Distribution of *Rhincalanus nasutus* Giesbrecht 1888 (Calanoida, Copepoda) During the Eastern Sulu Sea Coastal Upwelling Season

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The distributional pattern of a large sized copepod, *Rhincalanus nasutus*, was investigated in Sindangan Bay and Dipolog Bay during a northeast monsoon-driven seasonal upwelling off the Northeast coast of the Zamboanga Peninsula, Eastern Sulu Sea. Three depth strata (0-65 m, 65-135 m, 135-200 m) at 10 stations were sampled for zooplankton using 100- and 200-µm meshed plankton nets with opening and closing mechanism. *Rhincalanus nasutus* ranked fourth most common taxon among the copepod-dominated samples. There was no significant difference between the abundance of the species caught in the two plankton nets. Adult females largely contributed to abundance peaks, while adult males comprised less than half the abundance of females. Younger copepodite stages (<CV) of both sexes were very few while naupliar stages were not observed. There was no significant difference in the depth-integrated abundance of *R. nasutus* between Sindangan Bay and Dipolog Bay, but it differed significantly with time of day. A crepuscular upward vertical migration is suggested by the early morning highest abundance in all depth strata sampled, and another smaller peak around sunset in deeper (65-200m) strata. Multiple linear regression analysis indicated highest *R. nasutus* abundance also associated with lowest tide and turbidity levels. A relatively homogenous set of conditions in environmental variables throughout the study area allowed us to speculate that the causes of such crepuscular behaviour may be attributed to low tide, least turbid waters, and position maintenance of *R. nasutus* in a highly advective upwelling system, but less likely associated with predator avoidance.

Key Words: abundance, distribution, Philippines, population, *Rhincalanus nasutus*, Sulu Sea

INTRODUCTION

The calanoid copepod *Rhincalanus nasutus* belongs to Family Eucalanidae which includes three other circumglobal genera: *Eucalanus, Pareucalanus*, *Subeucalanus* (Bradford-Grieve 1994; Mulyadi 2004). *Rhincalanus nasutus* is common in the tropical and subtropical regions of all oceans, inhabits epi- to bathypelagic depths (0-2000 m depth), and is also found in shelf and slope areas and in shallow coastal embayments (Schnack-Schiel et al. 2008). Very recently, Shimode et al.
(2012) noted that for Kuroshio Current spring populations, 88.5% of the total catch are mainly younger stages (C1-C4) found in 0-200 m depths at daytime. Adults are generally found >200 m depth at daytime. High densities of tropical *Rhincalanus nasutus* populations have been observed both day and night in the 0-150 m layer in the Banda Sea and Arafura Sea during upwelling events in February (Baars et al. 1990). Schnack-Schiel et al. (2008) noted that in the Red Sea and the Gulf of Aqaba, regardless of time of day, the bulk of the population are found at 400-500 m, but high densities of younger stages (C3-C5) are found in 0-200 m depths at daytime. Depths (>400 m) with oxygen concentrations below 0.01 mL O₂ l⁻¹ seem to limit the extent of its deepest habitat in the ocean (Sameoto 1986). Wishner et al. (2008) speculated that oxygen concentration, depth, sampling location, season, potential food availability, and predators influence *R. nasutus* distribution.

