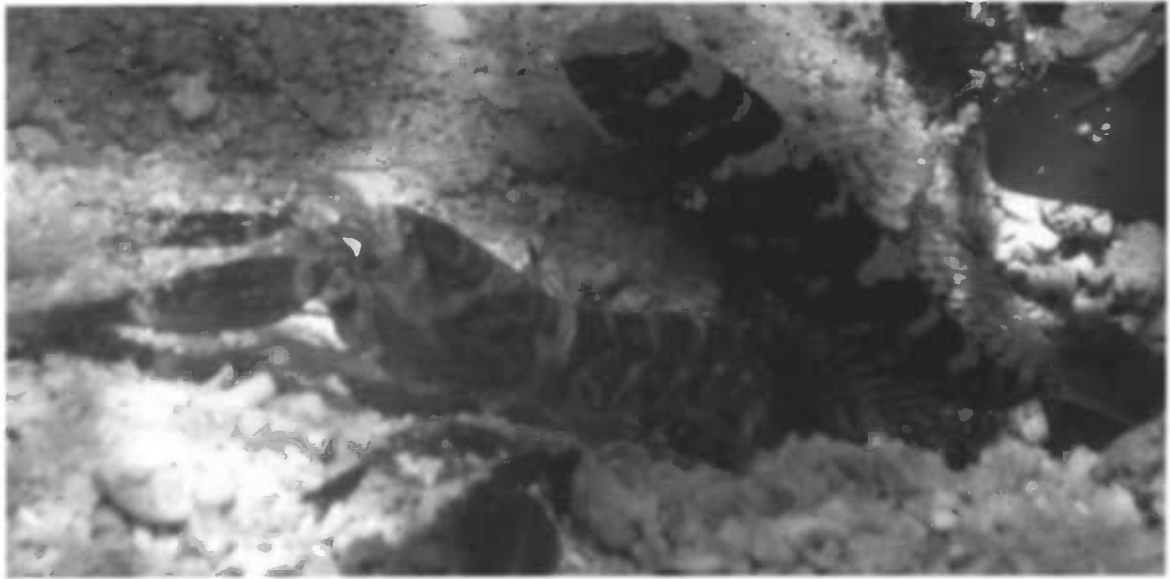# *Alpheus edamensis*: the gardeners of the Indonesian Archipelago



Nutrient cycling by *Alpheus edamensis* in *Thalassia hemprichii* dominated seagrass beds in the Indonesian Archipelago

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## Abstract

The harvesting behaviour of *Alpheus edamensis*, in the seagrass beds in the Indonesian Archipelago has been investigated to gain a greater understanding of the persistence of seagrass beds, nutrient sinks/sources, in a nutrient poor environment. The harvesting of the daily produced biomass of *Thalassia hemprichii* was based on the pilot of Stapel and Erftemeijer (2000). In total 75 observations of 15 minutes each were made, 25x25cm quadrants of seagrass, 50x50 cm quadrants of loose leaves were collected and water samples were taken from interstitial sediment pore water as well as the water column. During this time we looked at sediment reworking and harvesting. The nutrient concentrations, Nitrogen and Carbon, of *T. hemprichii* were measured at the Radboud University in Nijmegen, the Netherlands. Water concentrations were measured with the help of Hasanuddin University (UNHAS) in Makassar, Indonesia. The gathered information resulted in an estimated 17.2% of the daily production of *T. hemprichii* being harvested by the *A. edamensis*. The pilot experiment estimated that 53% of the daily produced seagrass was harvested. We suggest that this difference is mainly caused because of the different season conditions. An unusually large storm hit Makassar around mid December, which was half through the experiment. This made it impossible to conduct research there for 10 days, while the storm passed. A large amount of sediment was deposited on the seagrass bed. This in turn buried much of the *T. hemprichii*. More than 40% of the normally aboveground produced leaf was under the surface and therefore unavailable for harvest by *A. edamensis*. This situation was temporary and unique.

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## 1.    Introduction

This report covers my field research on nutrient cycling in seagrass beds by alpheid shrimps. It took place from November 2003 until February 2004 in Sulawesi, Indonesia as part of my doctoral studies as a marine biologist. This research was conducted at and supported by the Faculty of Mathematics and Natural Sciences at Groningen State University (RuG) in the Netherlands, the Faculty of Environmental Studies at the Radboud University (RU) in Nijmegen, the Netherlands and the Hasanuddin University (UNHAS) in Makassar, Indonesia. This research was supervised by Prof. Dr. W.J. Wolff (RuG), Prof. Dr. A. Noor (UNHAS), Dr. J. Stapel (RU) and J.A.Vonk, Drs. (RU).

### 1.1    Problem definition and justification

The marine environment found in the tropics is generally nutrient-poor with the exception for areas close to the coast and the areas nearby the discharge of rivers (Erftemeijer and Herman 1994). Seagrass beds occur in shallow coastal areas around the world (den Hartog 1970). The generally high productivity of seagrasses, paralleled by a high nutrient demand in nutrient-poor environments, has attracted attention since the expansion of seagrass research in the early 1970s (Stapel *et al.* 2001). Islands, away from the shore, such as our study area are considered to be lacking seasonal river and anthropogenic influences (Erftemeijer and Herman 1994). It would be expected that the organisms residing in such a nutrient poor environment have adopted strategies to be efficient with the nutrients available. For seagrasses this is not always straightforward because, none of the nutrient efficient strategies hypothesized are strongly developed in seagrasses (Stapel & Hemminga, 1997, Hemminga, *et al.* 1999). One such strategy hypothesis is: "Plants with slow leaf turnover rates have lower annual nutrient requirements" (Chapin, 1980). Other suggested strategies for higher "Mean Residence Times" of nutrients in leaves are 1) reducing litter production, 2) synthesizing low-nutrient tissues and 3) efficient nutrient resorption from senescing tissues (Aerts, 1990). Seagrasses do not use these strategies, on the contrary, they have a low leaf life span, synthesize most of their nutrients in the leaves and reabsorb fewer nutrients than terrestrial plants (Hemminga *et al.* 1999).

A high turnover rate should not be cause for much of a problem in leaf, or nutrient economics. The problem of the nutrient dynamics arises from the export of these loose leaves from source seagrass beds to their subsequent deposition in nearby unvegetated areas due to water currents. Because of the long-term

persistence of these seagrass beds, the nutrient loss caused by this export of loose leaves cannot be higher than the nutrient input in the seagrass bed, otherwise all seagrass beds would cease to be. So how does a seagrass bed become a significant sink/source for nutrients?

Organisms on a seagrass bed can recycle loose leaves and detritus over time. Griffis and Suchanek (1991) mentioned the harvesting of leaf material by the thalassinid shrimps as a significant nutrient sink. Duarte (2005) refers to seagrass beds as one of the "hot spots" in the ocean for carbon burial by benthic organisms. However, little is known about the influence of burrowing organisms, such as *Alpheus edamensis* (de Man, 1888), and the consequences it may have on the nutrient conservation strategies in seagrasses.

The alpheid shrimp and its symbiotic partner a gobiid fish (Stapel and Erftemeijer 2000) reside in mixed tropical seagrass beds. Stapel and Erftemeijer (2000) found, in a pilot study, that the alpheid shrimps' harvest could consist of 53% of the daily production of the seagrass *Thalassia hemprichii* and 45% of daily-incorporated leaf nitrogen in mixed seagrass beds, dominated by *T. hemprichii* in the Spermonde Archipelago, near Makassar, Indonesia. These findings thus suggest a major pathway of which little is known in the nutrient cycle of the mixed seagrass beds.

Seagrass beds display seasonal changes much like terrestrial plants. They have annual cycles where growth rates, shoot densities and root/shoot ratios change (Lin and Shao, 1998). These seasonal fluctuations can differ per location and seem to significantly affect benthic invertebrates (Erftemeijer and Herman, 1994). Our experimental research was conducted during a time when a particularly big storm hit our research area. It is important to realize that our experiment is conducted during a different time of the season than that of the pilot study mentioned above and that availability of plant material fluctuates over the year.

As there is little information on how burrowing organisms influence the nutrient cycle in seagrass beds, the main goal of this research was to establish the amount of seagrass the alpheid shrimp collects on a daily basis. The fieldwork took place during the months November, December and January 2003-2004. The research design was based on previous research done by Stapel and Erftemeijer (2000), the scientific literature available, and the suggestion of a major pathway for nutrients, a possible sink. A "sink" is an area where nutrients accumulate over time and a "source" is an area where nutrients leave at a greater rate than they accumulate. A secondary goal was to establish a scientific basis for observing the shrimp and its behavior in the seagrass beds for future research.

## 1.2    Location

As a part of this research report it is also import to convey a bit of history and social background of the research area.

Indonesia is an archipelago of more than 17,000 islands (6,000 inhabited) that straddles the Equator. It has a population of over 242 million (July 2005 est.) of which 27% has an income below poverty (1999) (CIA-The World Factbook: http://www.cia.gov/cia/publications/factbook/geos/id.html). (These and more facts about Indonesia can be found on this website.)

The research was performed around Bone Batang (5°01' S, 119°19'30"E), a small island off the coast of Sulawesi, approximately 15 kilometer from the coastline and the city Makassar. Bone Batang is located north of the populated island Barang Lompo and is a small island without trees, terrestrial grasses and humans. The island consists mainly of coral rubble and sand, with seagrass meadows and corals in the surrounding waters. The seagrass meadows consist of the following species: *Thalassia hemprichii, Halodule uninervis, Halophlia ovalis, Enhalus acoroides, Syringodium isoetifolium* and *Cymodocea rotundata* (Vonk, *Unpublished*). This mixed species seagrass system also includes several macro algae species like *Sargassum sp.* and *Padina sp.* and is partially surrounded and protected by shallow coral reefs that fringe about ¾ of the Island.

The human population from the other islands frequently visits this seagrass bed at Bone Batang. They search for sea cucumbers, gastropods, and other potentially useful materials. Unfortunately things like buoys and herrings used for marking of the experimental sites are also seen as useful materials. The reef around it is used for fishing of all sorts. These include traditional fishing, bomb fishing and poison fishing. These last two practices of fishing are generally very harmful to the marine ecosystems.

