PROSOPE

H. CLAUSTRE : head of mission and project leader

**PHOSPHATE ACTIVITY : A. VAN WAMBEKE**

Method | References | Data

*Alkaline phosphatase activity*

# *METHOD*

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Alkaline phosphatase activity (APA) was measured fluorometrically using methyl-umbelliferone phosphate (MUF-P). Stock solutions of 5 mM in methylcellosolve were stored at –20°C and diluted with boiled-filtered sea water prior to use. The increase in fluorescence was measured during the linear increase with time over 9 hours, with excitation set at 365 nm and emission at 460 nm, with a Kontron SFM 23B spectrofluorometer. Calibration curves were made with MUF standards. On the horizontal transect, we ran concentration kinetics using a concentration set from 25 to 1000 nM. The following transformation of the Michaelis –Menten equation was used to determine kinetic coefficients :

$S/V = (K+Sn)/Vm + S/Vm$

Plotting turnover time S/V versus S resulted in a straight line from which the in situ turnover time (TT) is the crossing point with y axis, the maximum hydrolysis rate Vm is the inverse of the slope and the sum of the half saturation constant plus the natural concentration K+Sn is the crossing point with the x-axis.

Due to the number of parameters to be processed simultaneously on the vertical profiles of the stations DYF and MIO, APA activity was measured with only one concentration added. We used a trace concentration (25 nM) of MUF-PO4 which insurred appropriate conditions for estimating in situ turnover time (TT). Indeed, TT measured with 25 nM MUF-P additions was not different from the value obtained from concentration kinetics (comparison made on surface layers of the stations of the transect with TT ranging 14 to 182 h, n=11, slope = 1.05, $r^2$ =0.99).

# *References*

Van Wambeke F, Christaki U, Giannakourou A, Moutin T, Souvemerzoglou S.Longitudinal and vertical trends of bacterial limitation by phosphorus and carbon in the Mediterranean Sea. Microbial Ecology, submitted

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# *Data set*

| n° CTD | station | bout n° | depth | TT APA | Vm APA | Km APA |
|---|---|---|---|---|---|---|
| | | | m | h | nM/h | nM |
| 2 | UPW | 15 | 15 | 786 | 1.1 | 848 |
| 11 | MED1 | 18 | 11 | 135 | 1.2 | 168 |
| 14 | MED 2 | 18 | 15 | 182 | 1.0 | 175 |
| 17 | MED 3 | 18 | 10 | 114 | 1.8 | 206 |
| 20 | MED 4 | 18 | 12 | 51 | 2.6 | 133 |
| 23 | MED 5 | 18 | 11 | 52 | 1.6 | 86 |
| 26 | MED 6 | 18 | 10 | 31 | 2.6 | 83 |
| 28 | MIO 1 | 18 | 15 | 27 | 4.0 | 111 |
| 63 | MED 7 | 18 | 14 | 11 | 7.9 | 88 |
| 68 | MED 8 | 18 | 15 | 21 | 4.6 | 95 |
| 70 | MED 9 | 18 | 15 | 19 | 6.0 | 114 |
| 73 | DYF 1 | 18 | 10 | 14 | 12.6 | 178 |
| 33 | MIO2 | 20 | 5 | 31 | | |
| 33 | MIO2 | 18 | 14 | 25 | | |
| 33 | MIO2 | 16 | 30 | 34 | | |
| 33 | MIO2 | 13 | 50 | 117 | | |
| 33 | MIO2 | 10 | 70 | 203 | | |
| 33 | MIO2 | 8 | 90 | 249 | | |
| 33 | MIO2 | 6 | 100 | 372 | | |
| 33 | MIO2 | 4 | 131 | 438 | | |
| 78 | DYF2 | 20 | 4 | 15 | | |
| 78 | DYF2 | 18 | 9 | 16 | | |
| 78 | DYF2 | 16 | 19 | 24 | | |
| 78 | DYF2 | 12 | 29 | 66 | | |
| 78 | DYF2 | 10 | 39 | 53 | | |
| 78 | DYF2 | 8 | 47 | 150 | | |
| 78 | DYF2 | 6 | 60 | 298 | | |
| 78 | DYF2 | 4 | 70 | 2674 | | |