Reports on whether *R. nasutus* performs vertical migration differ (Schnack-Schiel 2008), with some (e.g., Koslow & Ota 1981; Castro et al. 1993; Palomares-García et al. 2013) describing the species performing diel vertical migration in near-shore areas, while others (e.g., Ohman et al. 1998; Farstey 2001 as cited in Schnack-Schiel et al. 2008) did not observe such behaviour. Summer upwelling in a shallow (<60 m) embayment of the Arauco Gulf, Chile, has been linked with offshore to inshore migration of the species (Castro et al. 1993). Maximum phytoplankton production tends to be associated with this coastal intrusion, and the vertical variation in abundance appeared to agree with tidal fluctuations (Castro et al. 2011). Subtropical populations from Kuroshio Current, Red Sea, Gulf of Aqaba, and California Current have been reported to perform ontogenetic vertical migration that involves an ascent of copepodite stage V in spring and reproduction in surface layers (Schnack-Schiel et al. 2008; Shimode et al. 2012). After the spring season, late copepodite stage V rich in wax esters descend back to deeper layers and become dormant (Schnack-Schiel et al. 2008; Shimode et al. 2012). Changes in light, temperature, integrated chlorophyll concentration in the top 200 m, stored lipid levels, predation, and endogeneous reproductive behaviour could trigger emergence from and entering into dormancy (Schnack-Schiel et al. 2008; Shimode et al. 2012).

An important prey for pelagic fish and invertebrates themselves (Lluch-Cota et al. 2007; Takagi et al. 2009; Espinoza et al. 2009), *R. nasutus* is predominantly herbivorous (Mullin 1993; Schnack-Schiel et al. 2008), but may also be omnivorous (Irigoin et al. 2005). The Banda Sea population feeds intensively at night (Baars et al. 1990), while those inhabiting the California Current seem to display crepuscular feeding as evident by early evening and morning peaks in gut fluorescence (Ohman 1988).

There has been little information on the ecology and behaviour of *R. nasutus* in the tropics, particularly in Philippine waters. Information on *R. nasutus* in the northeast monsoon winds-driven seasonal upwelling in Eastern Sulu Sea would help in understanding the relationship between oceanographic processes including zooplankton production and the sardine fisheries in the area. This study aims to examine the spatial and temporal patterns in the abundance and population structure of *Rhincalanus nasutus* in the upper 200 m in the eastern Sulu Sea during an upwelling event. Environmental parameters of the study area were also concurrently observed to examine their relationships with the abundance of *R. nasutus*.

**MATERIALS AND METHODS**

The two sampling sites of this study are situated in Eastern Sulu Sea, particularly in the waters off the Northeastern Zamboanga Peninsula in Mindanao, Philippines, with the western site located within the vicinity of Sindangan Bay (8.24-8.35°N, 122.85-122.95°E) while the eastern site was at Dipolog Bay (8.55-8.64°N, 123.22-123.28°E) (Figure 1). The topography and oceanographic features of the study area, and Northeast monsoon winds have been reported to cause an Ekman transport-driven coastal seasonal upwelling that occurs from December to March (Villanoy et al. 2011). This upwelling area supports a seasonal sardine fishery which is dominated by *Sardinella lemuru* or the Bali sardine.

On board a rented commercial fishing vessel, a physical and biological oceanographic cruise was conducted on 4-7 February 2012 in the sampling sites (Table 1). Five stations in each site were reached following a consecutive mixed day/night sampling scheme. Every sampling station was marked using a hand-held Global Positioning System unit. At each sampling station physico-chemical parameters (temperature, salinity, dissolved oxygen, chromophoric dissolved organic matter (CDOM), turbidity, chlorophyll a, depth) were observed in the upper 200 m using a General Oceanics (Florida, USA) multi-parameter vertical profiler. The chromophoric dissolved organic matter parameter measures dissolved tannins while turbidity determines the amount of suspended living and non-living particles.

Immediately after the observation of physico-chemical parameters, vertical collection of zooplankton was made using a paired North Pacific (NORPAC) net both having 0.45 m mouth diameter but differed in mesh sizes; one with 100 μm and the other 200 μm. These nets were used to ensure collection of small and large size fractions of zooplankton. A General Oceanics opening and closing mechanism was attached onto the towing line so that three
Table 1. List of sampling stations for North Pacific-net tows in Eastern Sulu Sea, off Northeastern Zamboanga Peninsula, Mindanao, Philippines. At all station stratified vertical tows were made in 200-135, 135-65, and 65-0 m layers with combined 100- and 200-µm nets.