Our study area was marked with bamboo sticks. We tried herrings with buoys attached to them, but those never lasted for very long. The area we studied was dominated by *T. hemprichii* with small patches of the other species mentioned above. All of the observations took place inside the *T. hemprichii* dominated areas. The site depth ranged 40 cm to 80 cm due to tidal difference at different days.

### 1.3    Weather conditions and tides

In general the study area is governed by a tropical climate with a distinct dry and wet season. Each year from November until April, northwesterly winds dominate and usually bring 70% of the total annual precipitation (Erftemeijer *et al.* 1994). The weather conditions changed rapidly during the research period in Indonesia, which lasted from November 2003 until January 2004. Halfway through the research period, around mid December, a heavy storm hit the research area. Storms are an annual occurrence in Indonesia during the wet season, also known as the rainy season. This storm, however, blew away a cabin that was present on Bone Batang Island for four years and was according to the local populace of exceptional force. The first part of the research covering the behavioral observations, was done before the heavy storm, other parts such as the seagrass density measurements, were done before and after the heavy storm. The nutrient concentrations were measured only afterwards.

The climatologically results, are shown in figure 1, which were taken from Houston Chronicle:

http://weather.chron.com/auto/chron/global/stations/97180.html). (This website keeps track of the weather conditions in Makassar; e.g. wind speeds, temperature, daylight hours, etc.).
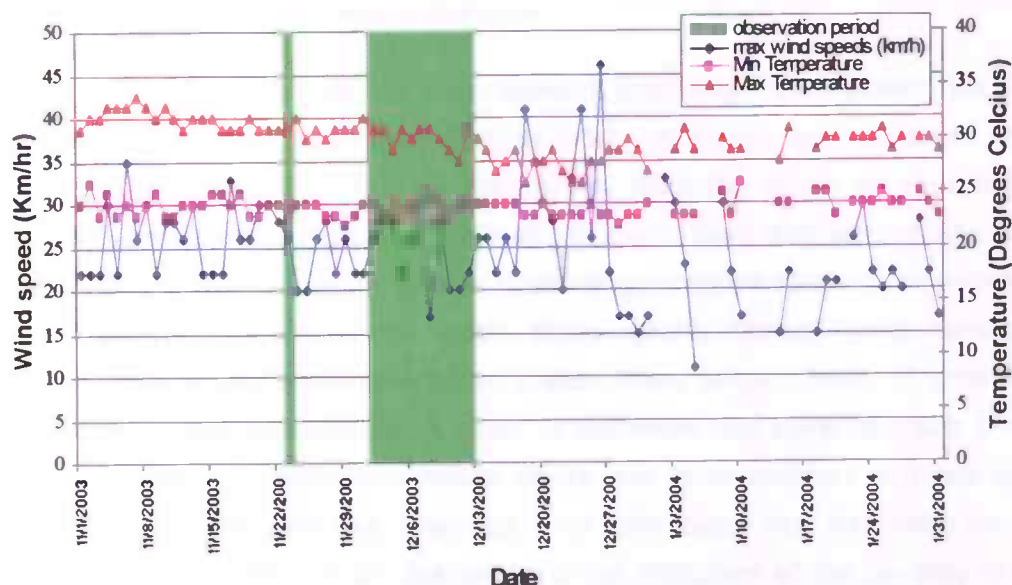
## Weather conditions and observation period



**Fig. 1** – Weather conditions measured in Makassar during observation period. These include Minimum Temperature (°C), Maximum Temperature (°C), and Wind speeds (Km/Hour). During the storm period some points are missing.

The predicted tide of P. Karangrang Lompo, figure 2, was created with the tidal information provided by Makassar Port.

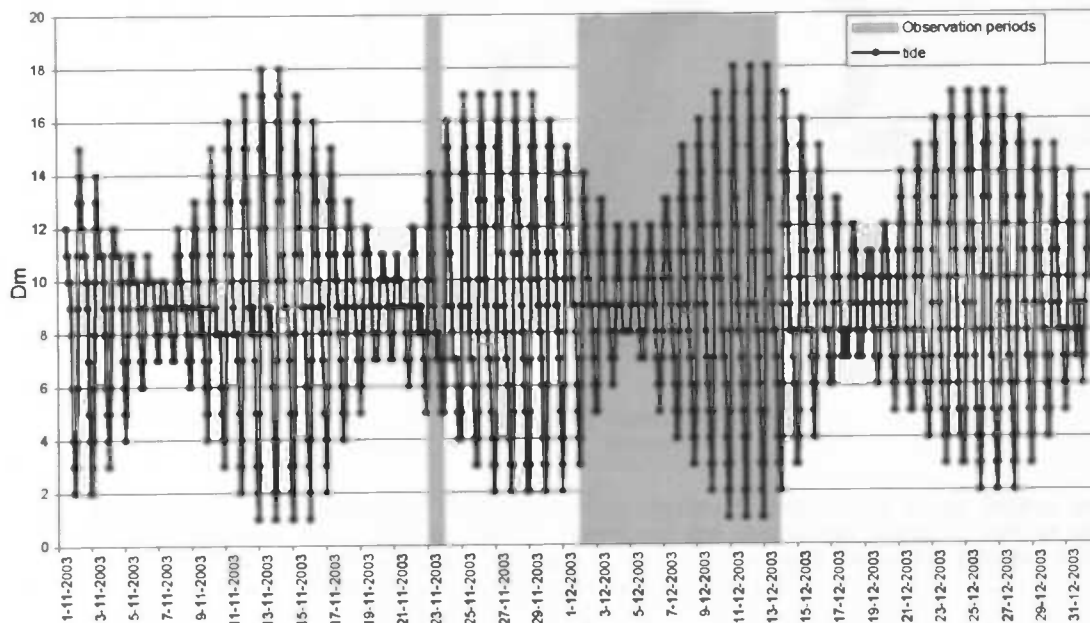## Predicted tide of P. Karangrang Lompo (An island north of Bone Batang)



**Fig. 2** – The predicted tidal oscillation curves at P. Karangrang Lompo in decimeters (Dm) for the period November till December *2003; P. Karangrang Lompo is nearby the research area. Observations were done for almost an entire cycle from 2-12-2003 till 14-12-2003, during neap tide. Circatidal in Makassar = 23.8 hours.

The heavy storm made our field research impossible for 2 weeks, because it prohibited any small seaworthy fishing boat, including our transport from Makassar to the island, from leaving the harbor. After the storm we returned to the research area to find mass mortality of sea cucumbers and sea urchins. It is worth mentioning that we found several types of gastropods shells in an excellent condition, suggesting their recent death. Water quality allowed for 5 meters of vision and much of the water was tinted green. Many large schools of small fish were present. It also seemed that a layer of sediment had covered much of the plant life (see Fig. 3), while the island above sea level seemed to have been eroded away. The fact that the small but very solid cabin that had been on the island for many years did not survive was another indication of the severity of the storm.

In the days without storm, field research was conducted from 8:00 am till 1:00 pm. After 1:00 pm the winds started picking up and the sea became rough. Our research vessel, a small boat, had thus far not been tested on large waves.

At our experienced Makassarese boatman's advise, we chose safety above all and returned before the winds and waves became too dangerous.
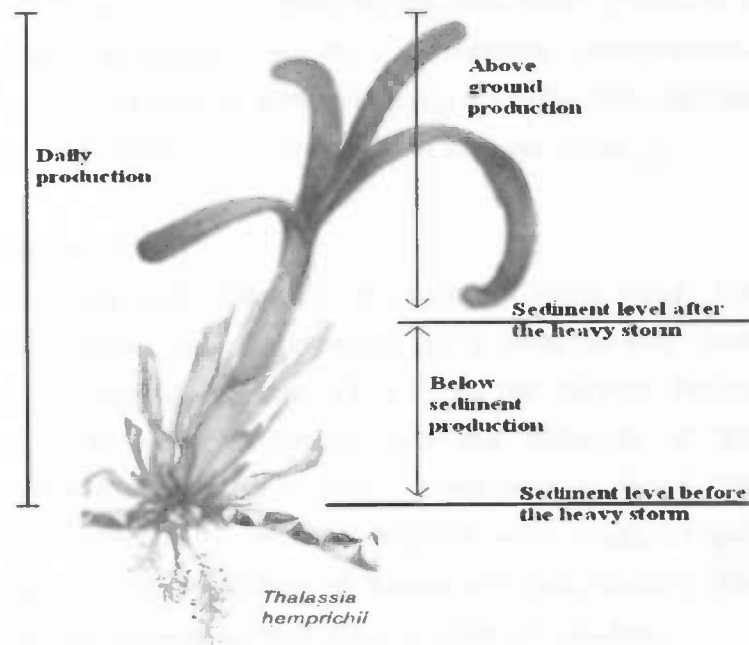


**Fig. 3** - *Thalassia hemprichii* - as taken from Richmond (2002). The figure depicts the increase sediment level after the heavy storm and the consequent burial of part of the seagrass. Also indicated are above ground and below ground leaf production plus the daily production, which was based on the dry weight of the whole leaf.

## 2.    Research design and methodology

The main research question, as formulated before, has been divided in a number of concrete research activities, i.e. basic behavioral observations, measurements of densities, the catching of shrimps, seagrass collection, sample preparations and measuring the nutrients concentration in the sea water.

### 2.1    Basic behavioral observations

The following methodology and collection of materials were used. The research consisted of eight to twelve field observations being done per day. Each observation was done while snorkeling and took 15 minutes per burrow. During this period the behavior of an alpheid shrimp and the behavior of the accompanying gobiid fish were recorded on a plastic sheet with a pencil. The recordings were done at Bone Batang. These observations were made to give insight into the activity pattern, the harvesting of leaves and bioturbation. The behavior was divided into several aspects as described in table 1 hereafter.

The size of the shrimp, the gobiid fish, the seagrass and loose seagrass leaves harvested was determined by having a 10 cm submersible tube, with markings for every centimeter, near the perimeter of the burrow. The gobiid fish was observed eating sand, epiphytes on leaves, and free-floating epiphytes and water, which might have been smaller epiphytes that were not visible to the observer due to minute size.

