1	Krill distribution in relation to environmental parameters in mesoscale structures in
2	the Ross Sea
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30 Abstract

Krill in Antarctica is one of the fundamental pelagic resources of the food web. It constitutes an important link between primary and secondary production and top predators. For this reason monitoring krill abundance is a very relevant activity because the availability of this resource permits the correct functionality of the local ecosystem. The acoustic methodology is one of the most suitable ways to monitor krill abundance at sea. Several acoustic surveys were made in the western Ross Sea, revealing the presence of Antarctic krill (Euphausia superba) in the northern-central part of this area and confirming the high relevance of ice krill (Euphausia crystallorophias) in the central coastal part of the Ross Sea. More recently, an acoustic survey was conducted in the western Ross Sea in January 2014 with the aim to estimate krill abundance after a time lapse of 10 years; synoptically with acoustic sampling, oceanographic parameters were collected by means of CTD probe at dedicated stations. The information about krill biomass and its spatial distribution in this area was analysed together with environmental parameters in order to explore possible relationships. A negative correlation between E. superba density and salinity was found in the water column, coupling acoustic and CTD information and also a positive correlation between E. crystallorophias and fluorescence was found, confirmed also in thematic maps of krill spatial distribution and fluorescence interpolated in the study area.

46 Keywords:

Krill; acoustics; Ross Sea; spatial distribution; oceanography.

- 60 **1. Introduction**
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62 The Ross Sea in Antarctica can be considered one of the last places on earth that remains almost 63 unaffected by the changes occurred after the industrial revolution, even if this is more true for the shelf area 64 respect to the slope one (Ainley, 2010). Other Antarctic regions, instead, start to show traces of impacts, from 65 human related activities and global warming in general, such as the Antarctic Peninsula and West Antarctica 66 region (Abram et al., 2013; Steig et al., 2013). The marine ecosystem of the Ross Sea in Antarctica has been 67 object in the past of several research projects and papers aimed to try to understand the complex dynamics 68 undergoing among the different organisms living there (Ryan & Cooper, 1989; Ichii et al. 1998). Several 69 acoustic surveys conducted in the western Ross Sea (Azzali & Kalinowski, 1999; Azzali et al., 2000; Azzali et 70 al., 2006) have put in evidence the consistence in terms of abundance of two euphausiids species in this area: 71 Euphausia superba and Euphausia crystallorophias. The euphausiids spatial distribution obtained by these 72 surveys seems strongly influenced by ice presence. In fact it could be seen how in November, when ice covers 73 the most part of western Ross Sea, the bulks of the two species are very near and located around latitude 74° S, 74 while in January, when polynya ice-free areas are at maximum extension, E. superba moves significantly to the 75 north, while E. crystallorophias do the same but over a much shorter extent. These two species together with 76 other pelagic species, secondary in occurrence, constitute a basic link between planktonic primary and 77 secondary production and top predators (Lyver et al., 2011; Murase et al., 2013; Lee et al., 2013). Being krill a 78 so important resource for a huge number of animals, studying the factors that could enhance or reduce its 79 reproductive and survival performances results crucial.

80 The environment has proven to be an important factor conditioning krill survival in Antarctica and in 81 other regions (Smith et al., 2012; Flores et al., 2012); in particular ice coverage seems to have a positive 82 influence on feeding and reproduction in krill species and also in achieving a successful competition for space 83 and food with salps (Siegel & Loeb, 1995; Loeb et al., 1997; Nicol, 2006; Lee et al., 2010). Moreover water 84 temperature rising in the last years may have potential influences on krill biomass and spatial distribution; 85 some statistical models indicate a possible reduction of around 20% in the area in which krill may be found 86 (Hill et al., 2013); this is due not only to a suitable temperature range for krill survival, but also to the ice 87 decrease consequent to global warming as reported above. Another result that seems linked to what reported 88 above is that of Fielding et al. (2014) who found a significant correlation between krill density during austral 89 summer and Sea Surface Temperature (SST) of the previous winter; once again krill density resulted positively 90 influenced by cooler winters.

For what concerns salinity, when this parameter presents low values it seems that we may have areas with high presence of Antarctic krill due to ice melting (Murase et al., 2013). Even Tarling & Thorpe (2014) found that the displacement magnitude of an Antarctic krill swarm was positively affected by the swarm area, vicinity of ice-edge and salinity; in particular the highest responses were found for large swarms and lowsalinity regions.