<table>
<thead>
<tr>
<th>Station Number</th>
<th>Location*</th>
<th>Date</th>
<th>Local Sampling Time (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>8.27°N</td>
<td>122.89°E</td>
<td>5 February 2012</td>
</tr>
<tr>
<td>2</td>
<td>8.34°N</td>
<td>122.86°E</td>
<td>5 February 2012</td>
</tr>
<tr>
<td>3</td>
<td>8.34°N</td>
<td>122.90°E</td>
<td>5 February 2012</td>
</tr>
<tr>
<td>4</td>
<td>8.35°N</td>
<td>122.95°E</td>
<td>5 February 2012</td>
</tr>
<tr>
<td>5</td>
<td>8.24°N</td>
<td>122.94°E</td>
<td>5 February 2012</td>
</tr>
<tr>
<td>6</td>
<td>8.55°N</td>
<td>123.22°E</td>
<td>6 February 2012</td>
</tr>
<tr>
<td>7</td>
<td>8.60°N</td>
<td>123.27°E</td>
<td>6 February 2012</td>
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<td>8.60°N</td>
<td>123.22°E</td>
<td>6 February 2012</td>
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<td>9</td>
<td>8.64°N</td>
<td>123.23°E</td>
<td>7 February 2012</td>
</tr>
<tr>
<td>10</td>
<td>8.63°N</td>
<td>123.28°E</td>
<td>7 February 2012</td>
</tr>
</tbody>
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* Approximate location of starting point
successive depth strata (0-65 m, 65-135 m, 135-200 m) were sampled. Towing velocity was between 0.64-0.81 m/sec. Filtered volume was computed by multiplying the vertical distance covered by the area of net’s mouth assuming 100% filtering efficiency. Zooplankton samples were fixed in 5% borax-buffered formaldehyde in filtered seawater, sealed and stored at room temperature for later analysis. From these samples the relative abundances of higher taxonomic groups were enumerated. Analyses on the distribution of the entire zooplankton assemblage are presented in Metillo et al. (2013). All *R. nasutus* were sorted out from the samples and their sexes and stages were identified under a stereomicroscope following Mori (1964), Bradford-Grieve (1994), and Mulyadi (2004). The abundance of all life stages of *R. nasutus* was expressed as number of individuals per cubic meter for the temporal analysis, but number of individuals per square meter was used for the spatial analysis for easy comparison of oceanic surface layer (0-200m) standing stock values reported in other studies (e.g., Schnack-Schiel et al. 2008; Shimode et al. 2012).

The abundance of all life stages of *R. nasutus* from the two net types was tested for similarity using two-tailed *t*-test with homoscedastic variance available in the Microsoft Excel software version 2003. Averages and standard deviations were computed for stations and depth strata with comparable abundance also using the same Excel software. Non-parametric Mann-Whitney *U*-test and Kruskal-Wallis *H*-test available in SPSS version 16 (SPSS, 2006) were used to compare abundance data between sampling sites and sampling time, respectively. The possible relationship between log-transformed *R. nasutus* density and physico-chemical parameters (temperature, salinity, dissolved oxygen, CDOM, turbidity, chlorophyll *a*, depth) was analysed using multiple linear stepwise regression also available in SPSS version 16 (2006).

RESULTS

Figure 2 shows vertical profiles of the physico-chemical parameters at the 10 sampling stations. Temperature profiles were similar across 10 stations (Fig. 2A), but variations were observed with other parameters at stations closest to shore at near surface depths (0-20m). For instance, low salinity values (0-10m) (Fig. 2B) and high CDOM (0-5m) (Fig. 2C) at stations 6 and 7 in Dipolog Bay. High turbidity was recorded at stations 6 and 7 still in Dipolog Bay and stations 4 and 5 in Sindangan Bay (Fig. 2D). In Sindangan Bay, low values were recorded for dissolved oxygen at station 1 (Fig. 2E) and chlorophyll *a* at stations 1 and 2 (Fig. 2F). The temperature profiles indicate a surface mixed layer in the upper 80 m (26.3–27.0° C) and a sharp decline of temperature underneath to reach 15.4-15.7° C at 200 m. Dissolved oxygen and chlorophyll *a* showed highest values in near-surface (0-25m) layers. The stations in Sindangan Bay showed relatively higher values of near-surface chlorophyll *a* (0.12-2.34 mg m⁻³) than those in Dipolog Bay (0.12-2.05 mg m⁻³). A mixed tidal pattern was observed throughout the sampling period, with peak heights occurring just after sunset (19:00 h) while lowest tides near sunrise (Fig. 3). A minor high tide occurred in the morning at around 08:00 h.