## Basic behavioral observations - overview

| Behavior | Designated letter | Description | Measured in |
|---|---|---|---|
| Sediment expulsion | R | Sediment expelled away from the burrow by the alpheid shrimp. | Number of occurrences |
| Incomplete sediment expulsion | H | Sediment expelled right in front of the burrow by the alpheid shrimp. | Number of occurrences |
| Burrow building | B | Burrow building consists of the alpheid shrimp dropping larger objects on top of the burrow or placing objects around the opening. | Number of occurrences |
| Seagrass cutting (cm) | C | The leaves that were cut by the alpheid shrimp and taken back into its burrow. | Centimeters estimated using a marked submersible tube. |
| Loose leaves harvesting | X | All loose leaves that was harvested by the alpheid shrimp and taken back into its burrow. | Centimeters estimated using a marked submersible tube. |
| Attempting to cut grass | T | All attempts by alpheid shrimp at cutting a seagrass leaf but failing followed by retreat into the burrow. | Number of occurrences |
| Disturbance (sec) | D | Disturbance of the shrimp was marked by a sudden retreat into the burrow usually combined with the gobiid fish retreat into the burrow. | Seconds the shrimp spends in the burrow after disturbance. |
| Fish attack | A | Several aggressive fish in the vicinity attacked researcher and shrimp alike. | Number of occurrences |
| Epiphyte eating | E | All attempts at eating large free floating and securely fastened epiphytes by the gobiid fish. | Number of occurrences |
| Gobiid fish Sand/Water eating | Z | Excessive intake of sand or water, which we presume to be filtering for edible materials. | Number of occurrences |
| Gobiid fish re-emerges | G | The re-emergence of the gobiid fish from the burrow. | Number of occurrences |
| Gobiid fish goes scouting | S | The gobiid fish was observed to temporarily leave the burrow to scout around the vicinity. | Number of occurrences |

**Table 1** – All observations done between 4-12-2003 and 14-12-2003 and based on preliminary field observations done on 23-11-2003. Basic observations as given in the first column were designated a letter for quick reference in the field. With the exception of actual harvesting and disturbance all occurrences were scored per minute. Harvesting was recorded in centimeters and disturbance in seconds.

### 2.2    Density measurements

Shrimp densities were determined by counting burrows inhabited by both shrimp and gobiid fish along a 1 by 10 meter transect (n = 7). This transect was created using 4 pegs and two pieces of string, each 10 meters long. A meter distance between the pegs was measured *in situ*. The area was left alone after placing such a transect to allow the shrimp and gobiid fish to become active again. If burrows were observed to be empty a new observation attempt was tried later that day. The observed lone shrimp (*Axius acanthus*), the alpheid shrimp and gobiid fish were marked on a slate with approximate coordinates. Three of these density measurements took place before the storm during the raining season, and four measurements were taken afterwards.

Some of the burrows contained more than one alpheid shrimp; this was only confirmed during basic behavioral observations. We could not be certain if a burrow had one, two, or more shrimp living inside; therefore all observations were recorded as activity 'per burrow' and not per shrimp. Very little is known about the pair-bonding of the *A. edamensis*, though some of the *Alpheus* sp. have been known to pair-bond for a long time, sometimes with as many as three shrimp per burrow (Karplus 1987). Though what should be mentioned in order to prevent an askew picture from being formed is, that this species is also believed to meet its partner underground via "coincidental" burrowing into a neighbors tunnel. The reason they think it finds its partner in such a way is because this *Alpheus* species closes its entrance to its burrow unlike *A. edamensis* does (pers. obs).

Burrow-casts were made by pouring liquid resin down the entrance of the burrow. The epoxy resin consisted of two components that were mixed on the spot and poured directly after mixing. The epoxy resin hardened over a two-day period after which it was dug out by hand. The excess coral attached to the hardened epoxy was chipped away with a hammer and chisel in order to establish the depth and the volume.

### 2.3    Catching of shrimps

During our stay on the seagrass beds, several attempts were made to capture the alpheid shrimp and the gobiid fish partner. The methods we used are described below and none of these methods had any success.

Capturing the pair was first attempted by using a diluted version of chlorine. Second, we tried undiluted version of chlorine. Third, we tried with a mesh cage (see fig. 4) positioned directly above the burrow equipped with a simple closing slide construction that could be operated at a distance of three

meters. This attempt at capturing the pair was combined with increasing the salinity around the burrow with NaCl to encourage evacuation of the burrow.

## Mesh Cage Construction



Mesh cage
sealed on all
six sides

Mesh lid that can
slide over the
opening

Opening

3-meter long rope used to
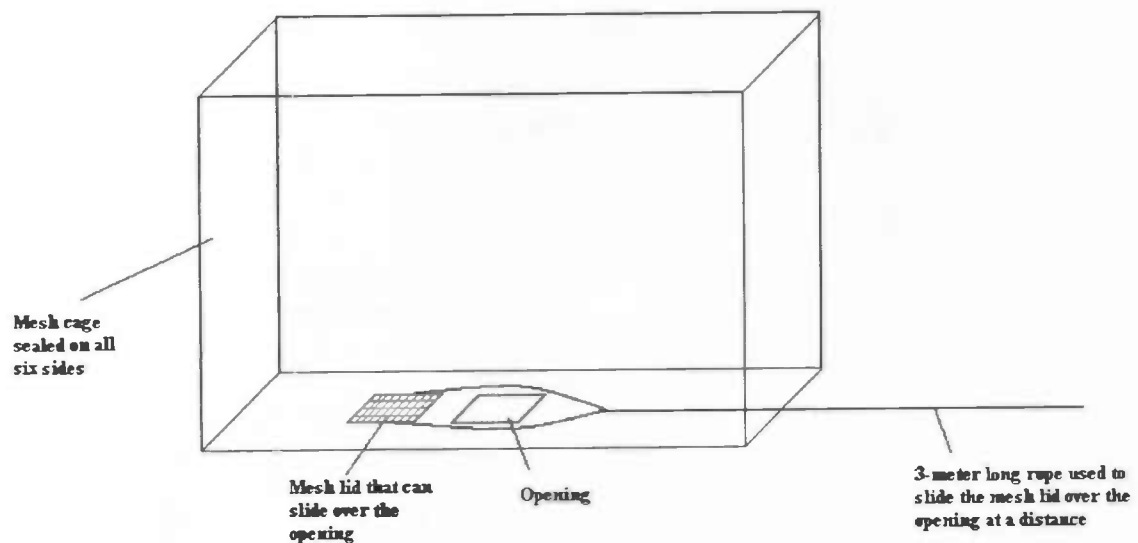slide the mesh lid over the
opening at a distance

Fig. 4 - A simple mesh cage construction used to catch shrimp and gobiid fish partner. When both were inside the cage it could be easily shut at a distance to decrease the potential for disturbance caused by the field researcher.

Fourth, we tried to directly drop the salt down the burrow and position the mesh cage afterwards. Fifth, we tried to add a fake burrow opening in the mesh, as is shown in Figure 5. Unfortunately the burrows were never sturdy enough to hold the fake burrow opening.



Dome
Fake Entrance/Exit
Entrance/Exit
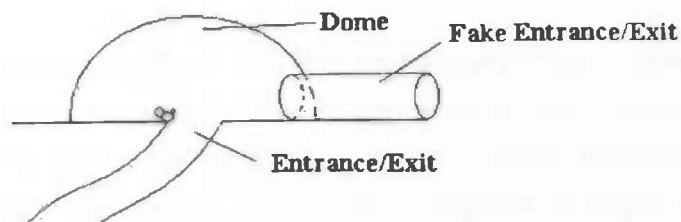
Fig. 5 – A simple dome constructed of sand and mesh glued together was attached to a small PVC tube.

Sixth, we put a simple dome on top of the burrow exit, just to see the reaction of the shrimp and gobiid fish. We made the dome to fit on top of the burrow. With this dome we tried to explore two things. The first was to see if the shrimp would recognize it as part of its burrow and thus walk on through the opening to deposit sand. The second was to attach such a dome to the pump, see

description below in Figure 6, to concentrate the suction of the pump. Seventh, and last, we tried to suck or blow away the shrimp from the burrow by using the simple pump, which was man-powered. Unfortunately we did not have the manpower necessary to create a water flow that was strong enough to blow the shrimp away or to suck it up.
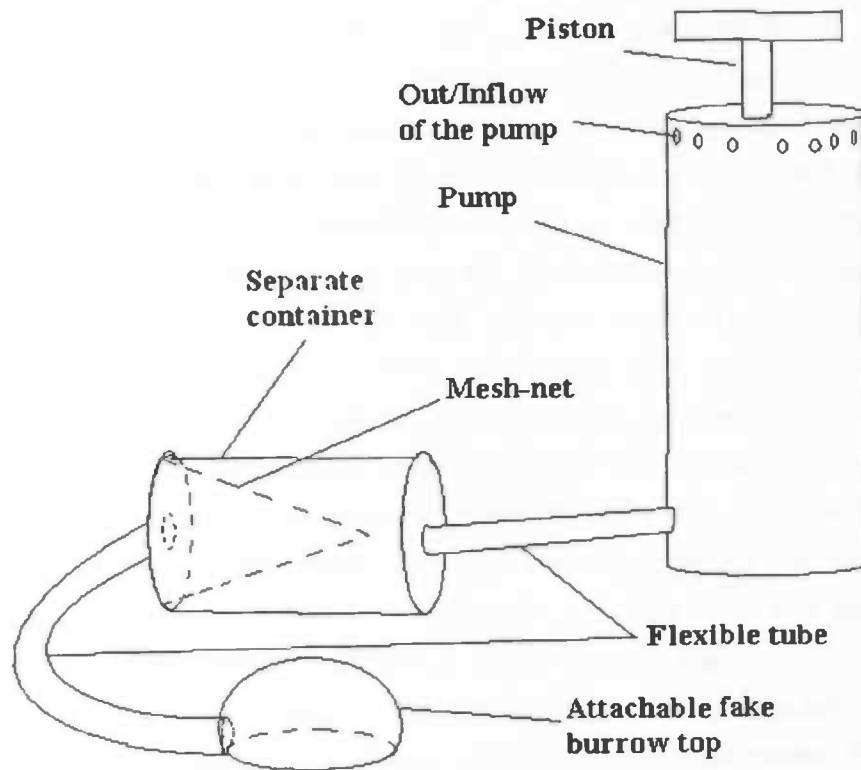


**Fig. 6 -** The pump's schematics. The principal parts of the pump are a large piston in a large PVC tube with holes in the top to allow for efficient water drainage. A small container containing a net that would ensure that any shrimp or fish sucked up would not be caught in the tube leading to the piston. Also a plastic see through top that would allow for light to come through and limit the suction area.

