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Research article

Evaluation of cyanobacteria bioenergy: a potential resource in Antarctica detected by Multi/Hyperspectral satellite Image.

Carla Micheli^{a, *}, Alessandro Belmonte^a, Vito Pignatelli^a, Luigi De Cecco^b, Viviana Piermattei^c, Marco Marcelli^c, Flavio Borfecchia^b

^a ENEA, Italian National Agency for New Technology, Energy and Sustainable Economic Development. Department of Energy Technologies, Laboratory of Biomass and Energetic Biotechnologies. Research Centre Casaccia C.P. 2400/00123 Roma, Italy

^b ENEA, Laboratory of Earth Observation, Research Centre Casaccia 2400/00123 Roma, Italy. ^c Laboratory of Experimental Oceanology and Marine Ecology, Department of Biological and Ecological Sciences (DEB), La Tuscia University, Molo Vespucci, Porto di Civitavecchia (RM), I-00053 Civitavecchia (Roma) Italy

E-mail carla.micheli@enea.it



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Abstract

This paper focuses on the natural conservation aspect of bioenergy conversion by polar cyanobacteria; they could represent a key requirement in the implementation of alternative renewable sources, since they have many advantages for fuel-level energy storage, particularly in polar habitat. We propose an innovative method for evaluating the potential bioenergy resource already existing in Antarctica, discovering cyanobacteria patches by remote sensing technique.

Cyanobacteria, which are considered potential precursors of liquid fuel and are alternative of bioenergy conversion, were collected in Antarctica during austral summer 2003. At the same time, *in situ* the microbial mat was identified by polar sensors (Hyperion, ALI and Lansat ETM+) having spectral/spatial resolution, which are able to detect



photosynthetic pigments that change in these populations not available elsewhere and suitable for their effective change assessment capabilities.

According to physiological, morphological and genetic features of collected samples, two groups of cyanobacteria (included in *Nostocales* and *Oscillatoriales* orders) were identified and, simultaneously mapped at three Antarctic stations Terranova Bay, Enigma Lake and Icaro in relation to pigment spectral responses to hyperspectral signatures. At the beginning of the 3rd era, the emergent need for an energetic biomass at an international level is highlighted using careful site selection and application of appropriate methods. Thus, in our results the technology links the perspective of knowledge of biomass as energy budget in the Antarctic ecosystem, together with the monitoring of integrated analyses of whole ecosystems from space. **Copyright © LJRETR, all rights reserved.**

Keywords: Cyanobacteria biomass and bioenergy conversion, Polar Hyperspectral Remote Sensing, Pigment spectral responses, Energy management, Photosynthetic microorganisms, Antarctica,

1. Introduction

Worldwide renewable energy demand is increasingly growing in direct proportion to its consumption. Accelerating the knowledge of energy availability, even in the polar lands, is now possible thanks to the development of a more efficient convergence of innovative and multidisciplinary programs [1]. Recently, the urgent governmental interest in using extremophile microorganisms for producing renewable energy has increased. They potentially produce biofuels more efficiency than plant biomass [2]. The development of new technologies to detect sustainable energy systems regarding renewable energy has became a global effort to resolve the enhancement of the energy deficit. Several alternatives have been proposed to develop a sustainable industrial society and reduce green house gases emission, such as the direct biological conversion of CO_2 with solar energy by photosynthetic microorganisms (microalgae and cyanobacteria) for the production of alternative fuel [3]. The polar photosynthetic cyanobacteria offer several advantages compared to traditional biofuel production from plant biomass such as their higher growth rate. Cyanobacteria are the most important photosynthetic organisms on the planet for cycling carbon and nitrogen fixation [4]. They have existed for 3.5 billion years and are rich in chemical diversity: some strains produce free fatty acids, the precursor to liquid fuels that might be one of alternative bioenergy products [5,6]. Although several strains are micro-algal fuel producers, others are notorious for causing nuisances such as dense and often toxic blooms across some of the most productive areas (in the seas, lakes and lagoons) particularly affected by both anthropogenic pollution/eutrophication and global change effects [7]. Many works [8,9] dealt with remote sensing of Cyanobacteria and algal blooms in sea or internal bodies waters which weakly affect the total reflectance signals, very few instead were focused on their detection over terrain where the noise signal arising from the substrate is very high, especially in polar areas characterized by visible high albedos from snow and ice covers.