The tendency registered for the Ross Sea shelf area from 1950's to more recent yeas showed an increase in air temperature and a decrease in shelf water salinities (Jacobs & Giulivi, 1998). Moreover, recently the study of water masses exchange at continental slope level, implicated in Antarctic Bottom Water (AABW)
formation demonstrated a change in the thermohaline characteristics on a decadal time scale (1995-2006) for
western and central Ross Sea (Jacobs et al., 2002; Budillon et al., 2011). In particular Ice Shelf Water (ISW)
freshened and its mean temperature increased (central Ross Sea) and the High Salinity Shelf Water (HSSW)
freshened too except for the 2003 saltier anomaly (western Ross Sea); this phenomenon influences the salinity
and density of AABW bringing cascade changes that may impact on the food web.

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105 **2. Materials and methods**

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107An acoustic survey for the evaluation of the biomass of Middle Trophic Level species in the Ross Sea108was conducted in January 2014 in the framework of the XXIX Italian Expedition to Antarctica organized and109coordinated by the Italian National Programme for Research in Antarctica (PNRA). The main aim was to110estimate the spatial distribution and abundance of Middle Trophic Level species and particularly *E. superba*111and *E. crystallorophias*.

112 The study area was monitored acoustically at three frequencies (38, 120 and 200 kHz) by means of a SIMRAD EK60 scientific echosounder on board R/V Italica; the frequencies in use are in agreement with the 113 basic guidelines for acoustic surveys identified by CCAMLR. Periodical pelagic trawls were performed 114 115 targeting the key species with the help of direct recognition of the two krill species on echograms at three 116 frequencies and with improved efficiency in capture due to the connection between the echosounder and the 117 integrated trawl monitoring system SIMRAD ITI, giving information on net position in the water column; in 118 this way the net could be cast with high precision on the swarms of interest visible on the echograms (Figure 119 1). The main acoustic settings used at the three frequencies are reported in Table 1.

Due to the limitation in the time period available for acoustic survey and to particularly bad weather conditions, the study area was reduced respect to other acoustic surveys conducted in the past by CNR-ISMAR of Ancona (Azzali et al., 2006). The ideal survey speed for acoustic survey, after noise test procedure, was identified in 8.8-9.5 knots, the variations in speed depended mainly on currents; monitoring was conducted during day and night and the only variations from standard transect routes were made for pelagic trawling and CTD sampling.

Hydrological casts and water sampling were carried out using a SBE 9/11 Plus CTD, with double
temperature and conductivity sensors, coupled with a SBE 32 Carousel sampler, carrying 24 bottles of 12 1
each. In Figure 2 the effective study area plus CTD and net samplings positions are shown.

130The pelagic net used to collect biological samples had a mesh of 1 mm in the codend and was equipped131with "V" type high performing pelagic doors. The biological samples collected consisted almost exclusively in132E. crystallorophias specimens in hauls 1 and 2 and E. superba specimens in hauls 3-5. Krill target species133individuals caught by the net underwent the following measures on board:

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Total catch weight

diameter of compound eye, sex determination

135 136 On a subsample of 100 individuals per species: Total length, carapace length, individual weight,

- 137 Acoustic data elaboration was accomplished through the scrutinization of echograms in LSSS software 138 discriminating pelagic species aggregations by means of multifrequency methodology. Proper filters were used 139 to remove noise originating from different sources (ice, rough sea, etc.) from echograms. Only those miles 140 monitored at survey speed were considered for the analysis. Krill swarms of the target species were identified 141 on the base of their specific frequency response profile (Azzali et al., 2004; Mitson et al., 1996); aggregations 142 from other scatterers were also defined and their frequency response analysed. In the cases of unknown 143 scatterers, hypothesis for scrutinization were made on the base of reference literature relative to the study area 144 (O'Driscoll et al., 2011), due to the lack of direct sampling with the net. In general, the other scatterers were: 145 other krill (probably Thysanoessa macrura), targets resonant at 38 kHz (probably Electrona antarctica) and a ground-truthed layer containing eggs and larvae not identified in detail. Acoustic density estimates in terms of 146 147 NASC (m²/nm²) were obtained per nautical mile, and also vertically stratified for 5 metres thick layers, in 148 order to be analysed together with environmental parameters in the water column in CTD vicinity. The vertical 149 limit for environmental parameters was set to 300 metres because this was the limit for echograms clean from 150 noise at all the frequencies in use (38, 120 and 200 kHz). The above cited data were log-transformed to 151 normalize the distribution and improve linearity of the variables for statistical analysis in the following way: 152 temperature values minus their minimum value were \log_{10} -normalized; fluorescence, salinity and oxygen 153 values were log₁₀-normalized; krill NASC values were log_{0.0001}-normalized. These data were analysed for 154 correlations by means of Pearson test. Krill spatial distribution was also analysed with environmental data 155 averages interpolated in the study area by means of IDW through thematic maps; ice coverage maps of the 156 survey period were also taken in consideration to try to understand krill dislocation.
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158 2.1 The Ross Sea water masses