Capturing the *A. acanthus* shrimp was much easier. This shrimp seemed to be attracted to seagrass leaves above its burrow. Simply placing a shovel in the sediment not too far from the burrow and then attracting it with seagrass set the trap. After the shrimp grabbed the seagrass, a signal was given to wedge the shovel, thus closing the burrow behind the shrimp giving it no way out, and therefore easy to catch with a simple fish net.

The burrows of the *A. acanthus* were sturdy and went straight down unlike the alpheid shrimp's burrow, which seemed to be more fragile. Placing a shovel in the near vicinity of the alpheid shrimps burrow usually resulted in the escape of both the alpheid shrimp and gobiid fish after which the burrow collapsed. When

checked again, 15 minutes later, the burrow remained collapsed and no activity was observed.

### 2.4 Seagrass collection

The seagrass, *Thalassia hemprichii*, was sorted accordingly to number of dead shoots and number of living shoots present per 25x25 cm quadrant. There was only one sample taken right before the storm hit the Spermonde Archipelago; all the other samples were taken after the storm. We chose for a practical approach with counting and measuring the surface area (SA) covered by the samples taken after the storm. Ten shoots were picked at random where possible and measured. The rhizomes, shoots, roots and leaves were separated, each into a separate category (see Fig. 7). We measured the length of the whole leaf, of the green part of the leaf, and the width. The number of shoots was counted for density purpose. This represented 20% to 25% of the total sample, which was enough to get a good estimate of the sample total.

The average number of leaves per shoot was determined by dividing the number of leaves by the amount of shoots measured. This gave us the average amount of leaves per shoot. This average multiplied by the number of days to create one leaf (~10 days) (Stapel *et al* 2001) gave us the average age of the seagrass in that area. From this average age and the total Dry Weight (DW) we can calculate DW created per day, which is the leaf production. The loss caused by herbivores was assumed to be constant and indiscriminate. We are interested in finding out how much of this daily leaf production is harvested by the shrimp. The shrimp does not discriminate between fully-grown leaves, old leaves and young leaves.
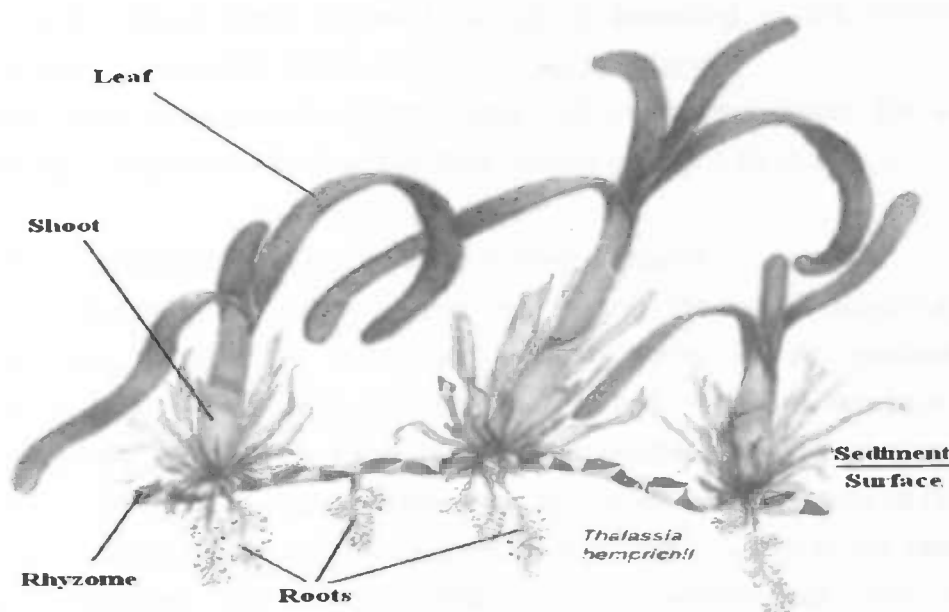
**Fig. 7** - *Thalassia hemprichii* - as taken from Richmond (2002) a simple drawing of Thalassia hemprichli with an indication of the location of "Leaf", "Shoot", "Rhizome", "Roots" and an approximation of what part is below the sediment surface and what part is above the surface.

Loose leaves found on top of the sediment in a 50x50 cm quadrant were identified to what species they belonged and also measured in length and width. Loose leaves were collected after the storm.

## 2.5 Preparation of samples

After collection, the plant material samples were oven-dried at a temperature of 60 degrees Celsius for at least 24 hours. We then proceeded with the analysis of our field samples with using a Carbon-Nitrogen-Sulfur (CNS) at the Radboud University (RU) in Nijmegen, the Netherlands. The samples were ground to a fine chalk like powder. We transferred a bit of our sample material into a tin capsule. After filling the tin capsule we proceeded to weigh, fold and compress it accordingly to make a compact ball of it. The tin ball sample was placed in an autosampler.

The autosampler worked as follows. The sample in the sampler drops into an incineration tube, where, under the influence of oxygen and the tin of the container, it is instantly incinerated at a temperature of 1800 degrees Celsius. The resultant gasses: $CO_2$, $N_2$, $N_xO_y$, $H_2O$ and excess $O_2$ flow through a Cu-column, where nitrogen oxides are reduced to elementary nitrogen and $O_2$ to CuO. These then pass through another column where water is absorbed. The gasses are led to a GC-oven, where the Nitrogen, Carbon and, if needed, Sulfur are separated in a Poropak QS-column, and flow through a Thermal Conductivity

Detector, which sends an electrical signal according to the concentration of Nitrogen, Carbon and Sulfur. (General Instrumentation:
http://www.instr.sci.kun.nl/startnl.html) (All information about the workings of the autosampler can be found on this website provided by the RU.)

## 2.6    Measurement of water nutrient concentrations

Pathways of nutrients and the cycling thereof in seagrass beds are important in order to understand what is going on. As mentioned in the introduction, our research area - a seagrass bed - is a sink/source in a location where water nutrients are low. How this is possible is exactly what research and theory have tried to explain in the past, but rather unsuccessfully so far. In order to understand this part of the nutrient cycling as it occurred in our research area, we measured the water nutrient concentrations with the use of a spectrophotometer, using a wavelength of 620 nm for the $NH_4$ and 880 nm for $PO_4$. We used the "Ammonium" (method) and the "Reactive Phosphate" methods as described in Short and Coles 2003, with a little variation of the wavelength. Samples were taken from three similar types of location at a depth of approximately 10 cm. This was done using a syringe equipped with a flexible rubber tube that was attached to a 10 cm hard tube filter. The filter was put in the sediment and the syringe was sucked vacuum by using a wedge between the piston and the syringe casing. Each series of samples was compared to a standardized set of concentrations (0, 0.5, 1, 2, 3, 5, 10 and 20 µM). The standardized set was then used to determine the concentrations of the water samples. The line of best fit and the accuracy of the standardized set, denoted by "y"and "$R^2$" respectively, are also present in the Figures 15 and 16.

## 3.    Research results

In this section we will present the results of the research activities as they were formulated before. The concrete research activities, were observing behavior, measure densities, catching the shrimp, seagrass collection, sample preparation and the measuring nutrients concentration in the seawater as listed in section 2.

### 3.1    *Behavioral observations of both alpheid shrimp and gobiid fish*

We looked at tidal height and time of day. Because so little is known about these shrimp and their partner fish, we labeled all visible behaviors in categories (table 1). Using these categories as well as tidal height and time of day we assembled the following figures (8-10). These figures give us a basic understanding of how tidal height and time of day may influence the behavior of the alpheid shrimp.

The shrimp is capable of moving its burrow opening around in the seagrass bed. We had limited time in the field and were unable to mark the shrimp for tracking/identifying in the field. We therefore chose to observe shrimps, at random, that were active around their burrow. Our first observation was done on the 23rd of November 2003 and we tested two ways of observing. One consisted of using a minute interval and marking occurrences the other was continuous monitoring marking times. All monitoring, thereafter, were done by marking occurrences per minute. Disturbance was added to the list of observations categories after quick consult in the field later that day. This resulted in half of the observations, done on that day, missing the "disturbance" category.

## Activity of both alpheid shrimp and gobiid fish
## per tide level and time of day



**Fig. 8 a1-c2** - Amount of activity for each type of behavior per burrow in the time recorded (15 minutes) for n=75 of both shrimp and fish. "1" – denotes tide comparison (dm), "2" – denotes time comparison (hour of the day). Both are comparisons to activity.

# Activity of both alpheid shrimp and gobiid fish
# per tide level and time of day (Conti.)



**Fig. 8 d1-f2** (conti.)– Amount of activity for each type of behavior per burrow in the time recorded (15 minutes) for n=75 of both shrimp and fish. "1" - denotes tide comparison (dm), "2" - denotes time comparison (hour of the day). Both are comparisons to activity.