Abundance of *R. nasutus* caught within station using 100- and 200-µm meshed plankton nets was not significantly different for adult females (*t*-test, *p* = 0.42), adult males (*t*-test, *p* > 0.28), and juveniles (*t*-test, *p* > 0.19). Thus, data presented in the temporal and spatial analyses are averages of counts obtained from the two nets. *Rhincalanus nasutus* was found ranking fourth with a relative abundance of 1.47% as against the other zooplankton categories in all samples. In decreasing order of percent relative abundance, the other top 9 categories are: Copepoda copepodes (78.5) > Ostracoda (4.24) > Chaetognatha (2.43) > Bivalvia molluscs (1.2) > Gastropoda molluscs (1.06) > Copepoda nauplii (0.82) > Polychaeta (0.54) > fish eggs (0.53) > Decapoda larva (0.52). The 0-200m depth-integrated mean abundance of *R. nasutus* revealed that at all 10 stations females dominated with the highest value of 3306 inds. m⁻² at station 2 and the lowest (59 inds. m⁻²) at station 4 (Fig. 4). Males comprised less than half the abundance of females, while juveniles (copepodite III–V) of both sexes were few and naupliar stages were not observed (Fig. 4). The maximum total abundance of all life stages at station 2 was 6175 inds. m⁻².

Combined depth-integrated abundance data within Dipolog Bay were not significantly different for all life stages (females: *U* = 59, *p* > 0.24, males: *U* = 45, *p* > 0.35, juveniles: *U* = 36, *p* > 0.14) as against those from Sindangan Bay suggesting horizontal homogeneity. Meanwhile, significant differences were observed in the abundance between the sampling times (early morning, morning, midday, evening and midnight) in all females (*H* = 14.80, df = 4, *p* < 0.005), males (*H* = 13.32, df = 4, *p* < 0.009) and juveniles (*H* = 13.84, df = 4, *p* < 0.008). Female and male abundances were highest in the early morning, morning and evening but lowest at midday. However, juveniles were found highest only during the early morning and morning, and the rest of the day registered low numbers.

Abundance of females, males, and juveniles collected at various times revealed a crepuscular pattern with an early morning peak (06:49-07:47) in the three depth strata sampled, and another elevated abundance just after sunset (17:44-18:33) at deeper strata (65-200m) (Fig. 5). Individuals were mostly found at the deepest strata (135-200m) from late evening to around midnight. Multiple
Figure 2. Vertical profiles of temperature (A), salinity (B), chromophoric dissolved organic matter or CDOM (C), turbidity (D), dissolved oxygen (E), and chlorophyll \( a \) (F) in the 10 sampling stations off Zamboanga del Norte, Eastern Sulu Sea, Philippines. Profiles with black, blue, red, light gray, dark gray crosses represent stations 1-5 in Sindangan Bay, respectively, while brown, purple, red, green and black lines represent stations 6-10 in Dipolog Bay, respectively.
Figure 3. Tidal amplitude pattern throughout the sampling period from 4-7 February 2012. Tide reference point is at Jolo, Sulu, Philippines.

