusted. The relative sensitivity factor can be varied from 1 to 230 depending on adjustment (Table 1). Note that sensitivity and settings are not strictly linearly related.

2 The DUAL-DPM: Operation

2.1 Getting started

Connect power supply to DUAL-DPM housing. Select appropriate voltage using the AC VOLTAGE switch on the back of the housing. Connect power supply to line power. If the red LED lamp lights up right after the power supply is turned on, the photomultiplier must be switched on manually by pressing the green push button. During use, the photomultiplier is switched off automatically when it receives too much light or when the voltage applied is too high.

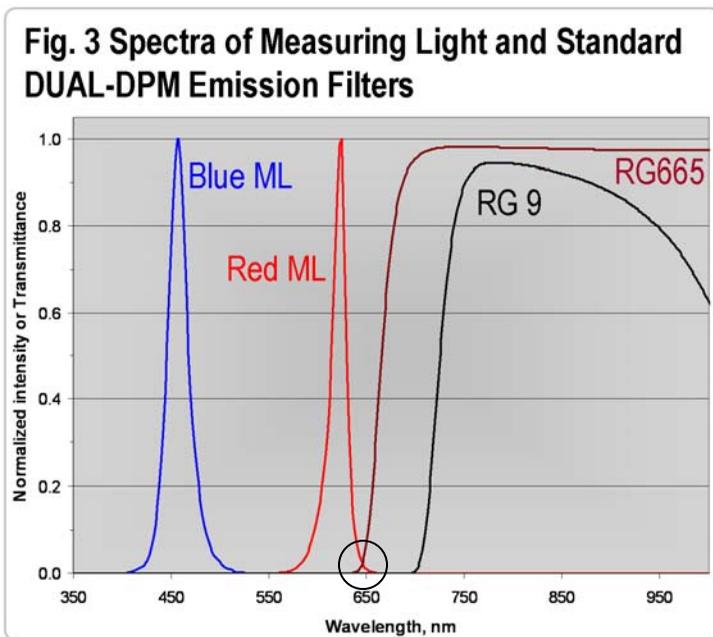
Particular care must be taken that the photomultiplier is not exposed to ambient light because the repeated exposure to high light intensities can result in a significant decrease in the signal/noise ratio of the DUAL-DPM unit.

2.2 Filters

Two long pass glass filters are delivered with the DUAL-DPM unit: the RG665 and the RG9 filters. These filters are needed to screen the photomultiplier from measuring and actinic light. The transmittance flank of the RG665 filter is situated at shorter wavelengths than that of the RG9 filter (Fig. 3). In

samples with low chlorophyll content the fluorescence emission maximum is located about 680 nm but at high chlorophyll concentrations, like in green leaves, the 680 nm peak is small because of reabsorption of chlorophyll fluorescence. Hence, at low chlorophyll concentrations, the RG665 filter can improve the signal because it transmits fluorescence of the emission maximum.

The RG665 cannot not be used in conjunction with red measuring light (DUAL-DR) or red actinic light because emission spectra of LEDs and the transmittance spectrum of the RG665 filter overlap (see: Fig. 3, circle). Instead, blue measuring light (DUAL-DB) and blue actinic light should be used.



2.3 Lightproof set-up

For best results, prevent ambient light from reaching the interior of the optical unit ED 101US/MD. It is strongly recommended to consider subsequent recommendations:

- Do not operate the DUAL-DPM at high light-exposed sites.
- During measurements, cover the cuvette holder by the black cuvette lid, and the filter slot by the triangle-shaped shield (both covers are part of the DUAL-DPM package).

3 Comments relating to the optimal use of the DUAL-DPM

The use of a photomultiplier instead of a photodiode detector is advantageous only in applications with very small signals because, contrary to the photodiode, the photomultiplier shows an increase of noise with increasing signal amplitude.

Actually, this can be a disadvantage of the photomultiplier in particular when saturating light pulses are applied for chlorophyll fluorescence quenching analysis: as the saturation light pulses are much more intense than the modulated measuring light, the pulse leads to a substantial increase in noise and too high sensitivity (or chlorophyll concentrations) may even result in automatic switch off of the photomultiplier.

To make optimal use of the photomultiplier for chlorophyll quenching analysis at very low chlorophyll concentrations the following points should be considered:

- Do not apply saturating pulse intensities higher than necessary to induce F_m .

- If the photomultiplier automatically switches off during application of a saturation pulse, a lower sensitivity setting should be selected.
- At low chlorophyll concentrations, non-specific background signals become a problem. To reduce background signals, clean surfaces of the cuvette and the Perspex rods. Also, avoid suspension media which show a fluorescence signal in the absence of chlorophyll.
- Always determine the background signal with an appropriate blank and consider the background level for quenching analysis.

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