## Activity of both alpheid shrimp and gobiid fish
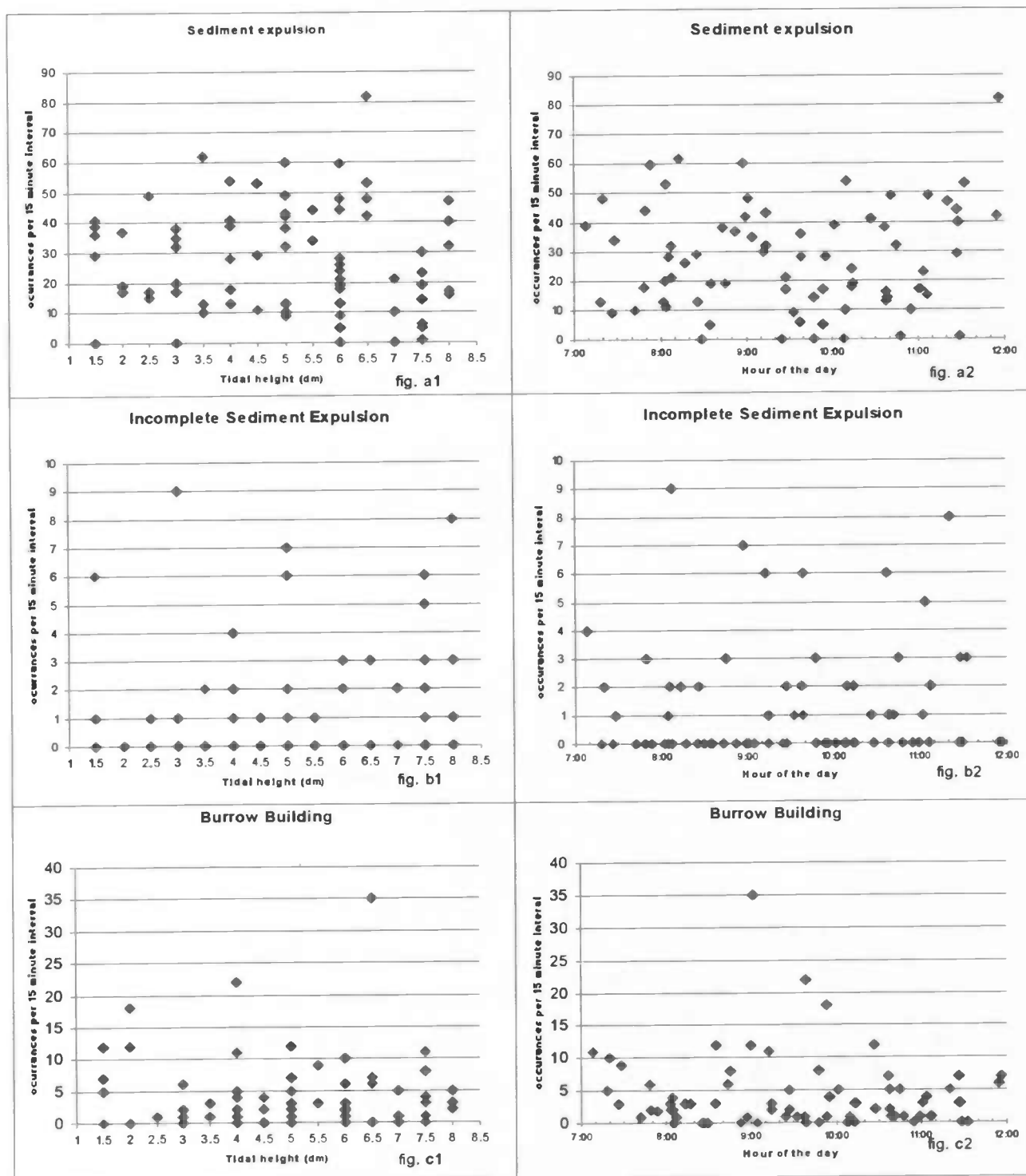## per tide level and time of day (Conti.)



**Fig. 8 g1-i2** (conti.)– Amount of activity for each type of behavior per burrow in the time recorded (15 minutes) for n=75 of both shrimp and fish. "1" - denotes tide comparison (dm), "2" - denotes time comparison (hour of the day). Both are comparisons to activity.

## Activity of both alpheid shrimp and gobiid fish
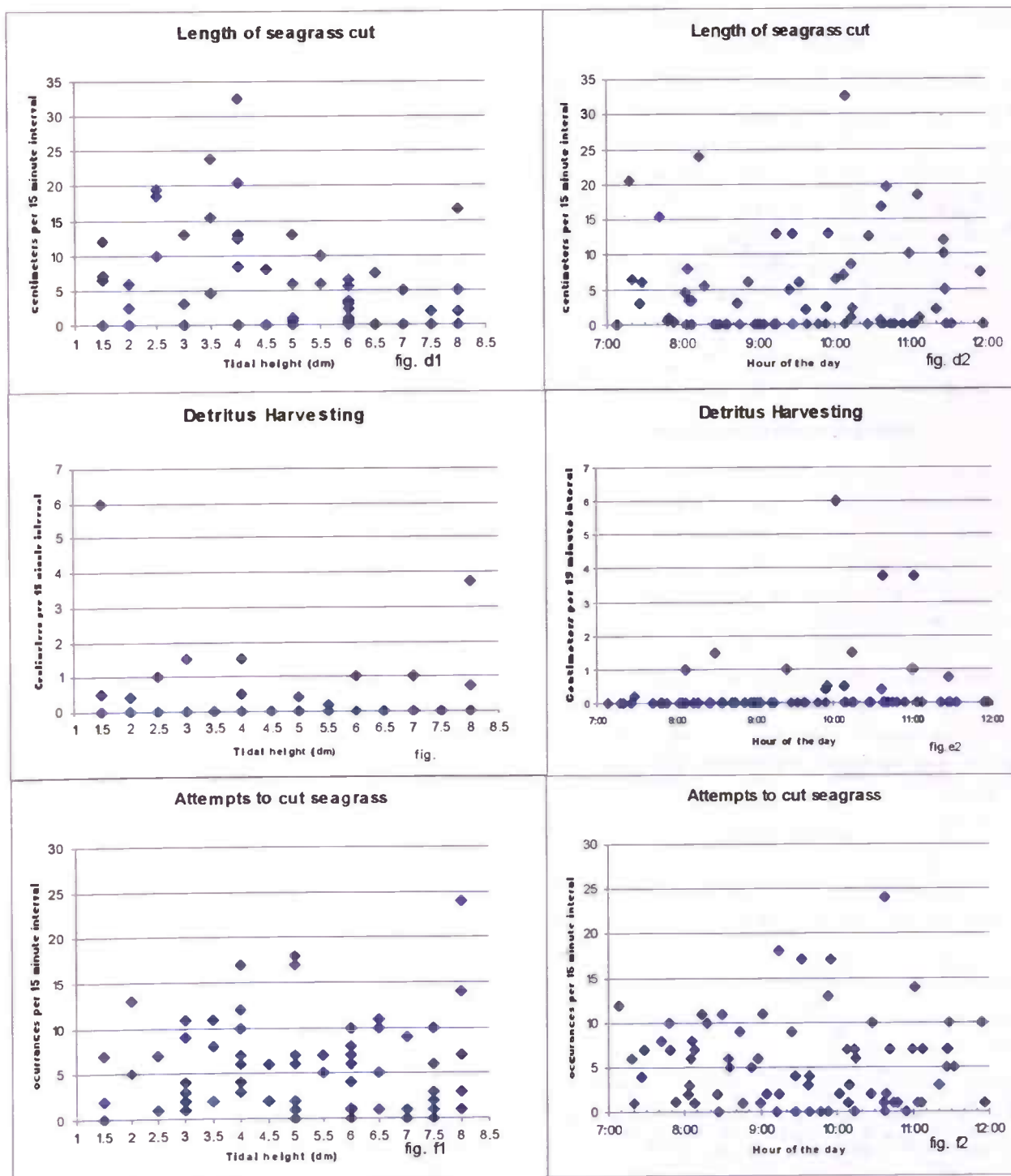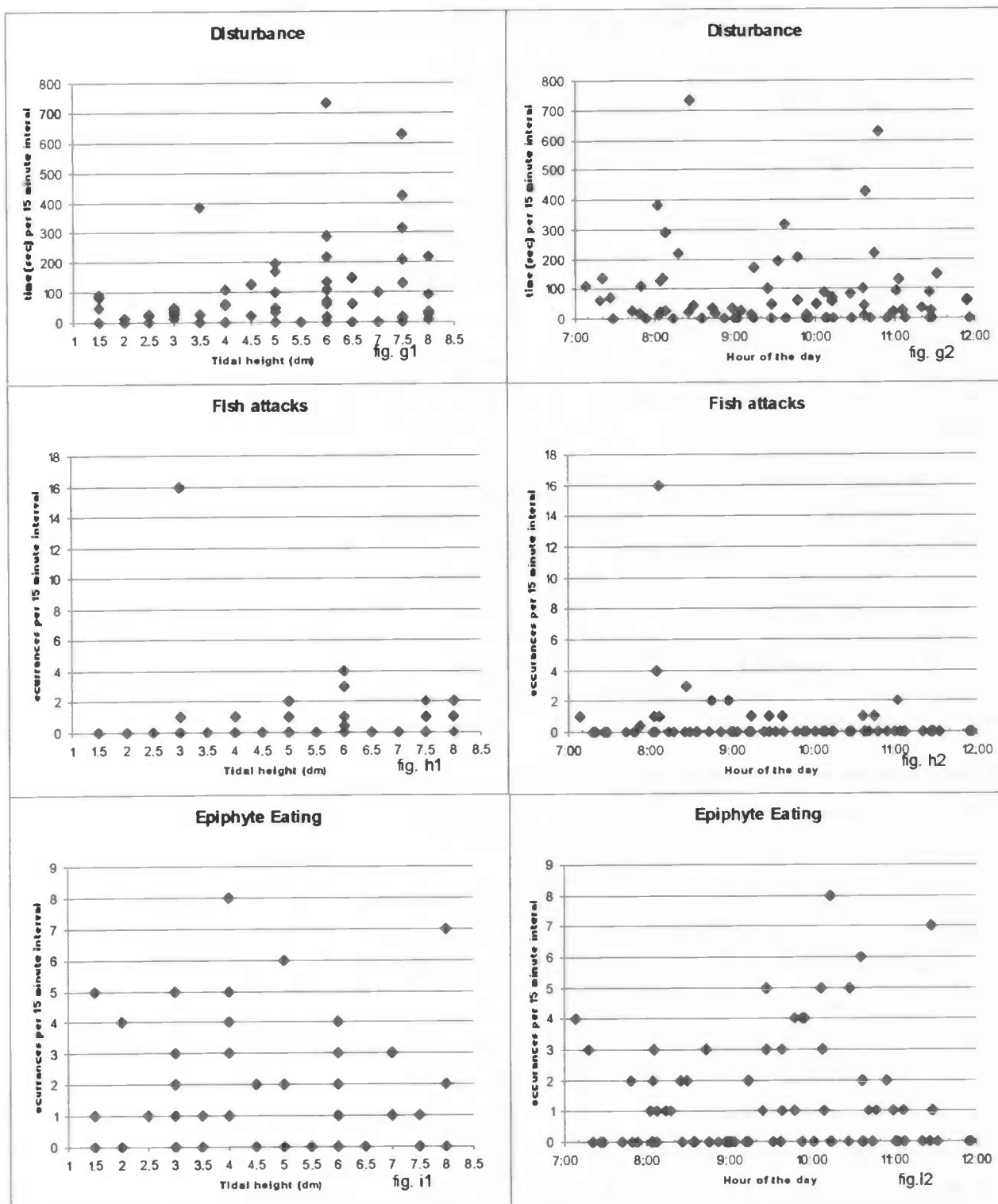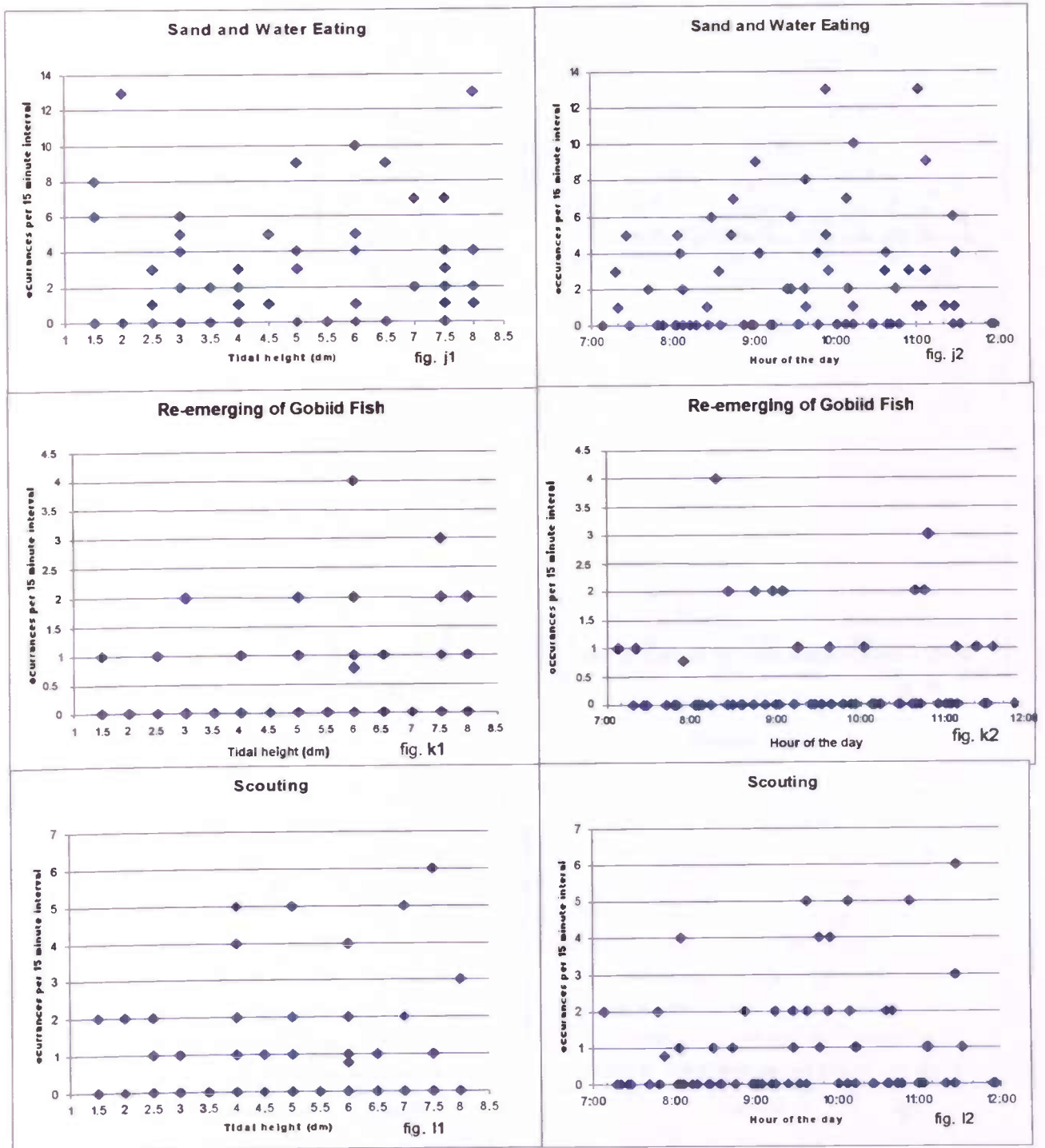## per tide level and time of day (Conti.)



