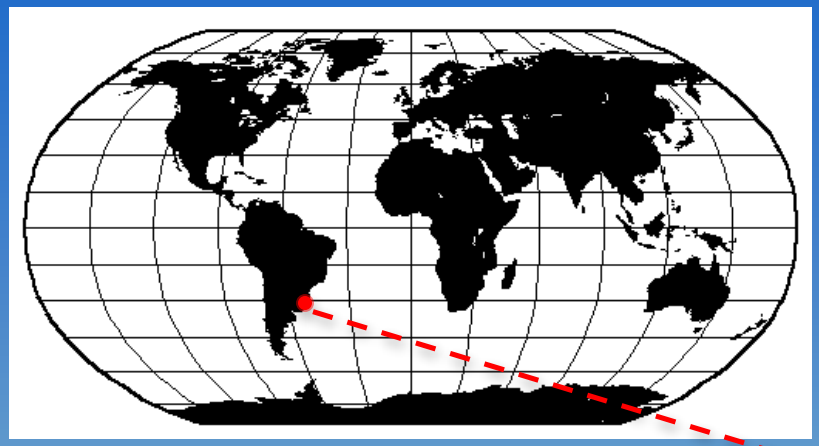
HPLC pigment measurements at Federal University of Rio Grande (FURG), Brazil

Virginia M.T. Garcia

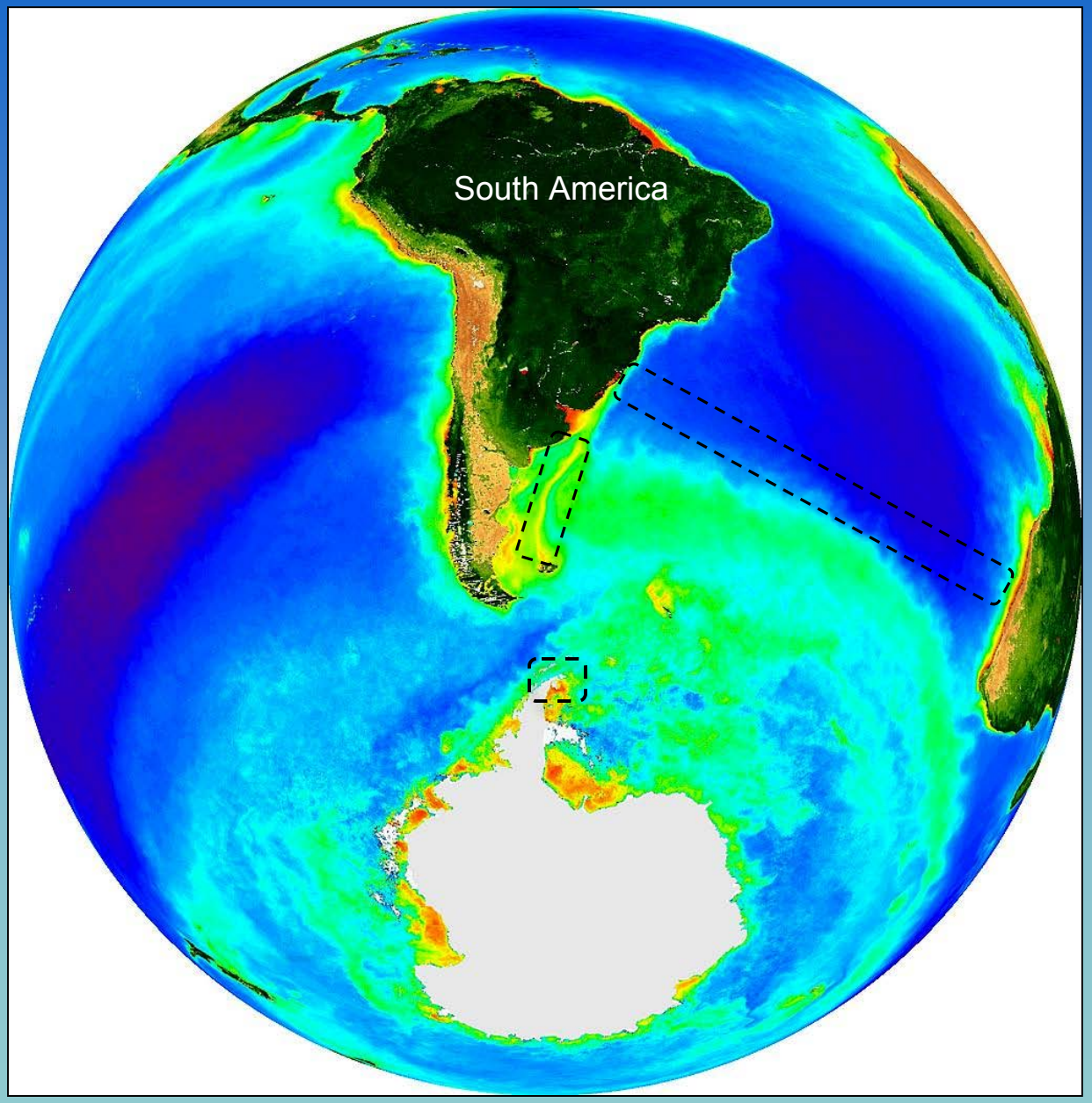
FURG Location in South Brazil



THE EQUIPMENT
"Shimadzu Prominence" HPLC
(to be used mainly in research/monitoring)

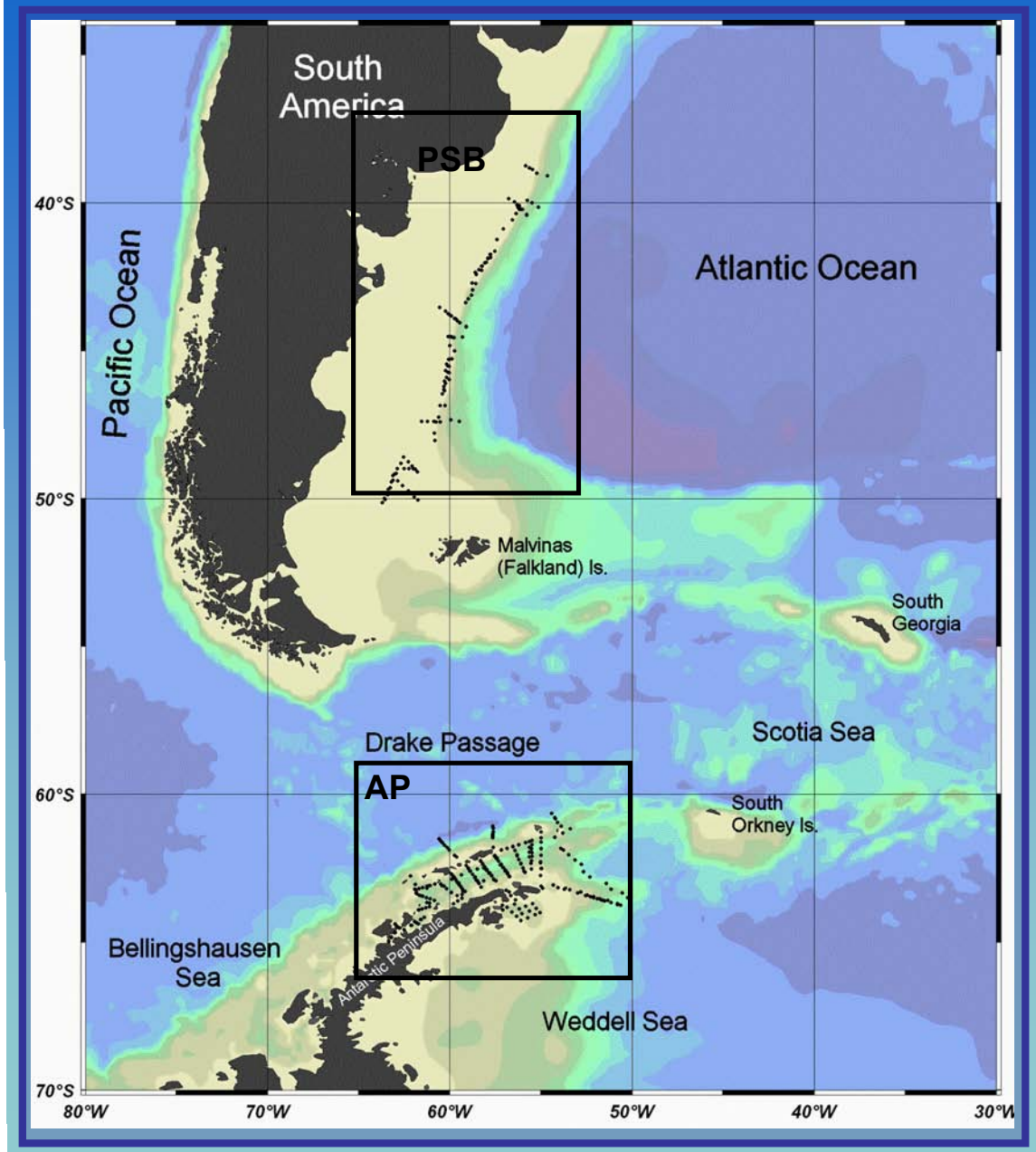
- ❑ Solvent delivery module
- ❑ Autosampler:
  - Injection volume: 0.1 to 100 µL
  - Sample cooler: 4 to 40ºC
  - Injection volume repeatability (RSD < 0.3 %)
  - Highest speed (15s injection cycle)
- ❑ Column Oven:
  - SunFire™ C8, 3.5 µm, 4.6x150mm column (Waters)
  - Temperature setting range: 4ºC – 85ºC
  - Temperature control precision: ± 0.1ºC
- ❑ Photodiode Array UV-VIS detector:
  - Measurement wavelength range: 190 to 800 nm
  - Cell temperature control: 9ºC to 50ºC
- ❑ Spectrofluorometric detector