159 Shelf waters in the Ross Sea generally have temperatures close to the surface freezing point (between -1.95 and 160 -1.75° C) and show salinity values higher in the western sector than in the eastern one. The high salinity in the 161 western sector could be explained by taking into account the large extent of sea-ice free areas (polynya) even 162 during the winter period (Kurtz and Bromwich 1983, 1985; Bromwich and Kurtz 1984, Budillon and Spezie, 163 2000). These areas, despite being involved in a large ice-formation process, are kept open by the wind action, 164 which takes away the ice as soon as it is formed. The rejected brine increases the salinity of the subsurface 165 waters, forming the High Salinity Shelf Waters (HSSW), the densest waters in the Southern Ocean. The 166 presence of the MCDW in the subsurface layer of the western sector of the Ross Sea plays an important role in 167 the HSSW formation: when the surface water freezes during the winter, the released brine is added to 168 subsurface waters that have relatively high salinity values due to the direct influence of the MCDW (Budillon 169 et al., 2011). HSSW is supposed to be formed during the winter particularly in Terra Nova Bay (Jacobs et al. 170 1985, Rusciano et al., 2014). Part of the HSSW moves north, reaching the continental shelf break, and takes 171 part in the formation of the AABW; other brunces may interact with the basal ice taking part in the formation 172 of a different water mass named Ice Shelf Water (ISW, Jacobs et al. 1970) characterized by temperatures lower 173 than the freezing point at the surface. The Antarctic Surface Water (AASW) is confined to the upper layer, 174 which became fresher and warmer during the summer because it was influenced by sea ice melting and by the 175 heat gained from solar radiation, which is the main constituent of the surface heat balance in summer in this 176 region (Budillon et al., 2003). Modification of incoming Circumpolar Deep Water (CDW) produces the 177 Modified Circumpolar Deep Water (MCDW) which intrudes across the continental shelf and it is the primary 178 source of heat and salt (Jacobs et al., 1985; Budillon et al., 2000) and nutrients (Smethie et al., 2005) for the 179 Ross Sea continental shelf. This inflow appears to be strongly influenced by the topography of the banks and 180 troughs along the shelf break (Dinniman et al., 2003) and, as expected, is often detected in the northern part of 181 the investigated area. Below this layer, the HSSW displayed a potential temperature close to the surface 182 freezing point and S>34.7.

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- 184 **3. Results and discussion**
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3.1 Krill spatial distribution and ice coverage

188 Sea-ice cover dynamics in the Ross Sea are reported in Figure 3; the evolution of the sea-ice cover is shown189 immediately before, during and after the acoustic survey.

- 190This figure shows how sea-ice free regions in the Ross Sea were quite developed since early January, leaving191place to a complete clearance by the survey period (23-25 January) with a quite large direct linkage between192the Ross Sea and the Southern Ocean.
- 193The acoustic density in terms of NASC (m²/nm²) of *E. superba* and *E. crystallorophias* is shown in Figure 4 as194bubble plot. Focusing the attention on krill species, the acoustic tracks performed at the northern border of the195Ross Sea with the Southern Ocean saw the almost exclusive presence of Antarctic krill, while the tracks made196in the central-western Ross Sea were characterized by ice krill alone.
- 197Part of the highest densities of Antarctic krill resulted in proximity with the residual ice still present near Cape198Adare; the importance of winter sea ice in particular to support and strengthen krill recruitment has been199widely demonstrated (Atkinson et al., 2004; Loeb et al., 1997; Flores et al., 2012*a*), but also, during austral200summer or late spring, krill can present high densities in the under-ice environments (Flores et al., 2012*b*) and201consequently higher abundances in areas freshly cleared from ice (Azzali et al. 2007).
- Ice krill was found south of Coulman Island, offshore Terranova Bay and Drygalski ice tongue. Unfortunately
 due to the reduction of the acoustic monitoring activity it was not possible to follow the situation in the area
 typically characterized by the overlapping between the two krill species.
- Generally speaking and accounting for the above mentioned spatial limits of this analysis, krill spatial distribution was in agreement with what found in the past when in January, respect to November-December, we assisted to an Antarctic krill, and to a minor extent ice krill, drift directed northwards in concomitance with the complete opening of the Ross Sea from sea-ice coverage (Azzali et al., 2006).
- The above shown spatial distribution of the two krill species could also be explained taking into account water masses dynamics in the Ross Sea; in fact the westward circulation along the shelf break seems to accumulate *E. superba* on the outer continental shelf and along the shelf break, while *E. crystallorophias* shows high

retention at Terra Nova Bay polynya and south of it; these results were obtained by Pinones et al. (2015)
 through Lagrangian particle experiments and are coherent to a certain extent with already cited observations in
 this area.