**Fig. 8 j1-l2** (conti.)– Amount of activity for each type of behavior per burrow in the time recorded (15 minutes) for n=75 of both shrimp and fish. "1" - denotes tide comparison (dm), "2" - denotes time comparison (hour of the day). Both are comparisons to activity.
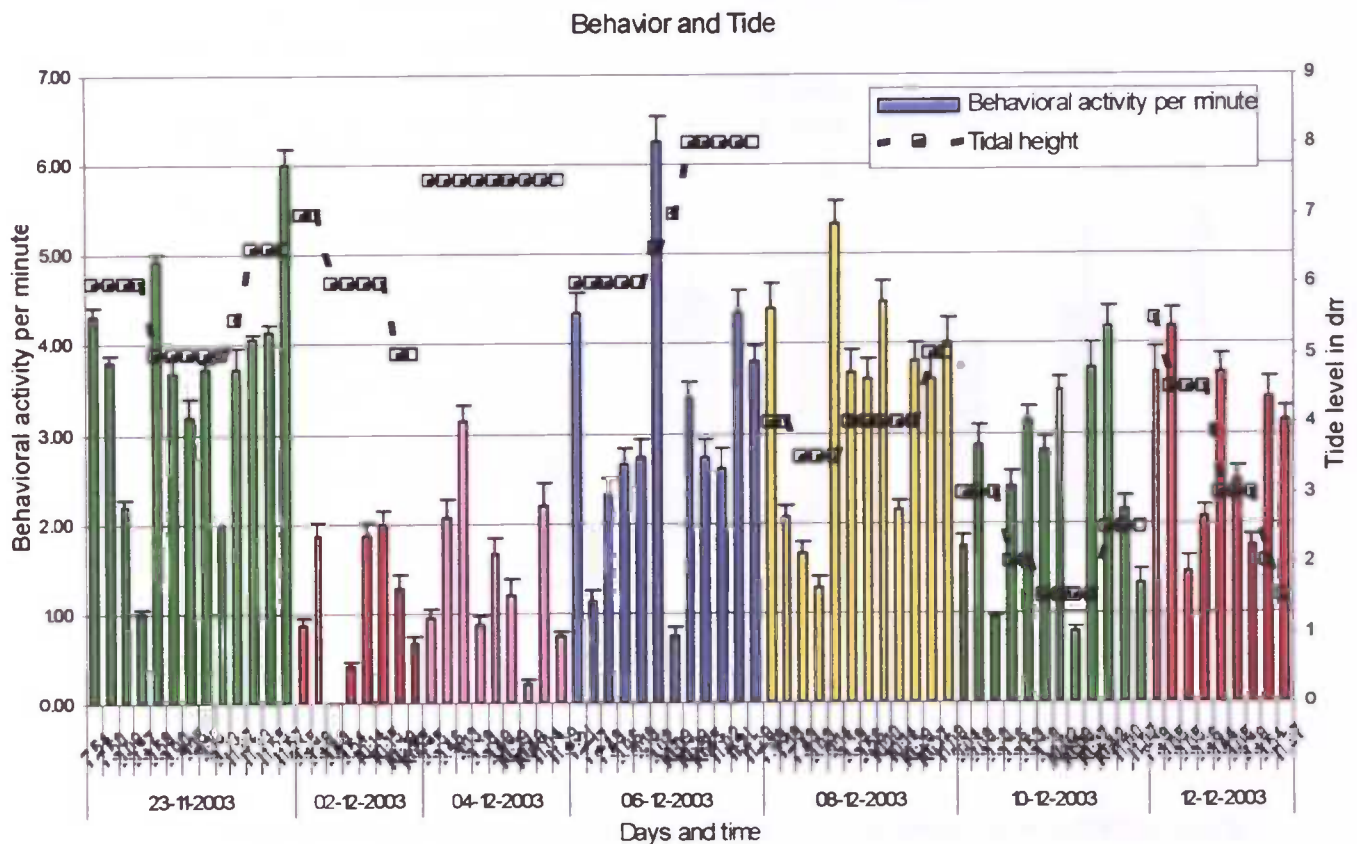
**Fig. 9** – Frequency of behavioral activity scored (burrow building + sediment expulsion + incomplete sediment expulsion + loose leaves harvest + seagrass harvest + attempting to cut seagrass) per minute on the time recorded in chronological order. Each day is given a different color bar. Tide level (dm) at the time of the observations of the activity is included in this graph. The figure only shows the frequency of behavioral activity scored for the alpheid shrimp. (n=1 per bar)

Using the categories, from table 1, we produced figures 8 a1-l2. We chose to pool some of them to perhaps gain a better view of frequency of "behavioral activity" scored per shrimp (see Fig. 9). We pooled "Sediment expulsion", "Incomplete sediment expulsion", "Burrow building" and "Attempting to cut seagrass" as defined in Table 1. We added the successful harvesting of seagrass and loose leaves as occurrences, not by size, and called it "behavioral activity". All these categories are activities that frequently occur outside the burrow.