STUDY REGIONS



HPLC vs FLUOROMETER
Chla measurements

SAMPLINGS FOR HPLC vs FLUOROMETER measurements



Patagonian Shelf-Break (PSB)

215 samples

Spring	Oct	2007
	Oct	2008
Summer	Jan	2008
	Jan	2009

Antarctic Peninsula (AP)

446 samples

Summer	Feb-Mar	2008
	Feb-Mar	2009

DATA AND METHODS

Chlorophyll data - Fluorometer

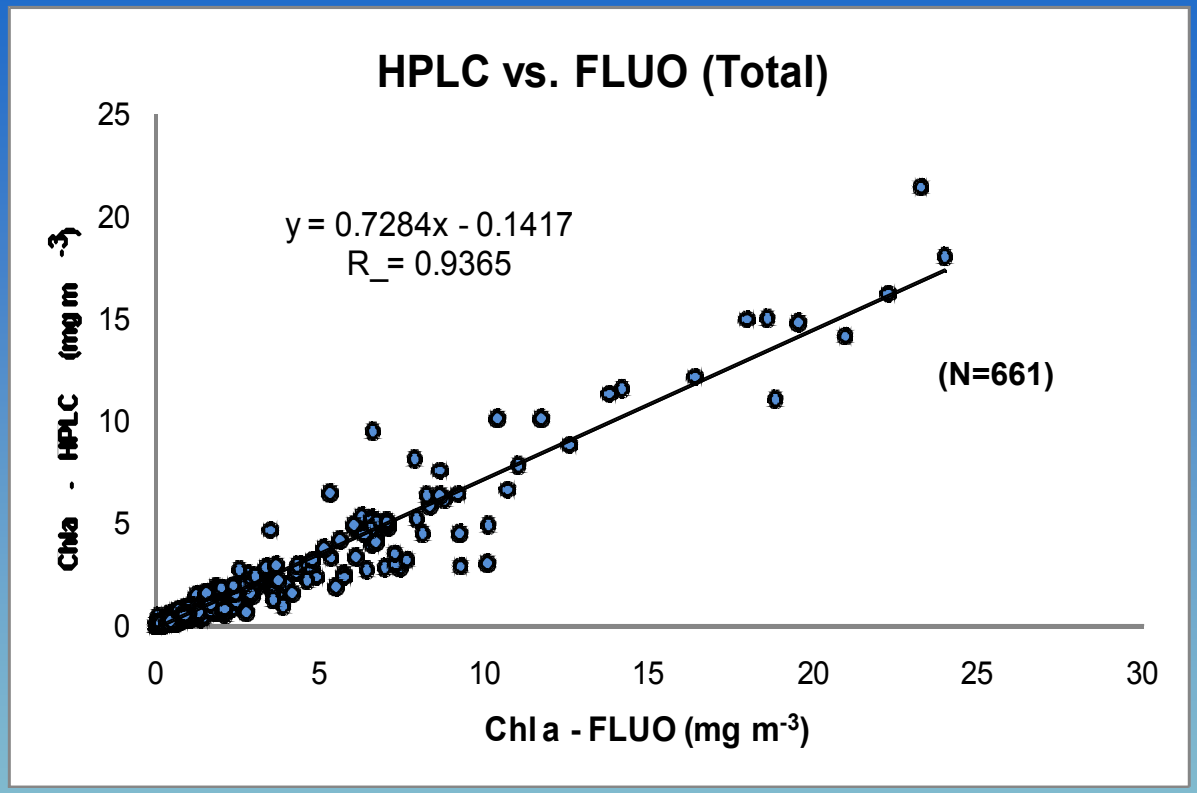
- Surface water samples for chlorophyll determinations were filtered onto 25 mm Whatman GF/F filters.
- The pigment was extracted in 90% acetone and fluorescence determined in a Turner Designs TD-700 fluorometer following the non-acidification method (Welschmeyer 1981).

Chlorophyll data - HPLC

- Method of FCUL/Univ. Lisbon (V. Brotas)

Remote Sensing / Match-up

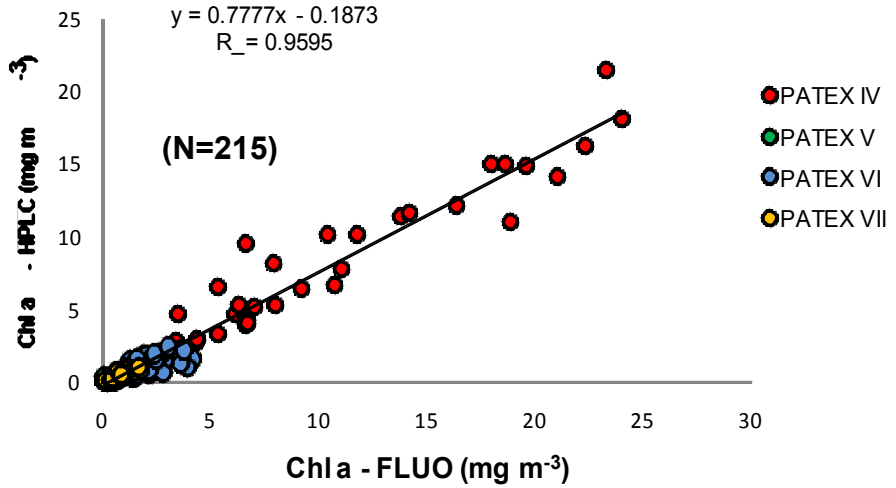
- Chl-a from MODIS/Aqua Standard OC3M algorithm
- Daily and eight-day 4 km resolution level-3 standard mapped image (SMI)
- Spatial: Chl-a value of the nearest pixel to the sample site
- Temporal: Within the same or 8 day period of the image

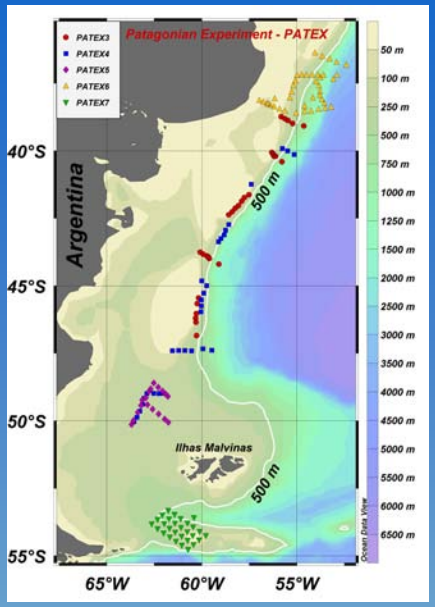


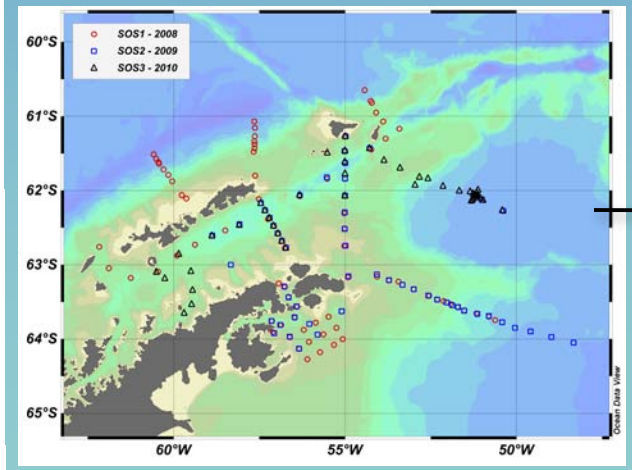
Chl c ?

Degradation Prods ?