Figure 4. Distribution and abundance (individuals m$^{-2}$) of adult female (black), adult male (white) and juvenile (gray) *Rhincalanus nasutus* collected at the 10 sampling stations in Eastern Sulu Sea from the surface to 200m depth. Dashed and dotted broken lines represent 200 m and 500 m isobaths, respectively. Numbers in the semi-circle legend are x 1000.
**Figure 5.** Distribution and abundance (individuals m⁻³) arranged by time of day collected of adult female (white bars), adult male (hatched bars), and juvenile (black bars) *Rhincalanus nasutus*. Time interval indicates duration of sampling in three depth strata. Error bars = standard deviations.
linear stepwise regression analysis yielded statistically significant inverse relationships between densities of the different life stages of *R. nasutus* and both tide and turbidity (Table 2). The highest *R. nasutus* abundance was associated with lowest turbidity (0.20 ± 0.04 mg m⁻³) and tide levels. The rest of the physico-chemical parameters showed no statistically significant relationships with abundance values of the different life stages of *R. nasutus* (*p* > 0.11 for all).

### DISCUSSION

The physico-chemical parameters observed in this study indicate influence of the Northeast monsoon-driven tropical upwelling event in eastern Sulu Sea (Villanoy et al. 2011). The high chlorophyll *a* values in the near-surface layers are similar to those observed in the Southeast monsoonal upwelling in Eastern Banda Sea (Arinardi et al. 1990; Baars et al. 1990). The upwelling event in the sampling area differs from that of the upwelling event in Vietnam tropical waters which showed maximum chlorophyll *a* depths from the surface to a much deeper depth of 40 m, and lower maximum chlorophyll values of 1.2-1.6 mg m⁻³ (Dippner et al. 2007). A more productive upwelling in Eastern Sulu Sea may be linked to area-specific physico-chemical, meteorological and oceanographic processes that promote intense upwelling.

The maximum total abundance (6175 inds. m⁻²) of all life stages of *R. nasutus* in this study is comparable to those reported for the tropical population of *R. nasutus* in the 0-150 m layer in Eastern Indonesian upwelling waters (ca. 4000 inds. m⁻²: Baars et al. 1990), the spring populations that could reach 5000 inds. m⁻² in the 0-1500 m layer in the Gulf of Aqaba (Schnack-Schiel et al. 2008), and up to 6218 inds. m⁻² in the 0-1000 m right in the path of the Kuroshio Current in the waters near Japan (Shimode et al. 2012).

The dominance of females in the *R. nasutus* population in this study coincides with the observations in the subtropical Red Sea and Gulf of Aqaba (Schnack-Schiel et al. 2008) and in the northwestern Pacific to the east of Japan (Shimode et al. 2012). These studies noted that these females are mostly in the non-dormant, intensely feeding and most likely reproducing stage. A gonadal analysis may help ascertain reproductive stages of females in this study.

The depth-integrated spatial distribution of *R. nasutus* among the 10 sampling stations in Eastern Sulu Sea varied strongly with sampling time with highest abundance generally observed in the morning and another minor peak in the evening just after sunset. The highest abundance in the morning and the low abundance at night is more-or-less similar to the absence of day/night difference observed for subtropical populations (Castro et al. 1993; Schnack-Schiel 2008). Although...
not studied here, horizontal advection, vertical mixing, internal waves may have also influenced horizontal distribution of \textit{R. nasutus} population in the sampled parcel of water in Eastern Sulu Sea. For instance, the mechanism of position maintenance shown by other upwelling calanoid copepod species in other regions is that copepods minimize the effect of offshore Ekman transport by downward vertical migration to depths where inshore moving upwelling currents prevail (Peterson 1998). The interaction of endogenous behaviour of \textit{R. nasutus} and physical oceanographic processes needs further investigation in the study area.