Average activity per burrow under different tide conditions
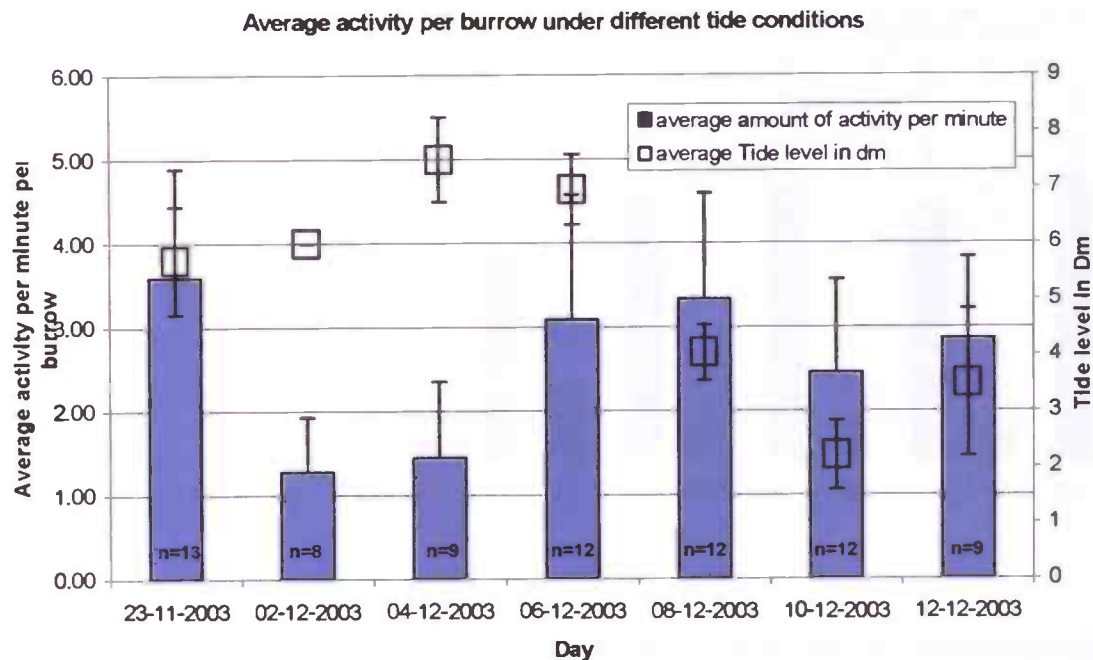
**Fig. 10** – Frequency of activity of the alpheid shrimp as measured per burrow per day per minute. The number of observed burrows is defined per bar (n). Average tidal fluctuation was taken from the tide predictions, Fig. 2, during our observation time in the seagrass bed.

Figure 9 shows us general activity per day. This behavioral activity, using table 1, can be subdivided into sediment reworking and harvesting. In figure 11 we plotted the sediment reworking using the observed sediment expulsion, incomplete sediment expulsion, and burrow building. This is the main part of behavioral activity. Figure 12 shows us the percentage of the behavioral activity that is spend on harvesting. Harvesting consists of "attempting to cut seagrass", "seagrass cutting" and "Loose leaves harvesting".

An important note to add to clarify harvesting is that leaving a burrow is always associated with attempting to cut seagrass, but sometimes this is interrupted by the find of *loose leaves*. Therefore we added this loose leaves gathering as a rate of success in the harvesting. Though unlike figure 10, figures 11 and 12 show us the percentage of occurrences per day. These percentages give an indication of changes in ratios per day.
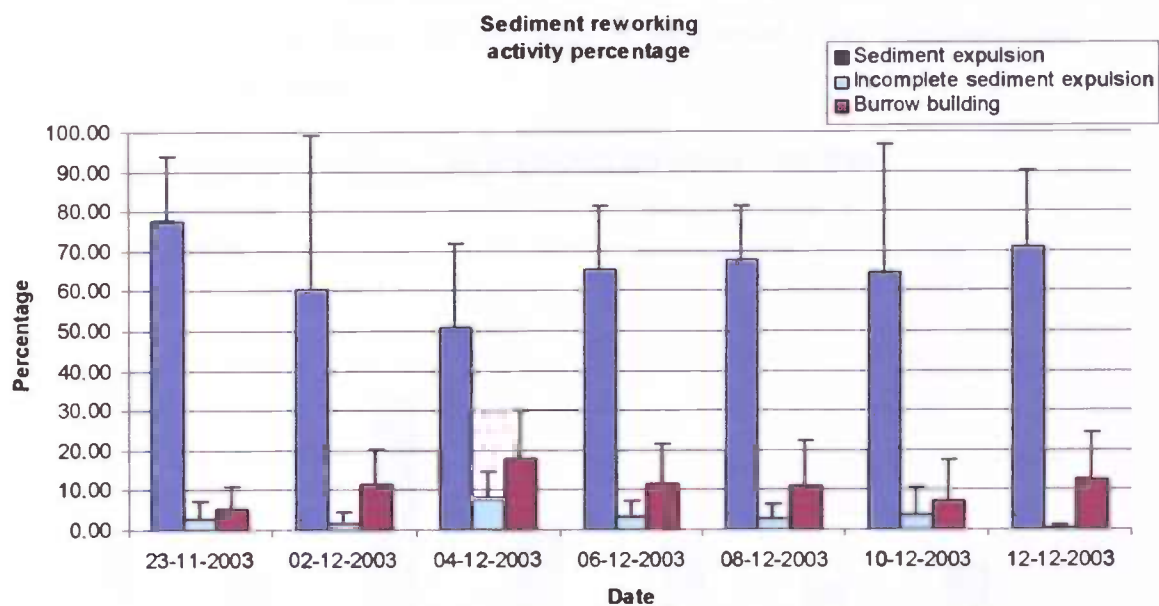
**Fig. 11** – Relative burrow activity of the alpheid shrimp divided into sediment reworking (Sediment expulsion and Incomplete sediment expulsion), time spend on construction (Burrow building).
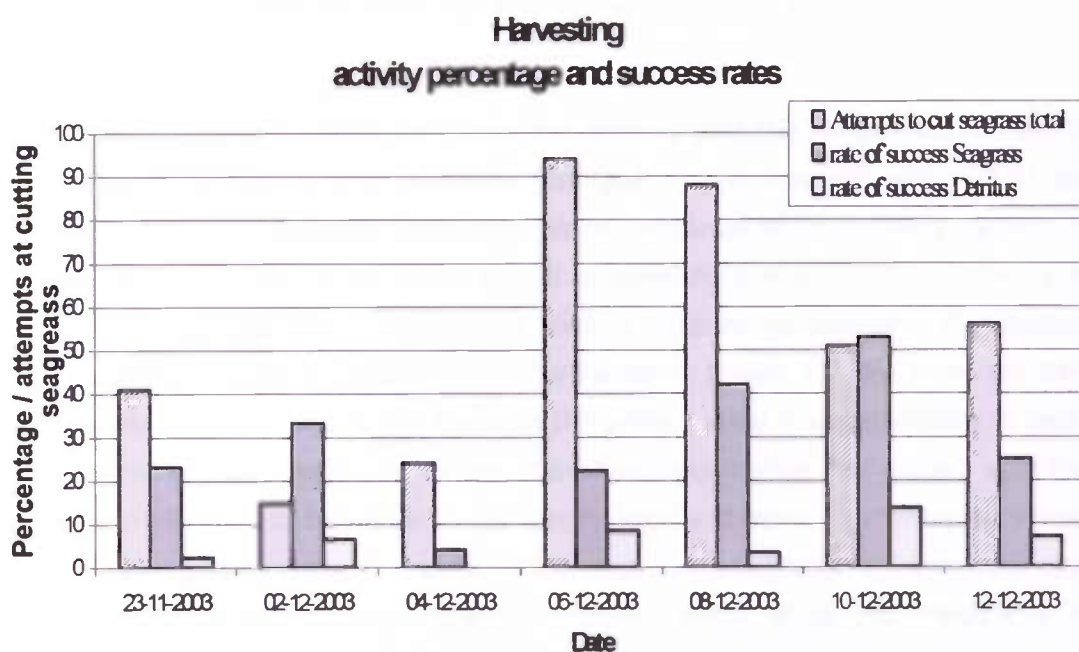


**Fig. 12** – The harvesting activity of the alpheid shrimp. "Attempts to cut seagrass" here displayed as total attempt observed on that day. The rate of success at cutting seagrass and the rate of success of retrieving loose leaves is a percentage of the attempts to cut seagrass. The rate of success is the chance that the shrimps attempt to cut seagrass successful and harvests the seagrass or loose leaf.

Figure 13 portrays the disturbance per day as a percentage per 15 minutes observation. This gives an indication of why some days displayed less behavioral activity than others.

**Percentage of time that the alpeid shrimp is disturbed**
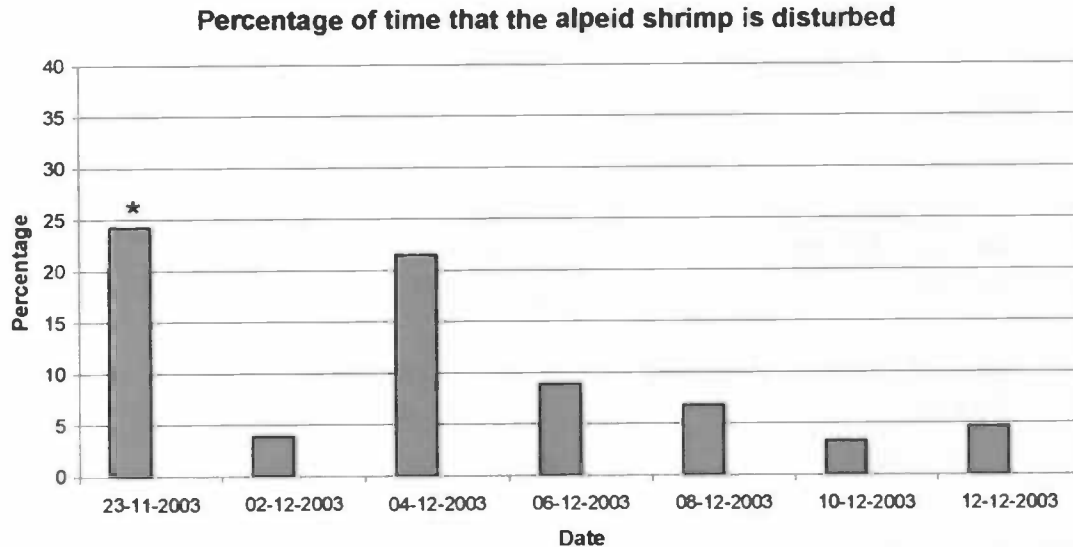


**Fig. 13** – Relative amount of time the alpheid shrimp is disturbed by outside factors. The time disturbed is spend in side the burrow and keeps the shrimp from undertaking any other action for that time period. The (*) indicates that only half the observations of that date could be used in this case due to lack of information in the other set of data.