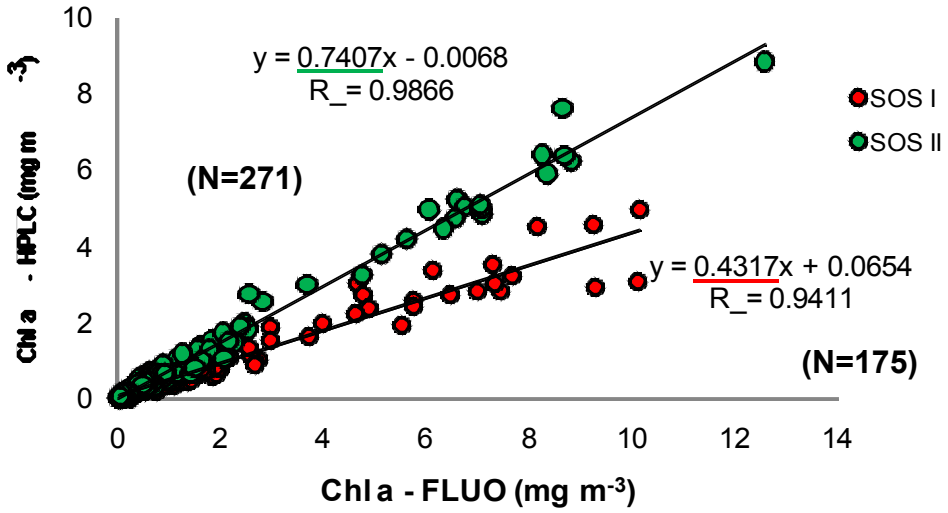
HPLC vs. FLUO (Patagónia)

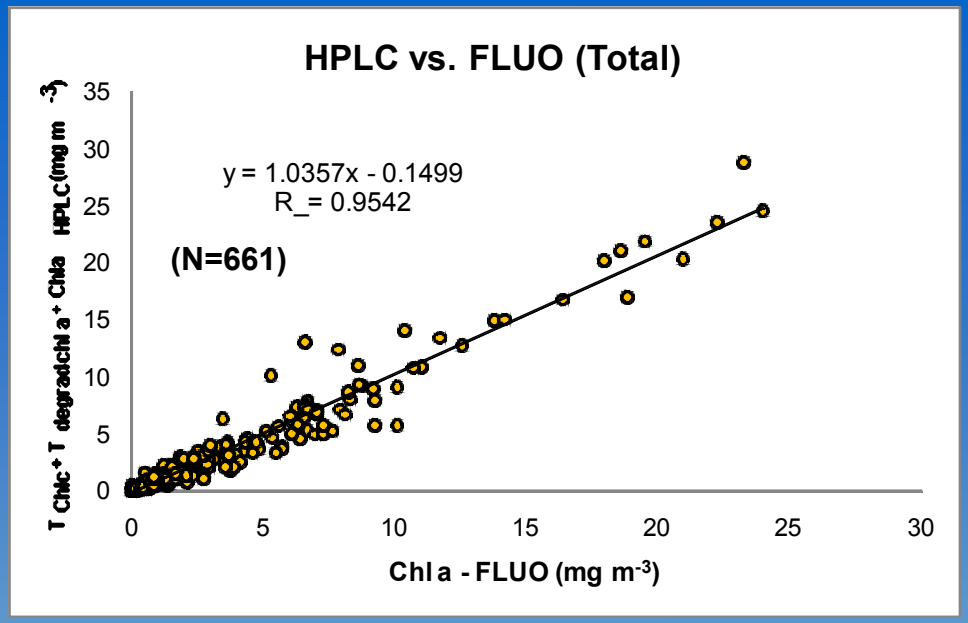


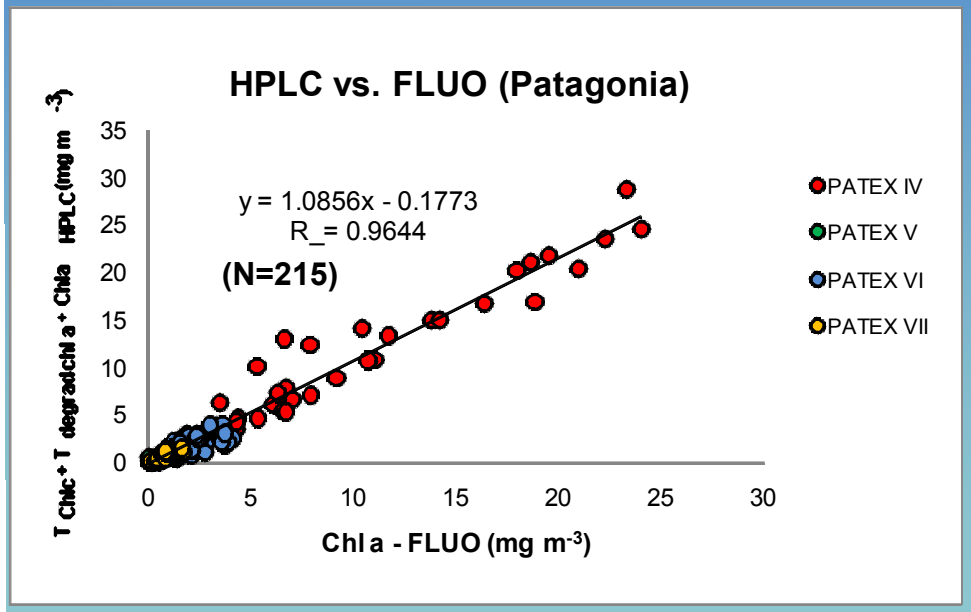


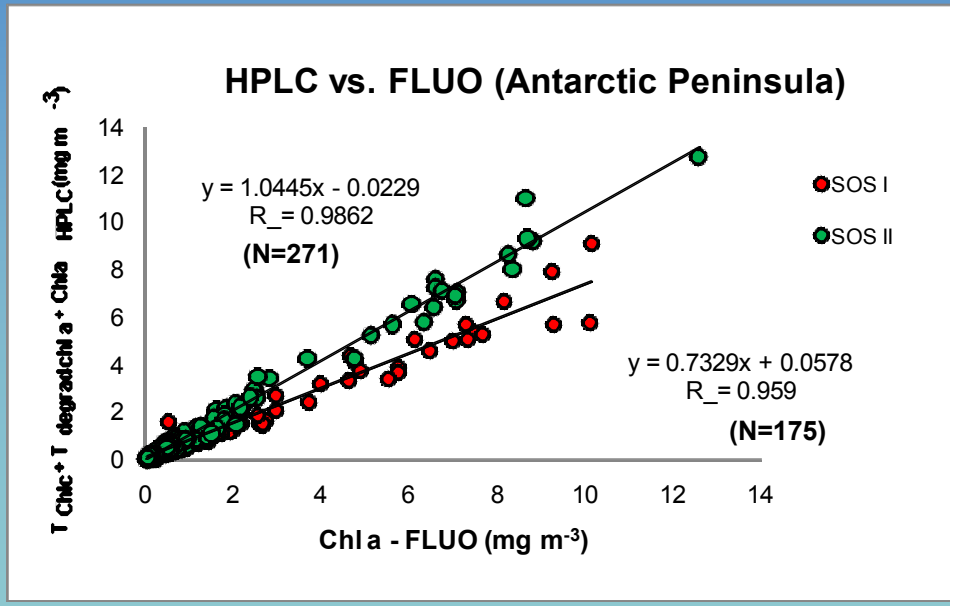


HPLC vs. FLUO (Antarctic Peninsula)