However the extensive mapping and characterization of cyanobacteria of polar and alpine habitats are of particular interest because in general they represent predominant species contributing to a major component of the ecosystem



photosynthetic biomass. In addition, the strong abiotic stresses in these harsh environments during paste evolutionary ages have shaped their particular resilience capability with the production of specific pigments (phycobiliproteins) and in particular biochemical compounds potentially valuable for bioenergy products.

In this context high spectral resolution imagery remotely sensed by polar satellite sensors and fluorescence data were tested for Antarctic cyanobacteria discrimination through spectral optical reflectance features of their specie-specific pigments. This improved perspective aims in the Antarctic ecosystem have repetitive monitoring through integrated analyses of whole ecosystem from space, by polar sensors, such as Hyperion, ALI and Lansat ETM+. Their spectral/spatial resolution is able to detect cyanobacteria populations at large spatio-temporal scales, not anywhere available and suitable for their effective change assessment capabilities.

The solar radiation absorbed by photo-synthetically active pigments (mainly chlorophyll a, b) is partly used as energy input in the photosynthetic process. The remaining fraction of absorbed energy is dissipated in two different ways: i) fluorescent emission, ii) heat dissipation. These processes occur in competition with each other, so that any variation efficiency of one of the three will involve a variation of yield in the other two. Hence the idea of measuring the yield of chlorophyll fluorescence to draw information to respect variations of efficiency relative to the other two processes, in particular in relation to photosynthesis. The fluorescence originates from the radioactive de-excitation of chlorophyll molecules of photo-systems: from an undulatory point of view, the radiation emitted in fluorescence has a wavelength of 10 nm on average greater than the incident radiation absorbed. The fluorescence patterns of the cyanobacteria are distinct from those of the plants, due to their key structural and functional properties. These include significant fluorescence emission from the light-harvesting phyco-biliproteins. These accessory pigments favor the absorption of electromagnetic radiation in wavelengths of chlorophyll-*a* taking advantage of a wider range of the available photosynthetic radiation [10,11].

In this paper, we report a study for assessing the distribution of benthic communities and phytal pigment content of cyanobacteria, on polar patches in Antarctica. By remote sensing techniques the hyperspectral signatures were detected by a priori knowledge of the fluorescence peaks of collected samples and the characterization of the sampling sites. Simultaneously, historical changes detected by harvesting of Antarctic populations at various times over the last two centuries were identified in different polar areas by phylogenetic patterns of Antarctic cyanobacteria [12].

This multi-disciplinary integrated approach involving innovative technologies aims at detecting patches of photosynthetic organisms for suitable energy production in the polar areas where the changes of the Earth System are amplified and can be seen anywhere, on the planet and in the paste, at suitable spatial/temporal scales. This applied technique could be used in the future to determine map of unknown diesel samples cyanobacteria to gain further information on the fatty acid profile in the view of a future biodiesel production.

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2. Materials and Method

2.1 Sampling sites and laboratory work

During austral summer 2003, cyanobacteria were collected from the surface layer of three stations selected in Antarctica: Terranova Bay, Enigma Lake and Icaro (Fig. 1, Table 1). Subsequently, all the species were grown in laboratory conditions and their morphological and biomass analyses (g/mL) were performed before identifying them by molecular and microscopic analyses [5,12,13].

Cyanobacteria are the major element of the biota across the full range of these lakes. In particular, in the zone of Enigma Lake, the biological community is dominated by the species included in the order of Oscillatoriales, whilst in the other two stations Nostocales are also present. All the communities showed the highest Chlorophyll a concentrations (680 nm) and blue-green color due to the pigments phycocyanin (612 nm) and phycobilin (500-660 nm) that is more typical of the Cyanobacteria Nostocales population, as shown in Table 1.

Table 1 Sampling stations, geographical coordinates, Cyanobacteria, and pigment content (Chlorophyll *a*, *Phycobiline*, *phycocyanine and phycoeritrine*).