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3.2 Biometric characteristics of the target euphausiid species

Length frequency distribution of *E. superba* and *E. crystallorophias* in the surveyed area are reported in Figure 5; they come from measurements on individuals sampled with the pelagic net. The size frequencies found reflect the typical situation for this period in the western Ross Sea and includes juvenile and adult stages for both species.

Length-weight relationship was calculated on the base of biometric measurements on the samples; the results are given in Figure 6. Unfortunately this relationship was not very good for *E. crystallorophias*; this is because of the very small size of the individuals of this species, so that even the high precision scale (0.1 g) present on board was not precise enough for individual weight measurements.

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3.3 Krill spatial distribution and relations with environmental parameters

In Figure 7 and 8 respectively Antarctic krill and ice krill spatial distribution deduced from acoustic survey are
 represented together with the main environmental parameters considered separately and averaged in the first
 300 metres.

233 Considering E. superba (Figure 7) found at the northern border of the Ross Sea, it can be seen from the maps 234 that this species shows the highest concentrations where fluorescence presents lower values, while for 235 temperature and salinity the tendency is contrasting. The bulk of the density that is more northwards is located 236 in conditions of lower temperature and salinity respect to surrounding areas, while the other bulk presents 237 higher temperature and salinity respect to the nearby areas. Dissolved oxygen presents quite constant values in 238 the area characterized by Antarctic krill. It seems that this species concentrates more in proximity of the shelf 239 break, but it is not clear the link between the environmental parameters, at least considering averaged values 240 for environmental parameters and krill density data limited to a relatively small portion of the sea.

E. crystallorophias (Figure 8) seems to show a preference for lower salinity values and higher temperatures
 even if this pattern is not very much evident. The highest values registered for ice krill density, however, match
 quite well with an area characterized by a peak in fluorescence, and consequently in primary production. No
 particular pattern can be observed in relation with dissolved oxygen.

- Pearson test applied on krill density (NASC) data together with the main environmental parameters in thewater column and in CTD station vicinity gave two main correlations in terms of significance:
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- 1. Antarctic krill density salinity -0.60, p<0.01

Ice krill density – fluorescence 0.47, p<0.01

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The inverse correlation between *E. superba* density and salinity could be explained again with the association

251 of this species with areas recently cleared from ice, as to say fresher waters due to the ice melting (Tarling &

Thorpe, 2014). In fact, hydrographic features were spatially diversified along the study sections: although someareas were completely ice-free, some other areas were still influenced by late ice melting.

254 The positive correlation between E. crystallorophias and fluorescence is probably of a trophic nature; it is quite 255 reasonable that ice krill tends to concentrate where primary production is higher and this result is also evident 256 in the thematic maps reported above. Weber et al. (1986) found that, even if physical factors as water 257 temperature, do affect phytoplankton spatial distribution at medium/large scales, there is a link at a small scale 258 between phytoplankton and krill spatial displacement explainable as a predator-prey relationship. The fact that 259 this relation is not present for Antarctic krill could be due to the limitation of the spatial extent for krill density 260 data that don't allow us to have a more complete picture. It has to be taken into account, also, that the 261 instantaneous picture given by acoustic and oceanographic data is probably not enough to follow the complex 262 dynamics of krill and water masses in the area. For example, acoustic surveys made at a distance of one month 263 in 1994 brought very useful information on krill movements and polynya dynamics during austral spring 264 (Azzali et al., 2006).