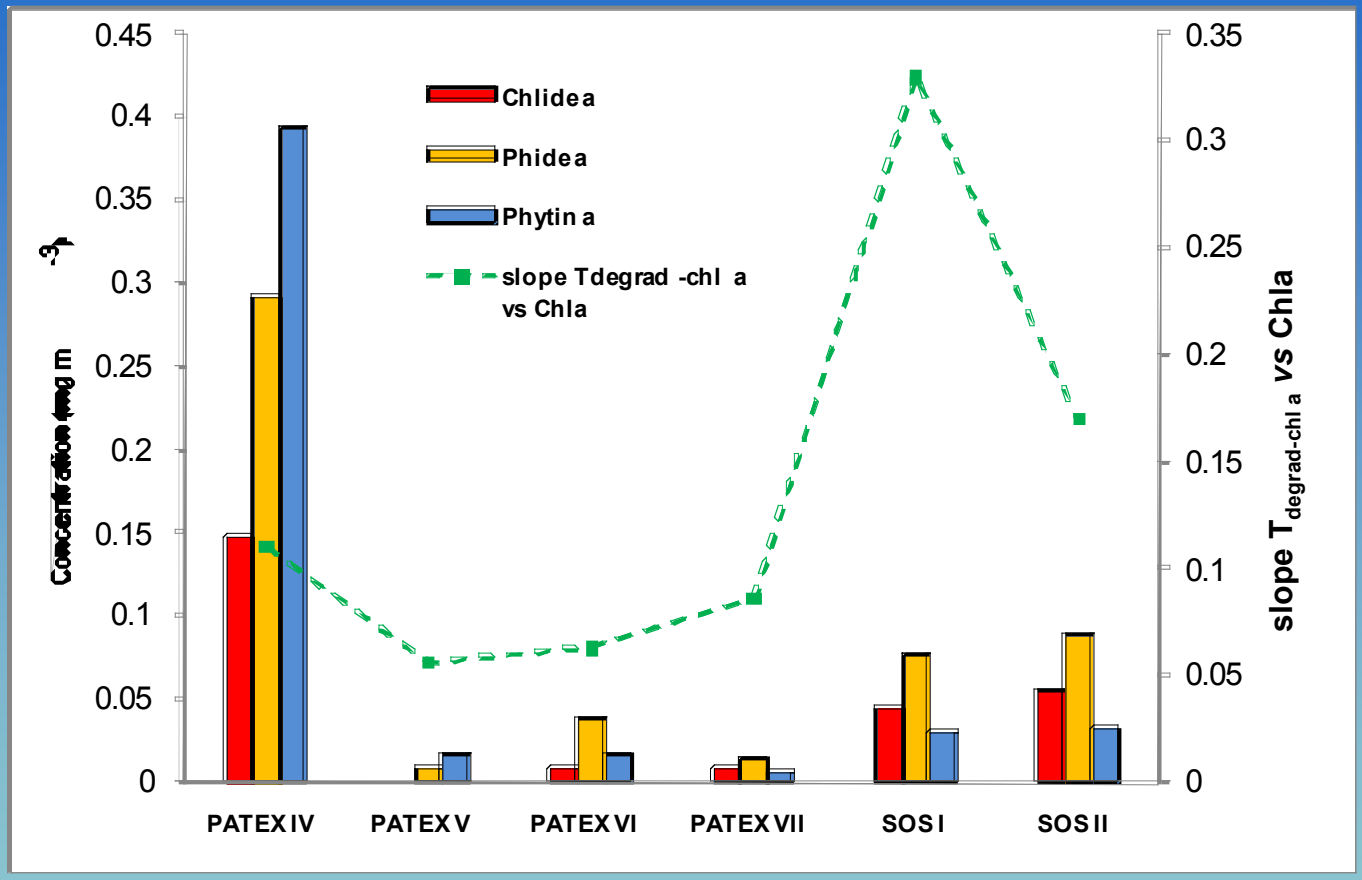




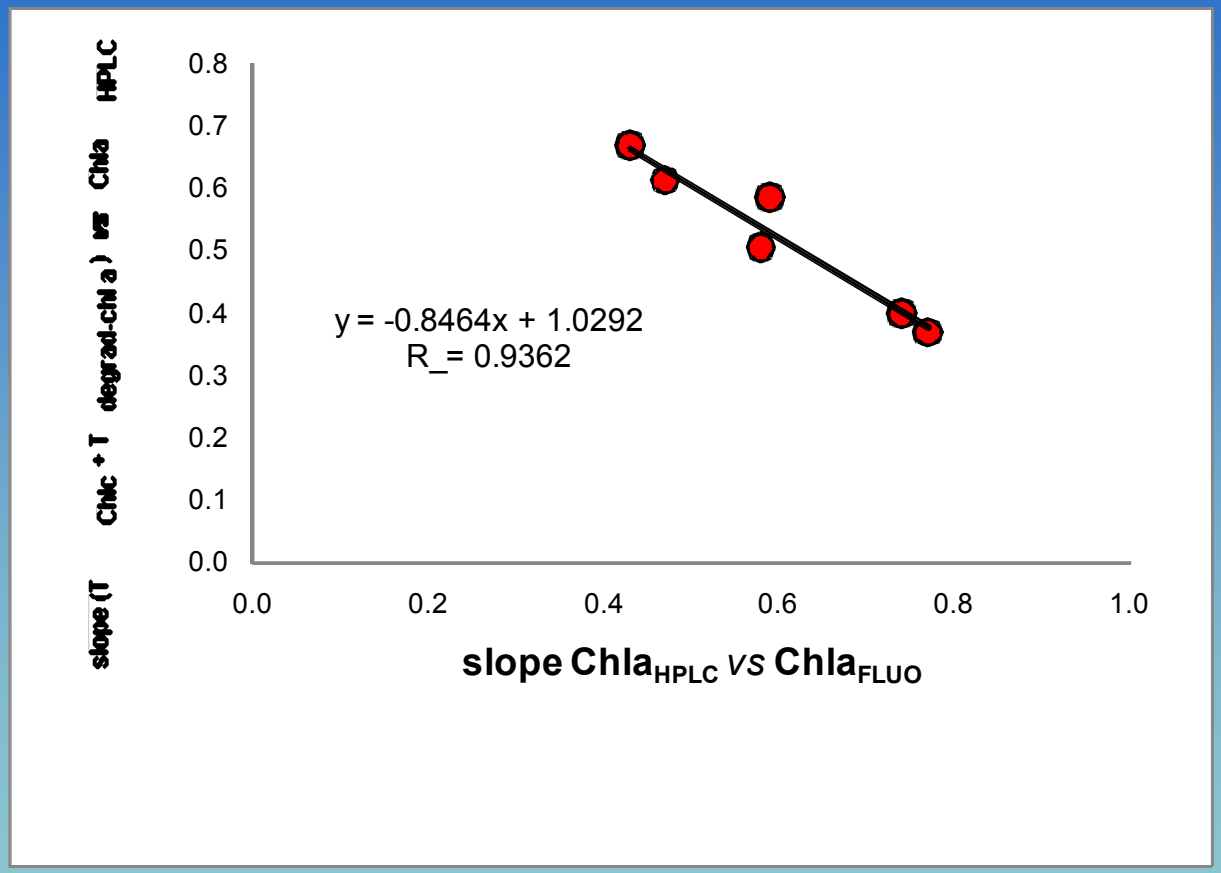


- PATEX IV (October 2007): y = 1,07x + 0.07; R^2=0.95; N=61
- PATEX V (January 2008): y = 0,96 - 0.097; R^2=0.80; N=35
- PATEX VI (October 2008): y = 0.80x + 0.17; R^2=0.68; N=68
- PATEX VII (January 2009): y = 0.92x - 0.07; R^2=0.75; N=68

Degradation products / chlorophyll *a*



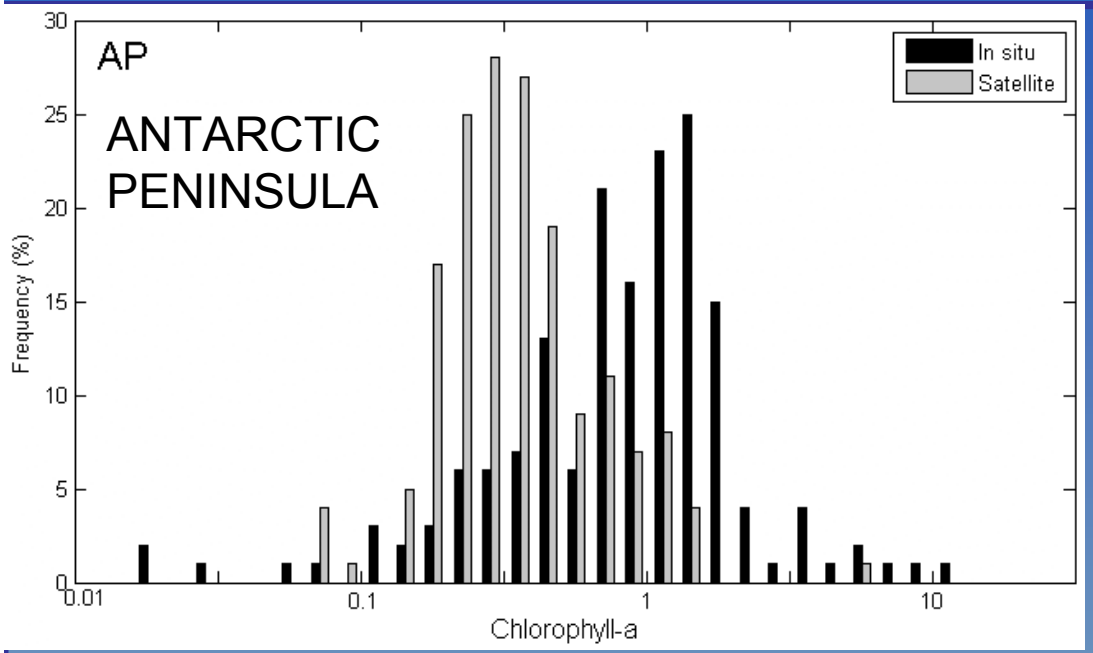
Total chlorophyll *c* ($T_{Chl\ c}$) or total degradation products of chlorophyll *a* ($T_{degrad\text{-}chl\ a}$)?

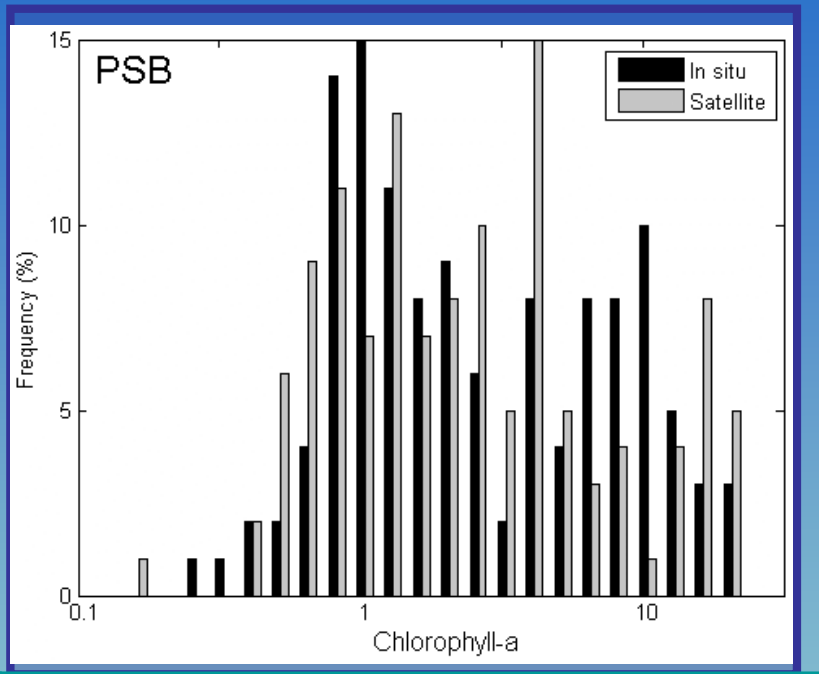


$T_{Chl\ c}$ = chlorophyll $c_1 + c_2 + c_3$
$T_{degrad\text{-}chl\ a}$ = chlorophyllide + pheophorbide+ pheophytin *a*