Sampling sites*	Geographical coordinates	Cyanobacteria	Chl a (nm)	Phycobiline (500-660 nm)
Terranova Bay	74°41'41.94''S; 164°06'7.02''E	Oscillatoriales: <i>Leptolymbya</i>	678	phycocyanin (612)
Enigma Lake*	74°43' 8.23"S; 163°55' 7.65"E	Oscillatoriales: <i>Leptolymbya</i> (ex <i>Plectonema</i>)	678	phycocyanin (612)
Icaro*	74°42' 8.23"S; 164° 6' 7.65"E	Nostocales: <i>Nostoc;</i> Oscillatoriales: <i>Leptolymbya</i>	678 678	phycoeritrin (580) phycocyanin (612)





Fig. 1 - Antarctic regions investigated in the three sampling sites (red dots) by Hyperion (Strip crossing the region of interest from upper-right to lower-left side), ALI (strips from upper left and upper right) and LANSAT ETM+ (background) and particular of region of interest (lower picture)

2.2 Sensors and EO data preprocessing.

The Hyperion is one of the first passive hyperspectral sensor satellite, operating since the year 2000 from the NASA EO-1 polar platform in line with the other ALI (Advanced Land Imager) [14,13]. It is able to acquire high resolution (30 m) images of the Earth's surface within 220 spectral bands with approximately 10 nm bandwidths and spanning from 400 to 2500 nm wavelengths. The ALI instead is a multispectral instrument having the same ground resolution (30 m.) and radiometric accuracy in its ten bands from VIS to NIR and SWIR, covering most of those in the Landsat system. The EO-1 is flying information and with the Landsat 7 satellite, they share the same orbit on which there is about a one minute delay and this allows the Hyperion and ALI to take a subset of the same images, also with an independent off-nadir acquiring programmable capability. The Hyperion's high spectral resolution spanned over the visible and NIR/SWIR ranges, which were poorly covered by other operating multispectral sensors, coupled with its SNR, radiometric (12 bit) and spatial accuracy. This makes it very attractive and effective for studying the pigmentdriven responses of vegetation and ecosystems where much more of the discriminative information is found in the visible, red edge, and near infrared parts of the spectrum. The usefulness of this special feature is further enhanced in polar areas monitoring due to favorable repeat cycle arising from polar orbits crossing over the Earth poles. Here during the long polar day's (about six months) it is then possible to acquire frequent hyperspectral images in order to better support various studies devoted to EO (Earth Observation) systems calibration, assessing the ecosystems dynamics, biological trends and biophysical parameter of interest as those related to algal blooms and polar cyanobacteria distributions. A strip taken by Hyperion sensor in the January 2003 and including the three in situ



surveys sites was selected taking into account the ground sampling date. A preventive selection of a subset (155/220) of useful spectral bands data acquired by Hyperion system was necessary to exclude the not working spectral channels. The atmospheric preprocessing of the raw data was performed with the FLAASH code using a "midlatitude-winter" atmospheric and "maritime" aerosol models with spectral polishing and wavelength.

2.3 Calibration of the samples

Sampling areas included in satellite imagery, the description of sites and bio-chemical features of cyanobacteria found are reported in Table 1. The samples were collected in the Antarctic summer of 2003 and suitably analyzed for cyanobacteria population characterization. In the three stations considered (*) because they are included in the Hyperion 7.5 Km strip of the 2003, the cyanobacteria distributions are quite different. In fact as you can see in Table 1, the Icaro station is characterized by the *Nostoc* presence which involves the high content of specific pigment (phycobilin). Pigment absorbance of the typical Antarctic cyanobacteria collected in the three regions to be detected by the satellite hyper-spectral sensor shows the following picks: Chlorophyll a at 678 nm and pigments included in the phycobilin in the range of 500-660 nm; they also contain the phycoeritrin at 580 nm, the phycocyanin at 612 nm and the allophycocyanin at 650 nm.

3. Results and Discussion

In Antarctica cyanobacteria are the major element of the biota across the full range of these lakes where the biological community is dominated by species included in the order of Oscillatoriales and Nostocales. All the communities showed the highest Chlorophyll a concentrations (680 nm) and blue-green color due to phycocyanin (612 nm) pigment as well as phycobilin (500-660 nm) content which typically occurs in the Cyanobacteria of Nostocales population. The spectral signatures of the three stations derived from the hypespectral preprocessed reflectances (VIS, NIR and SWIR) data are reported in Fig. 2 where, in its upper part there is their zoomed version of the visible-NIR spectral range. Although here their shapes are similar, probably due to different substrate contribution, they differ for mean values and show a relative y-shift.