- Anyway a sudden response by krill to high fluorescence values, and consequently greater chlorophyll concentrations, has been confirmed by experiments using acoustic data and net sampling on krill (Tarling & Thorpe, 2014); these experiments demonstrated a quite instantaneous effect of phytoplankton patches on krill displacement.
- Correlations similar to those found here were already obtained through multiple multivariate regression analysis applied on the same species and in the same area with 2004 dataset (De Felice, 2010), even if in that case *E. superba* density resulted related to water density and not salinity (but this two last parameters are often correlated) and *E. crystallorophias* was correlated with a-chlorophyll satellite data and not *in situ* fluorescence data. Anyway, as already said, interpolated in situ fluorescence data, even if not analysed statistically in detail, show a quite clear relation with ice krill abundance in 2014.
- 275 No particular correlations were found between krill density and temperature, maybe for similar limits to those 276 already mentioned above. In other areas other authors found relations between krill density and SST from the 277 previous winter (Fielding et al., 2014) and similar results considering historical series of data and applying 278 GAMs were obtained by Trathan et al. (2003). In general water temperature and consequently ice cover in 279 winter seems to have a strong effect on the following spawning and recruitment of krill (Lee et al., 2010), but 280 in order to verify if this works also for the western Ross Sea there is a need for an annual and more extended 281 survey. This delayed effect could also be due to more factors acting together as was found by Schroeder et al. 282 (2014); they saw a relation between previous winter temperature, salinity and particularly depth of 26.0 283 potential density isopycnal on subsequent krill spawning success in California waters. A similar mechanism 284 could be also hypothesized here considering the role of ice melting similar to the one given by upwelling in 285 California, releasing high abundances of primary producers with subsequent cascade effects on the local food 286 web.
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288 **4.** Conclusions

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290The acoustic survey performed in January 2014 in western Ross Sea, even if limited in extension, gave291important information concerning krill abundance and spatial distribution. In particular the latter one

confirmed what already seen during previous researches in this area in the period 1989-2004. The possible relations between krill density and oceanographic variables could only be explorative due to the limitation in the studied area; anyway a significant negative correlation in the water column between *E. superba* density and salinity was found, probably connected to fresher waters from recent ice melting and also a positive correlation between *E. crystallorophias* and fluorescence, probably ascribable to trophic factors. These results can be useful to plan future deeper investigations on krill dynamics in this area in relation to environmental conditions.

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421	Tables and Figure Legends
422	
423	Table 1 Main settings used during the acoustic survey
424	
425	Figure 1 Echograms showing E. superba (right) and E. crystallorophias (left) swarms with lines representing
426	headrope and footrope of the pelagic trawl fishing on the aggregations. The example on the right clearly shows
427	the possibility to cast the net with high precision at the swarm stratum
428	
429	Figure 2 Geographic positions of pelagic trawls on krill aggregations and CTD stations in the study area
430	
431	Figure 3 Sea-ice cover dynamics during January 2014 in the Ross Sea and the surrounding area
432	
433	Figure 4 Spatial distribution of <i>E. superba</i> and <i>E. crystallorophias</i> in the study area
434	
435	Figure 5 Length frequency distributions in 0.5 cm classes for E. superba (left) and E. crystallorophias (right)
436	
437	
438	Figure 6 Length-weight relationships for E. superba (left) and E. crystallorophias (right)
439	
440	
441	Figure 7 Antarctic krill spatial distribution in relation with temperature, salinity, fluorescence and dissolved
442	oxygen (average values 0-300 m)
443	
444	
445	
446	Figure 8 Ice krill spatial distribution in relation with temperature, salinity, fluorescence and dissolved oxygen
447	(average values in the water column)
448	
449	

Table 1

Transducer type	ES38B	ES120-7G	200-7G
Beam type	split	split	single
Draft	6.5 m	6.5 m	6.5 m
Threshold	-85 dB	-85 dB	-85 dB
(display)			
Colour palette	BI500	BI500	BI500
Pulse length	1.024 msec	1.024 msec	1.024 msec
Ping rate	0.8 – 1.2 sec	0.8 – 1.2 sec	0.8 – 1.2 sec
Power	2000 W	250 W	150 W
Avg. sound speed	1461 m/s	1461 m/s	1461 m/s
Avg. absorption coefficient	10.3 dB/km	29.9 dB/km	43.0 dB/km
Noise estimation	-136 dB re 1 W	-140 dB re 1 W	-151 dB re 1 W
Data logging range	10-500 m	10-500 m	10-500 m







Figure 3

62.5

60°S

58·S

56'S





56's

Feb.92 2014 RossSea

W-071

Ice C □ C[%] 100







2.0

1.5

(6) ∦ 1.0

0.5

0.0

y = 0.0 $R^2 = 0$ Euphasia superba

3 LT (cm) Euphasia crystallorophia

2 LT (cm)

0.4

0.3

(IB) 0.2

0.1

0.0

y=0.013978 R²=0.61913

587 588

589

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Figure 8