SATELLITE MATCH-UP RESULTS

Chlorophyll-a (mg/m3)

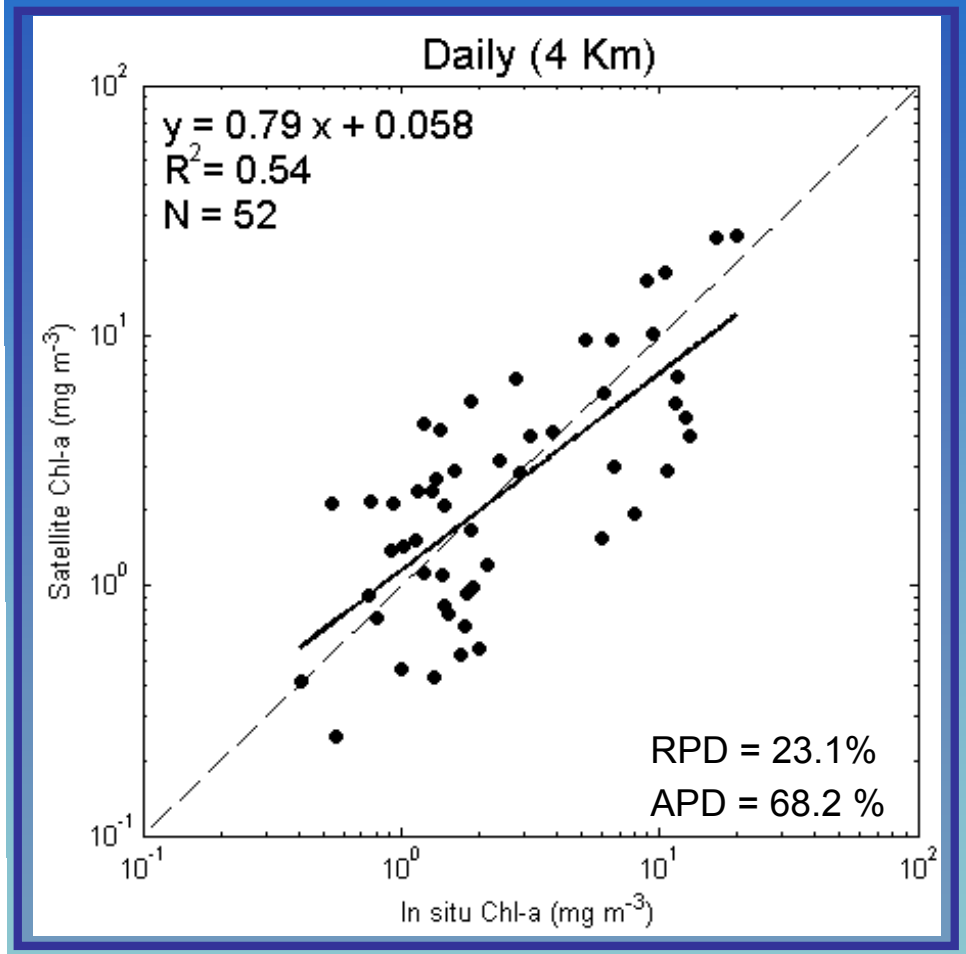


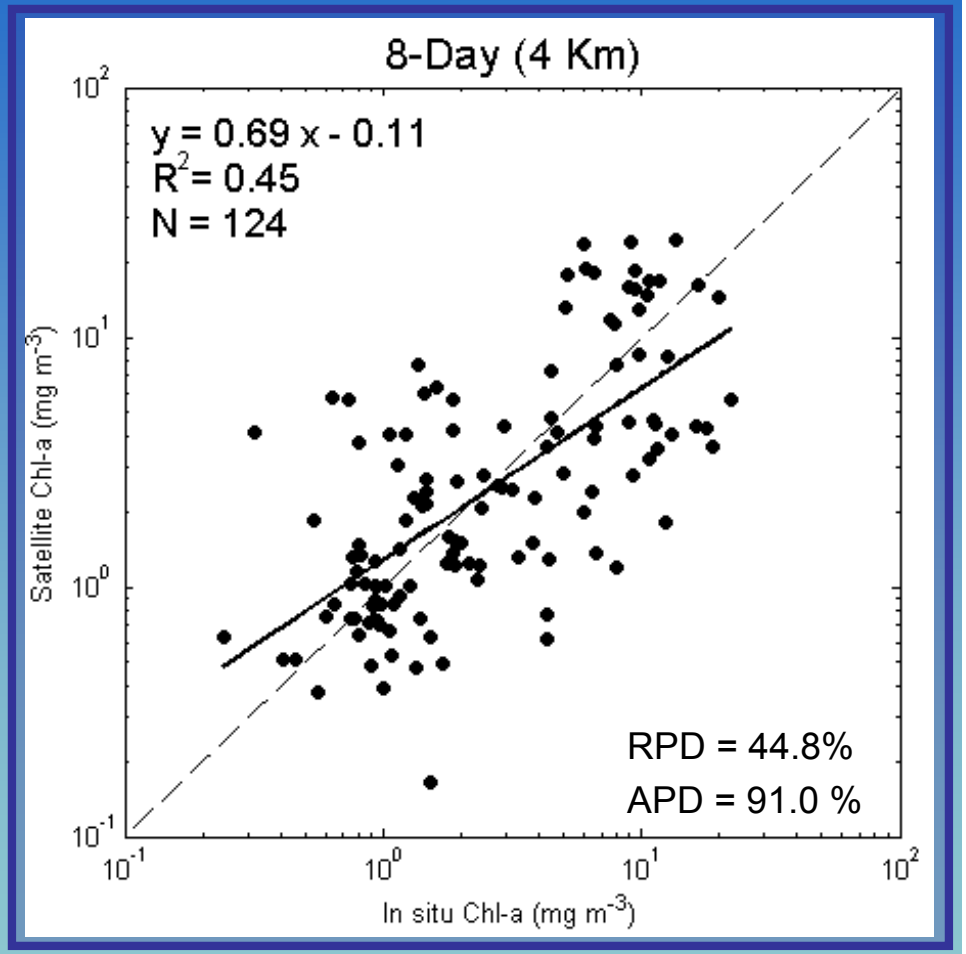


In Situ mean	1.24	In Situ mean	4.45
Satellite mean	0.45	Satellite mean	4.54
In Situ range	0.018 - 11.78	In Situ range	0.241 - 22.3
Satellite range	0.073 - 5.11	Satellite range	0.165 - 24.34