To capture the useful reflectance signal of the target pigments minimizing at the same time the different contribution of high-albedo substrates in the hyperspectral data, specific bands ratio responses were implemented. According to previous work [8,9], where some reflectance bands ratio have been suitably exploited through laboratory analyses to assess the phycocyanin content of similar cyanobacteria species, the R_{654}/R_{617} , R_{709}/R_{620} , R_{700}/R_{600} indices were approximated using Hyperion bands wavelengths. All these indices were located in the upper spectral range of interest (A box in graph of Fig. 3). Being designed to highlight the specific absorbance/reflectance features of phycocyanin pigment typical of Oscillatoriales Cyanobacteria population they should be able to detect its distribution around all the stations. To also account for the specific phycobilin response of Nostocales, two



additional spectral ratio indices, located in the lower part of the spectrum (B box in graph of Fig. 3), were introduced R_{550}/R_{510} and R_{580}/R_{520} to obtain a five indices ratio multilayer distribution.



Fig. 2–Remotely sensed hyperspectral signatures of the three sampling stations in the VIS, NIR and SWIR wavelengths and the upper spectral range of interest.



Fig. 3 Ratio indices spectral signatures of the three sampling stations and the zoomed version of the visible-NIR spectral range.



The five ratio-indices local responses of the three sites were analyzed in order to check their spectral separability, in particular that of the Icaro station which could be associated to the presence of Nostocales cyanobacteria population absent in the others two stations. As you can see in Fig. 4 the ratio indices responses of the Icaro station in general are lower than others, except for $R_{654/617}$ and $R_{709/620}$ indices which show closed values for all the stations. The two introduced indices seem to be effective to discriminate between phycocyanina prevalent Oscillatoriales of the Zucchelli/Enigma stations and Nostocales prevalent Cyanobacteria of the Icaro station. Being their intensity higher than that of the others, a better signal to noise ratio could be exploited. In addiction The $R_{700/520}$ index also shows adequate separate responses of the three stations. The spectral signatures' separability was tested through a transformed divergence distance (TDD) with the following result for the three stations, reported in Table 2.

Zucchelli (Terranova Bay)	1456	1529
Icaro		1975
Enigma lake		

Table 2- Spectral Transformed Divergence distance between the ratio reflectance signatures of the three stations

The lowest TDD between the Zucchelli and Icaro stations (Table 2) agrees with their minimum geographic distance while the highest geographic separation could be explained by the maximum TDD values involving the Enigma station. In this context although it so happens that the geographic distance from the coast is the main factor determining the total spectral diversity of the sampling station sites, the effective influence of the different Cyanobacteria populations should be based on their actual ground concentration to be measured through devoted in situ campaigns.

Even though the attained transformed divergence values are not too high, in order to obtain a local distribution of the three kinds of Cyanobacteria covers a supervised classification using ML (Maximum Likelihood) parametric algorithm was attempted. The thematic map of the region of interest obtained from ratio indices image classification is shown (Fig. 4) in overlay to false color from hyperspectral band selection. As you can see from the classification map the Nostocales prevalent cyanobacteria population rightly extends over the coastal region while the other two are differently mixed in the remaining areas (Fig. 4).

It should be pointed out that in this work only qualitative in situ calibrations (identification of different Cyanobacteria populations) on the three stations have been exploited and in order to confirm and improve the implemented methodology more quantitative and additional ground measurement would be necessary. In addition to their genus/specie the different cyanobacteria concentration should be measured and point reflectance spectra from

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different substrates should be acquired by means of hand held radiometers to suitably support more effective EO data processing and covers characterization. From these results the exploitation of hyperspectral data remotely sensed by polar satellite platforms for detecting and mapping the distribution of Antarctic cyanobacteria is now feasible.



Fig. 4 - Classification map of the Region of interest showing the distribution of three kinds of cyanobacteria covers found in the 3 station at 99% of confidence level.

4. Conclusion

Cyanobacteria, which are considered potential precursor of liquid fuels as well as a number of useful chemical products, an alternative of bio-energy conversion and green chemistry, were collected in Antarctica during austral summer 2003 and were detected by polar sensors (Hyperion, ALI and Lansat ETM+). They represent one way of natural biological resources to utilize for renewable energy which potentially could cover for a part of demand of its deficit. Accelerate the knowledge of energy availability in the polar continents is possible thanks to the development of a more efficient convergence of innovative techniques [15, 16]. The new hyperspectral sensors offer availability to study the polar ecosystems faster and cheaper in the interests of multi-disciplinary point of view. Such study aims to disseminate and discuss the results in this harsh polar area for the first time, encouraging new techniques and also to promote the scientific community a broad debate on questions concerning the research of non conventional sources of renewable energies.