Under the relatively homogenous set of conditions in the environmental variables throughout the study area, variation in \textit{R. nasutus} abundance appeared to be more closely linked with time of day, coupled with tide and turbidity, than the horizontal pattern of environmental variables. It is difficult from the present results to evaluate the relative importance of these three factors (time of day, tide, and turbidity). At least, however, the possibility of nocturnal upward migration in chlorophyll-rich near-surface layer may be highly unlikely, since the abundance peak of \textit{R. nasutus} in such a layer (0-65 m) was only observed in the early morning, a time not much advantageous in terms of predator avoidance, but not at night when predation pressure by visual predators may be much lower. As a related, but not identical, phenomenon, Ohman et al. (1988) reported that California Current \textit{R. nasutus} populations peaked in gut fluorescence at night, with peaks just after sunset and before sunrise, and they attributed such nocturnal feeding to endogenous feeding rhythms and vertical migration into the euphotic zone. Because the data presented here is from a mixed day/night sampling scheme, there may be a need for continuous sampling at regular intervals for 24 hours or more at fixed stations. The early morning abundance peaks in the entire sampled strata were associated with low tide and lowest turbidity levels. The pattern of association between peaks at early morning (0200 –0800 h) and at evening (1600 – 2000 h) coinciding with the respective sinusoidal patterns of flood and ebb tides has been observed in the sub-tropical \textit{R. nasutus} populations from the Patagonian Gulf of Ancud, Chile, as a mechanism of population maintenance in food rich areas (Castro et al. 2011). The inverse relationship between \textit{R. nasutus} abundance and turbidity may be explained by the negative effect on copepod populations as a consequence of the light-limiting effect of turbidity on their phytoplankton food source (Schlacher et al., 2009) and reduced copepod feeding rates and egg production effects of increased levels of non-living lithogenic materials (Arendt et al. 2011). However, we did not find a negative correlation between turbidity and chlorophyll \textit{a} nor a positive correlation between copepod abundance and chlorophyll \textit{a}. Perhaps larger spatial and temporal scales of sampling may reveal direct evidence on the relationship between turbidity and \textit{R. nasutus} abundance. However, we can speculate negative effects on feeding, reproduction and behavioural avoidance in \textit{R. nasutus} individuals in response to highly turbid water. Furthermore, the high \textit{R. nasutus} abundance in less turbid waters may be explained by reduced zooplanktivity due to high predation risks on fish zooplanktivores at low turbidity conditions as predicted by a unimodal successful predation response to different levels of turbidity (Pangle et al. 2012). However, this reasoning is purely speculative in the absence of data on the species and abundance of planktivores and piscivores in the study site, and their behavioural responses to different turbidity levels and prey/predator densities.

We invoke the same influence of predation and turbidity in explaining the deeper \textit{R. nasutus} abundance at sunset. The deep (65-200 m) minor peak at sunset and a much deeper (135-200 m) abundance at night coincided with high tide and elevated turbidity which may be associated with intense zooplanktivity causing \textit{R. nasutus} populations to avoid the 0-65 m stratum. This reasoning, however, may contradict the assumption that predation risk by visual predators (like sardine) on \textit{R. nasutus} may be minimal at night (except when there is strong moonlight), regardless of the depth.

This study is limited by the small temporal and spatial scales of sampling both for \textit{R. nasutus} populations and hydrodynamic processes. Future studies on sampling deeper strata to ascertain vertical migration as against horizontal advection, the cycle and extent of the upwelling event, mechanisms of population retention and residency, the role of feeding behaviour and reproductive patterns would further enhance our understanding of the distribution of \textit{R. nasutus} in Eastern Sulu Sea.

ACKNOWLEDGEMENTS

We are deeply grateful to the Philippine Department of Science and Technology (DOST) for research funds; the crew of the commercial fishing vessel; the physical oceanography team led by Dr. Cesar Villanoy of University of the Philippines Diliman; and the University of Tokyo, Japan for the international travel funds support to the senior author who presented this paper at the Seminar on Coastal Ecosystems of Southeast Asia 2012 held in Jakarta, Indonesia. This study was also partially supported by Indonesian Institute of Sciences (LIPI) and Japan Society for the Promotion of Science (JSPS: Asian CORE Program).
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