RESULTS

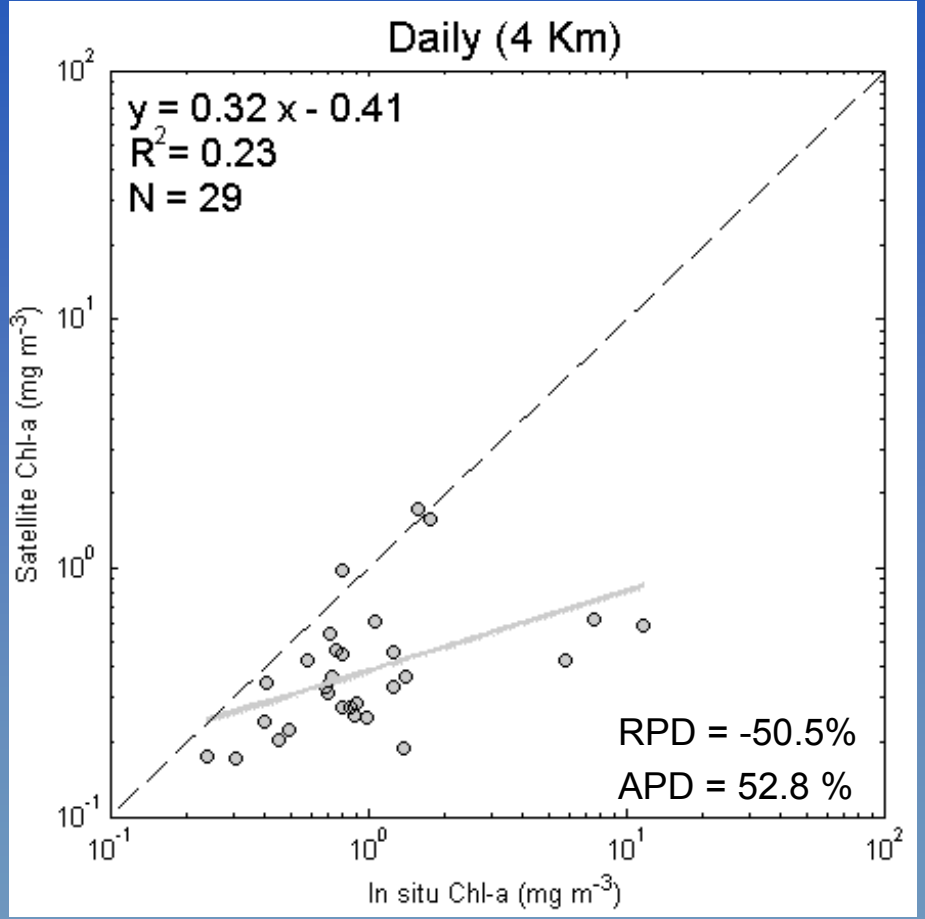
PATAGONIAN SHELF-BREAK

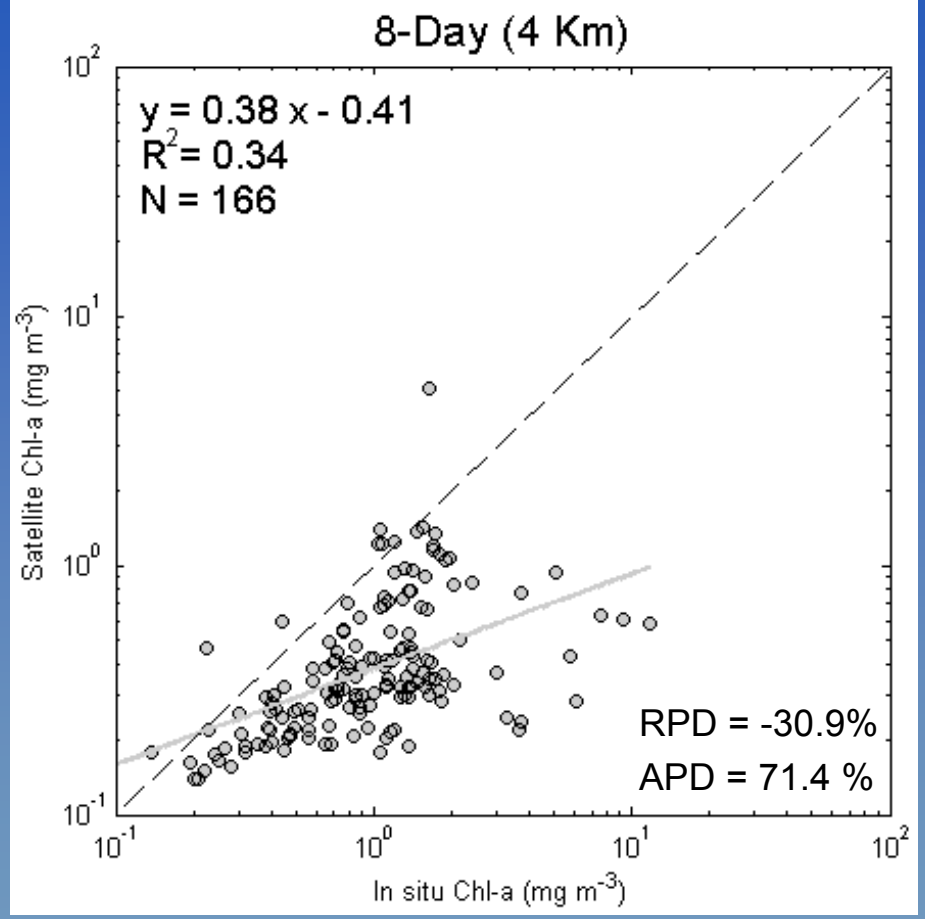




RESULTS

ANTARCTIC PENINSULA

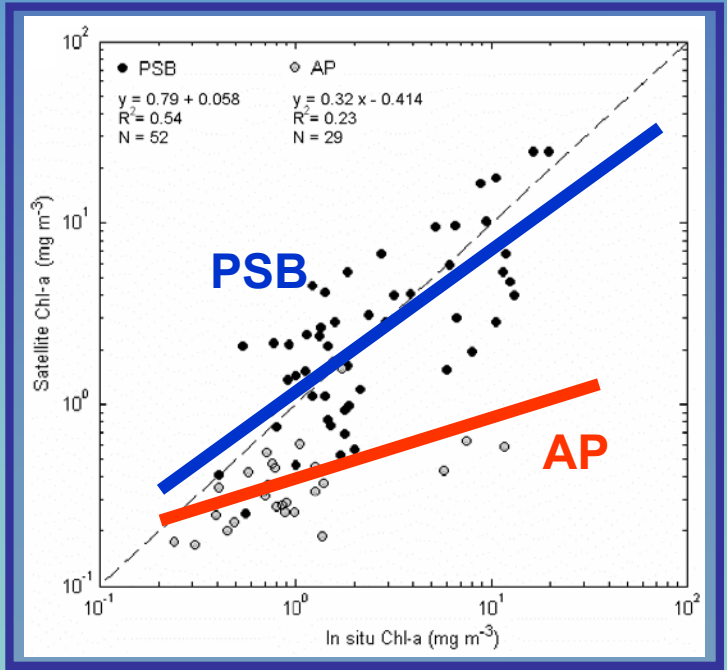




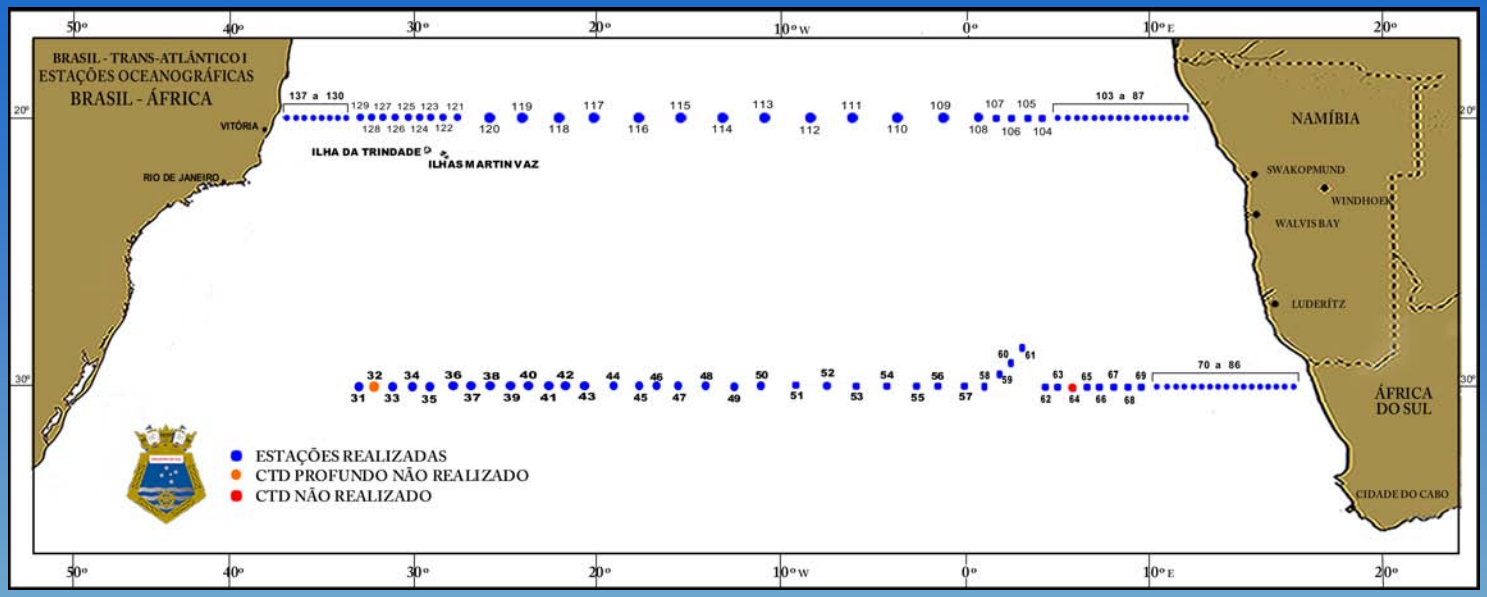
$$RPD = \frac{1}{n}\sum_{n=1}^{N}\left(\frac{(Chla_{sat} - Chla_{situ})}{Chla_{situ}}\right)\times 100 \quad ; APD \frac{1}{n}\sum_{n=1}^{N}\left|\frac{(Chla_{sat} - Chla_{situ})}{Chla_{situ}}\right|\times 100$$