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References

- [1] C. Micheli, Considerations on energy from biomasses, Energy Conver and Management 23 (1983) 65-66
- [2] E.B. D'Alessandro, N.R. Antoniosi Filho, Concepts and studies on lipid and pigments of microalgae: A review, Renewable and Sustainable Energy Reviews 58, (2016) 832–84
- [3] I.M. Machado, S. Atsumi, Cyanobacterial biofuel production, J Biotechnol. 162 (2012) 50-6 doi:10.1016/j.jbiotec.2012.03.005.
- [4] K. Sciuto, I. Moro, Cyanobacteria: the bright and dark sides of a charming Group, Biodivers Conserv 24 (2015) 711–738
- [5] C. Micheli, F. Spinosa, A. Paperi, R. Buccioni, B. Pushraraj, Biodiversity and Fatty acid production in cyanobacteria, Rapp. Comm. int. Mer Médit. 38 (2007) 380.
- [6] R.C. Saxena, D.K. Adhikari, H.B. Goyal, Biomass-based energy fuel through biochemical routes: A review. Renewable and Sustainable Energy Reviews 13 (2009) 167–178
- [7] T. Cibic, L. Bongiorni, F. Borfecchia, A. Di Leo, A. Franzo, S. Giandomenico, A. Karuza, C. Micheli, M. Rogelja, L. Spada, P. Del Negro, Ecosystem functioning approach applied to a large contaminated coastal site: the case study of the Mar Piccolo of Taranto (Ionian Sea). Environ Sci Pollut Res 23, 13 (2016):12739–12754 DOI 10.1007/s11356-015-4997-2
- [8] S. Mishra, D.R Mishra, W.M. Schluchter, A Novel Algorithm for Predicting Phycocyanin concentrations in Cyanobacteria: A Proximal Hyperspectral Remote Sensing Approach Remote Sens, 1 (2009) 758-775
- [9] J. Gower, R. Doerffer and G.A. Borstad, Interpretation of the 685 nm peak in water- leaving radiance spectra in terms of fluorescence, absorption and scattering, and its observation by MERIS. Int. J. Remote Sens. 9 (1999) 1771-1786.
- [10] T.T. Wynne, R.P. Stumpf, M.C. Tomlinson, P.A. Tester, J. Dyble, G.I. Fahnenstiel, Relating Spectral shape to cyanobacterial blooms in the Laurentian Great lakes, Int J. Remote Sens 29,12 (2008) 3665-3672
- [11] T.T. Wynne, R.P. Stumpf, M.C. Tomlinson, G.I. G.L. Fahnenstiel, J Dyble, D.J. Schwab, S.J. Joshi, Evolution of a Cyanobacterial Bloom Forecast System in Western Lake Erie: Development and Initial Evaluation. Journal of Great Lakes Research 39 (2013) 90-99. DOI: 10.1016/j.jglr.2012.10.003



- [12] C. Micheli, R. Cianchi, R. Paperi, A. Belmonte, B. Pushraraj, Antarctic cyanobacteria biodiversity based on ITS and TrnL sequencing and its ecological implication. Open Journal of Ecology 4 (2014) 456-467 <u>http://dx.doi.org/10.4236/oje.2014.48039</u>
- [13] B. Pushparaj, A. Buccioni, R.Paperi, R. Piccardi, A. Ena, P. Carrozzi, C. Sili, Fatty Acid Composition of Antarctic Cyanobacteria. Phycologia 47 (2008) 430-434.
- [14] F.L. Lobo, C.C. Barbosa, E.M.M.L. Novo, J.S. Yunes, Mapping potential cyanobacterial bloom using Hyperion/EO data in Patos Lagoon Estuary, Acta Limnol. Bras.21, 3 (2009) 299-308.
- [15] F. Borfecchia, M. Frezzotti, Satellite Image Mosaic of the Terra Nova Bay Area, Victoria Land, Antarctica. Memorie Società Geologica Italiana, 46 (1989) 521-523.
- [16] F. Borfecchia, A. Cimbelli, L. De Cecco, A.B. Della Rocca, S. Martini, R. Barbini, F. Colao, R. Fantoni, A. Palucci, S. Ribezzo, Integrated remote sensing mission in the Venice Lagoon. Proc. SPIE 2959, Remote Sensing of Vegetation and Sea, 162 (1997); doi:10.1117/12.264266; <u>http://dx.doi.org/10.1117/12.264266</u>