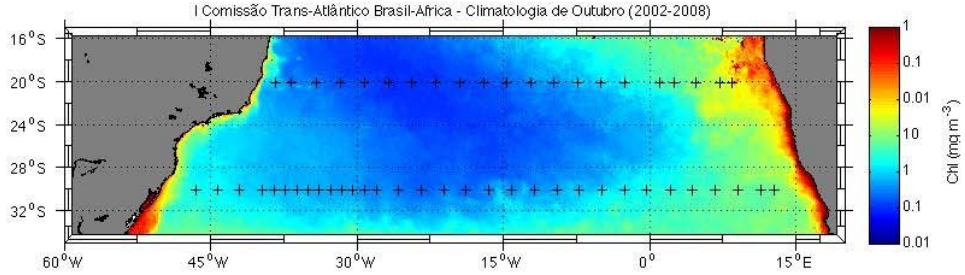
RESULTS

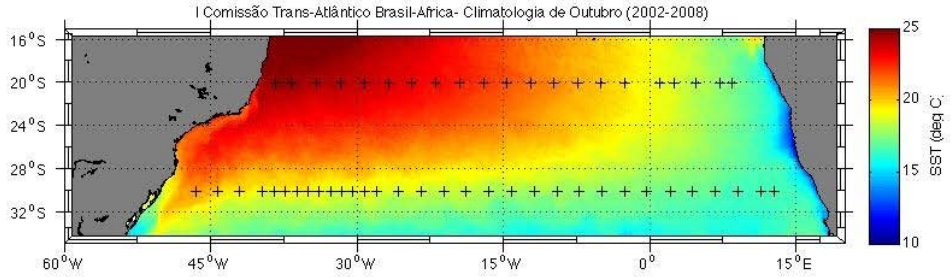
Patagonian Shelf-Break (PSB)	Antarctic Peninsula (AP)
MODIS **OC3M**	
Tend to overestimate low Chl-a and underestimate Chl > 2.0 mg m^{-3}	Tend to underestimate in situ Chl-a
Linear **relationship**	
R^2 = 0.5	R^2=0.23
High uncertainty	
APD ~ 68%	APD ~ 70%



FUTURE WORK


BRASIL - TRANS-ATLÂNTICO I
ESTAÇÕES OCEANOGRÁFICAS
BRASIL - ÁFRICA
VITÓRIA
RIO DE JANEIRO
ILHA DA TRINDADE
ILHAS MARTIN VAZ
NAMÍBIA
SWAKOPMUND
WINDHOEK
WALVIS BAY
LUDERITZ
ÁFRICA DO SUL
CIDADE DO CABO
137 a 130
103 a 87
70 a 86
ESTAÇÕES REALIZADAS
CTD PROFUNDO NÃO REALIZADO
CTD NÃO REALIZADO


I Comissão Trans-Atlântico Brasil-Africa - Climatologia de Outubro (2002-2008)
Chl (mg.m-3)


I Comissão Trans-Atlântico Brasil-Africa- Climatologia de Outubro (2002-2008)
SST (deg C)

Thank you

http://www.goal.ocfis.furg.br

Concluding remarks

- Global empirical algorithms perform better in the PSB than in the AP, though the uncertainty in the satellite estimates in both regions is high

- Knowledge of the bio-optical properties and its spatial and temporal variability is needed to further develop a regional algorithm

- We are currently measuring several bio-optical properties during the Internacional Polar Year (Project SOS-CLIMATE) to implement semi-analytical algorithms to derive
  - chlorophyll
  - dissolved organic matter
  - inorganic particulate material

from space

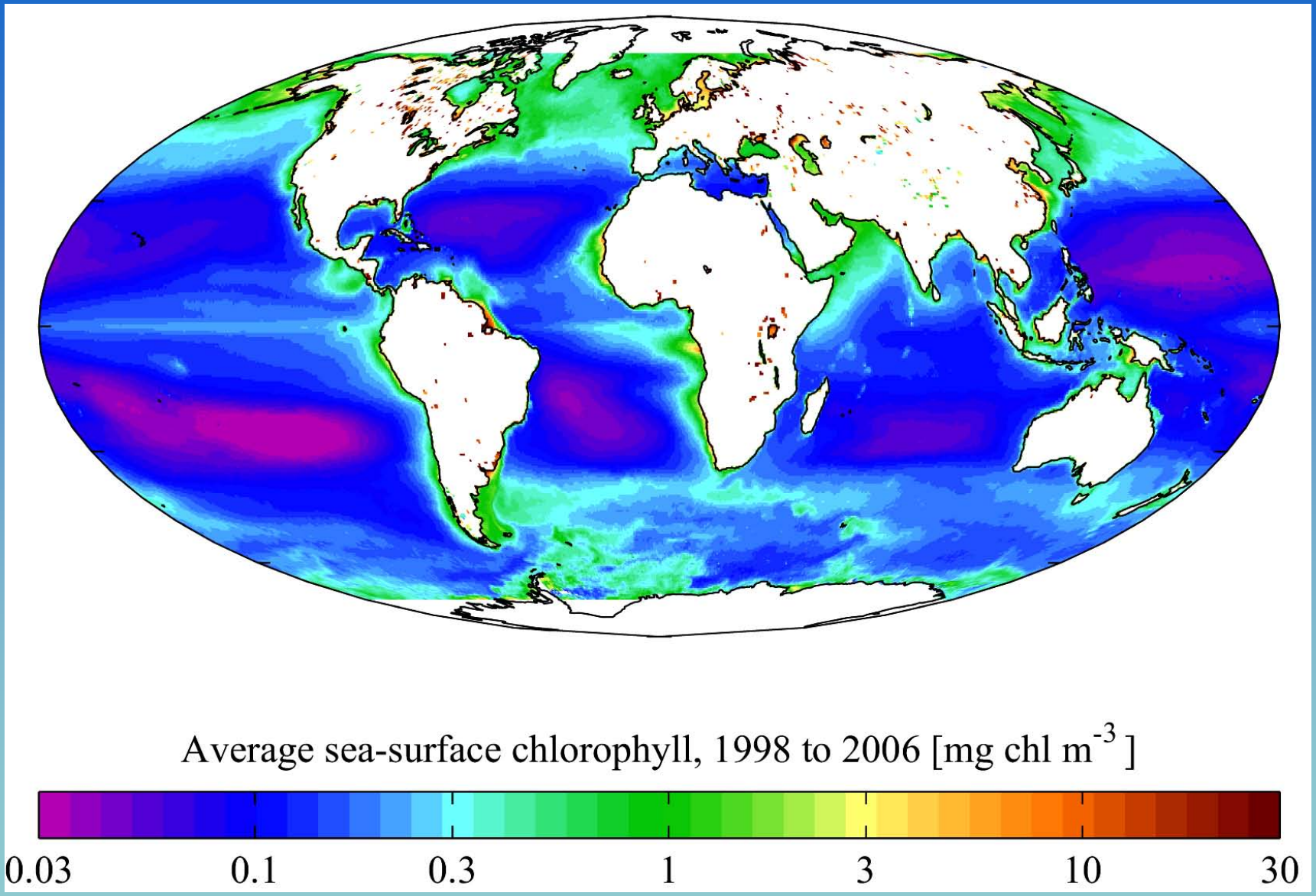
Possible causes of mismatch

- In situ samples are point measurements while satellite pixels cover a larger area (4×4 km^2).
- In situ and satellite measurements are not strictly concurrent in time
- **Specific optical properties of the region**

E.g. Antarctic Peninsula

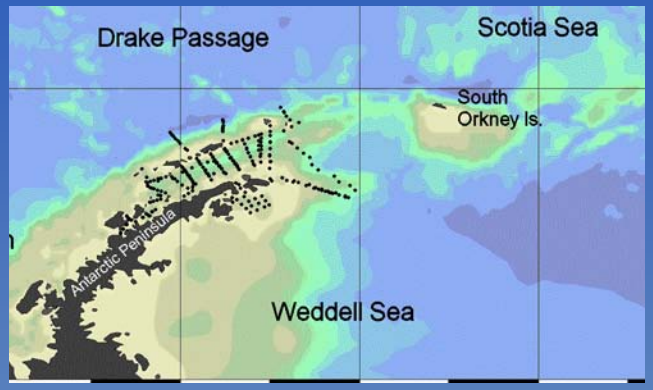
- Distinct optical characteristics of the water particles, including phytoplankton assemblages (Dierssen and Smith 2000)
- Fluorometric methods may result in biased results, particularly in the presence of certain accessory pigments (Marrari et al. RSE 2006)

Global Mean Image of Sea-Surface Chlorophyll



INTRODUCTION

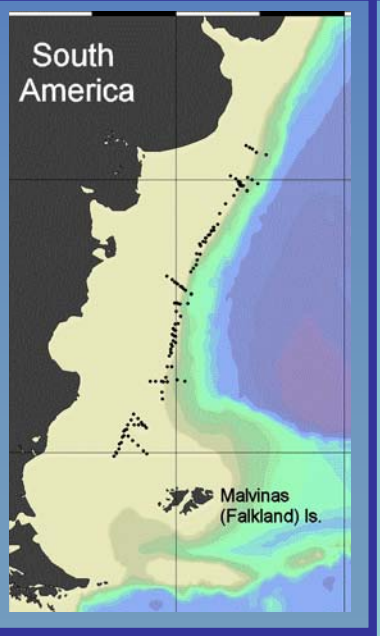
Southern Ocean



Antarctic Peninsula (AP)

- One of the most productive areas of the SO

- Supports large concentrations of phytoplankton, zooplankton, seabirds, seals, and whales



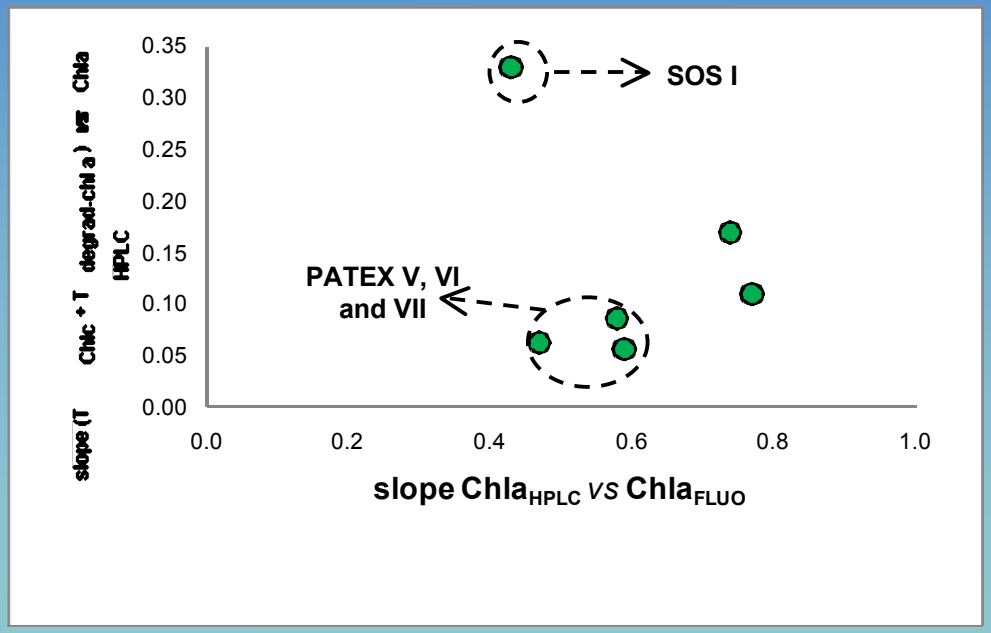
Patagonian Shelf-Break (PSB)

- Strongly influenced by nutrients supplied by Malvinas Current

- Frontal areas associated with intense ocean CO_2 uptake in spring-summer (↑ Chl-a - strong CO_2 sink)

Total chlorophyll *c* ($T_{Chl\ c}$) or total degradation products of chlorophyll *a* ($T_{degrad\text{-}chl\ a}$)?

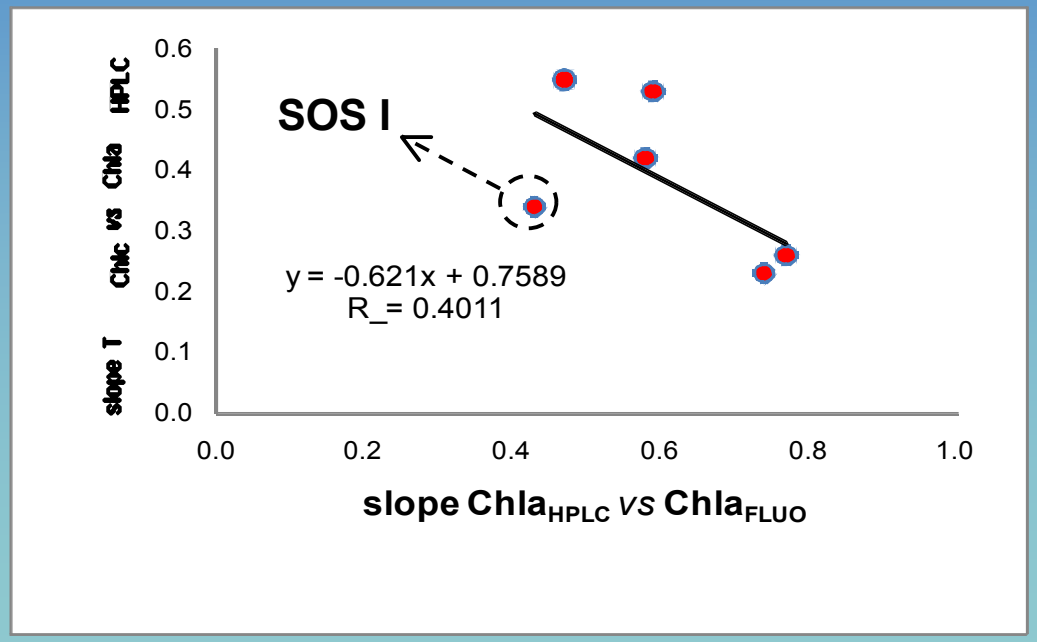
			Chla $_{HPLC}$ *vs* Chla $_{FLUO}$		$T_{degrad\text{-}chl\ a}$ *vs* Chla $_{HPLC}$	
Cruise	**Date**	**N**	**Y = ax + b**	**R^2**	**Y = ax + b**	**R^2**
PATEX IV	Oct-07	61	y = **0.77** x - 0.06	0.95	y = **0.11** x + 0.25	0.72
PATEX V	Jan-08	35	y = **0.59** x - 0.003	0.78	y = **0.056** x - 0.01	0.62
SOS I	Feb-08	271	y = **0.43** x + 0.065	0.94	y = **0.33** x - 0.05	0.84
PATEX VI	Oct-08	68	y = **0.47** x + 0.19	0.63	y = **0.063** x - 0.003	0.38
PATEX VII	Jan-09	51	y = **0.58** x - 0.01	0.74	y = **0.086** x - 0.001	0.16
SOS II	Feb-09	175	y = **0.74** x - 0.007	0.99	y = **0.17** x - 0.005	0.92



$T_{Chl\ c}$ = chlorophyll $c_1 + c_2 + c_3$
$T_{degrad\text{-}chl\ a}$ = chlorophyllide + pheophorbide+ pheophytin *a*

Total chlorophyll *c* ($T_{Chl\ c}$) or total degradation products of chlorophyll *a* ($T_{degrad\text{-}chl\ a}$)?

			Chla $_{HPLC}$ *vs* Chla $_{FLUO}$		$T_{chl\ c}$ *vs* Chla $_{HPLC}$	
Cruise	**Date**	**N**	**Y = ax + b**	**R^2**	**Y = ax + b**	**R^2**
PATEX IV	Oct-07	61	y = **0.77** x - 0.06	0.95	y = **0.26** x - 0.05	0.95
PATEX V	Jan-08	35	y = **0.59** x - 0.003	0.78	y = **0.53** x - 0.06	0.90
SOS I	Feb-08	271	y = **0.43** x + 0.065	0.94	y = **0.34** x + 0.014	0.88
PATEX VI	Oct-08	68	y = **0.47** x + 0.19	0.63	y = **0.55** x - 0.07	0.80
PATEX VII	Jan-09	51	y = **0.58** x - 0.01	0.74	y = **0.42** x - 0.03	0.65
SOS II	Feb-09	175	y = **0.74** x - 0.007	0.99	y = **0.23** x - 0.006	0.96



$T_{Chl\ c}$ = chlorophyll c_1 + c_2 + c_3
$T_{degrad\text{-}chl\ a}$ = chlorophyllide + pheophorbide+ pheophytin *a*

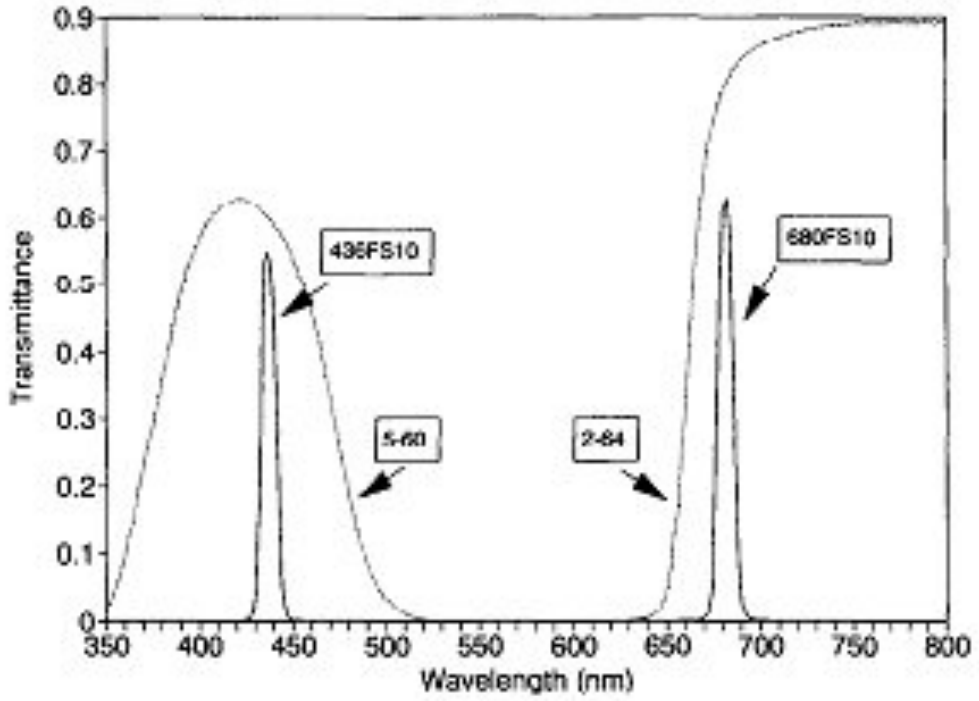


Fig. 3. Transmittance characteristics of excitation-emission filters used in conventional fluorometric acidification technique (Corning 5-60/Corning 2-64) and in the newly proposed method (436FS10/680FS10 interference filters, Andover Corp.).