## 3.2 Behavioral observations overview

The figures 8-13 thus far show the activity and the influences of certain environmental factors on this behavior. The goal of this research was to find out about harvesting of seagrass and its possible importance to nutrient cycling in seagrass beds. The above figures 8-13 tell us something about the consistency of behavior through the days. The general idea of happenings and time distribution among different types of behavior can be found in figure 14. We have omitted standard deviations in figure 14, because the added value is questionable at best. Some behavior can take place or not. Constant monitoring (24 hours) and the presence of all types of behavior would justify standard deviation, however during the 15-minute observations it was too often that some behavior did not occur and thus the standard deviation range was between 0 occurrences and which ever is maximal. Figure 8 shows this range and variation. Figure 14 is therefore an average of all the 15-minute observations to give a general impression.

## Shrimp and gobiid fish behavior overview
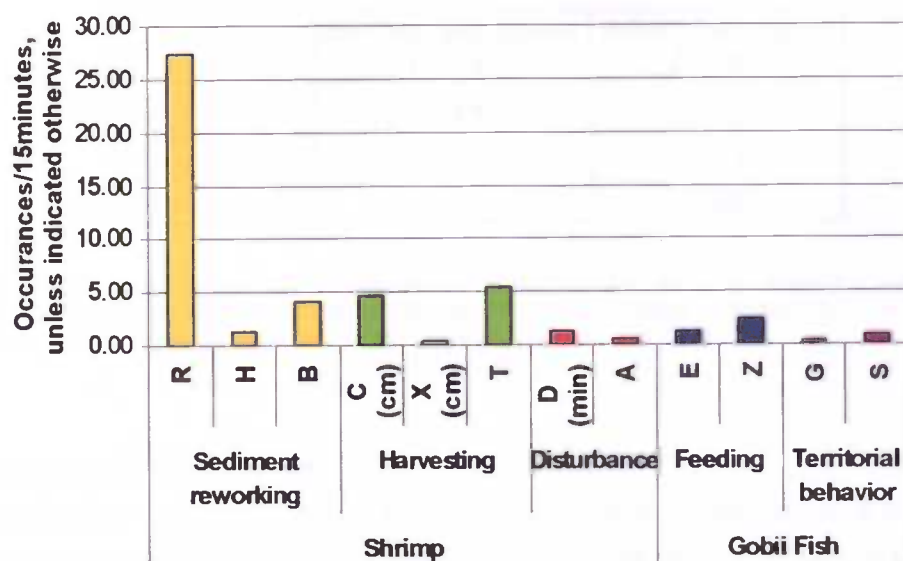


**Figure 14** - Behavior categorized for the alpheid shrimp according to Sediment reworking, Harvesting, Disturbance. Behavior categorized for the gobiid fish according to Feeding and Territorial. Total of 1125 minutes of observation at burrows (n = 75). All results are occurrences per 15 minutes unless indicated otherwise. The letters used to indicate certain behavior correspond to those described in table 1 in Materials and Methods.

### 3.3    Shrimp density measurements

Table 2 is an overview of the shrimp and gobiid densities measured in several places in the seagrass beds around Bone Batang. Table 3 uses the same information given in table 2, but shows the measurements that were taken before and after the storm.

### Shrimp and gobiid densities

| Density/m² | Transect (n=7) | | | | | | Average | S. D. |
|---|---|---|---|---|---|---|---|---|
| | 1&2 | 3 | 4 | 5 | 6 | 7 | | |
| **Alpheus shrimp** | 0.55 | 1.1 | 0.40 | 0.10 | 0.20 | 0.20 | 0.44 | 0.34 |
| **Gobiid fish** | 0.65 | 1.2 | 0.40 | 0.10 | 0.40 | 0.40 | 0.54 | 0.34 |
| **Axius Sp.** | 0.15 | 0.60 | 0.30 | 0.50 | 0 | 0.20 | 0.27 | 0.21 |

**Table 2-** Densities per m² of the alpheid shrimp, gobiid fish, and species of *Axius acanthus* shrimp, also found in the area. The *Axius* sp. is included because it harvests anything that comes above its burrow and seems to be as abundant as the alpheid shrimp. Scoring was done along n=7 1 by 10 meter transect.  The first two observations were pooled in situ and later observations were kept separate.

**Density shift**

| Density/m² | Before the storm (n=3) | After the storm (n=4) |
|---|---|---|
| **Alpheus Shrimp** | 0.73 | 0.23 |
| **Gobiid Fish** | 0.83 | 0.33 |
| **Axius Sp.** | 0.30 | 0.25 |

**Table 3-** Densities per m² for alpheid shrimp, goblid fish and *Axius* sp. making a distinction between densities measured before the storm hit the island, transect 1-3 n=3, and measurements taken after the storm hit, transect 4-7 n=4, see table 2.

### 3.4   Seagrass daily production

Leaves were collected, in the field, separated and measured, in a lab, for later reference of the harvesting behavior of the shrimp. Shoot surface area and weights are given below in table 4. With the exception of one sample, due to damaging in the transport, ten shoots were picked at random out of the sample and measured. These were kept and dried separate of the entire sample and used for reference of surface to dry weight measurements.

**Shoot surface area and weight**

| Sample # | Shoots counted # | Total leaf mm² | Green part leaf mm² | Green part leaf % | DW of counted shoots g | Weight of Leaf surface g/m² leaf |
|---|---|---|---|---|---|---|
| 2 | 10 | 55804 | 24993 | 45 | 1.663 | 29.80 |
| 4 | 10 | 34975 | 16120 | 46 | 1.150 | 32.88 |
| 5 | 10 | 34688 | 13162 | 38 | 0.993 | 28.63 |
| 6 | 8 | 22810 | 9707 | 43 | 0.676 | 29.64 |
| **Average** | | | | 42.84 | | 30.24 |
| **SD** | | n = 4, 38 | | 3.58 | | 1.84 |

**Table 4 -** Shoot surface area and weight. The 10, where possible, separate shoots were taken per sample at random and were measured in width and length of both the total leaf as well as the green, photosynthetic, part of the leaf. The samples were dried separately from the other leaves to establish Dry Weights (DW). The information on leaf length above the surface is unknown in the sample before the storm (sample 3).

Table 5, below, give the widths of all the leaves counted. The average width of the leaves counted was used to calculate the surface area of leaves cut by the shrimp. The width of the leaves harvested by the shrimp was not established in situ.

**Leaf width**

| | Sample 3 | Sample 2 | Sample 4 | Sample 5 | Sample 6 | Total |
|---|---|---|---|---|---|---|
| Sum of width (mm) of all leaves | 1231.5 | 484.5 | 323.5 | 313 | 214 | 2566.5 |
| # Leaves | 175 | 56.0 | 43.0 | 44 | 33 | 351 |
| Average width (mm) | | | n = 351 | | | 7.3 |

Table 5 - Leaf width. Sample width was pooled and divided by the amount of leaves counted to estimate the average width of seagrass leaves.

The dry weight of the entire sample, excluding the 10 shoots removed from the sample for more detailed measurements, was weighted. The dry weight of the entire sample is composed of the DW of the samples in table 4 and the rest of the sample. Table 6 is the DW per sample per $m^2$ and an average is given below.

**_Thalassia_ biomass**

| Sample # | DW leaf sample (25x25 cm) g | DW leaf g/$m^2$ |
|---|---|---|
| 3 | 1.72 | 27.52 |
| 2 | 4.81 | 76.96 |
| 4 | 5.04 | 80.64 |
| 5 | 4.14 | 66.24 |
| 6 | 2.37 | 37.92 |
| Average | | 57.86 |
| SD | n = 5 | 23.83 |

Table 6 - *Thalassia* biomass. Dry weight of entire sample calculated per $m^2$.

The production of this DW, see table 6, was established using Stapel *et al* (2000) leaf production time. It takes approximately 10 days to produce a fully-grown leaf. Therefore we can assume that the amount of leaves in a shoot determines the amount of days it took to produce the plant in its current state. This estimate of the plant's age, together with the DW of the seagrass leaf per $m^2$, calculated in table 6, makes it possible to determine the amount of DW produced per day, see table 7.

**Thalassia production**

| Sample # | Shoots counted # | Leaf counted # | Average leaf/shoot # | Days to produce all shoots Day | Production leaf g DW leaf/ m²/ day |
|---|---|---|---|---|---|
| 3 | 41 | 175 | 4.27 | 42.7 | 0.64 |
| 2 | 10 | 56 | 5.60 | 56.0 | 1.37 |
| 4 | 10 | 43 | 4.30 | 43.0 | 1.88 |
| 5 | 10 | 44 | 4.40 | 44.0 | 1.51 |
| 6 | 8 | 33 | 4.13 | 41.3 | 0.92 |
| Average | | n = 5, 79 | | 45.4 | 1.26 |
| SD | | | | 6.0 | 0.49 |

**Table 7** - *Thalassia* production. The production of a shoot was calculated in days using an estimate of 10 days of production per leaf given by Stapel *et al* 2001.

### 3.5    Seagrass harvest

Figure 14 presents the average length harvested by the shrimp. Table 8 calculates the length of seagrass cut per day based on the average lengths cut per 15 minutes during the 15-minute observations.

**Shrimp leaf length harvest**

| | mm leaf/ shrimp/ 15 min | mm leaf/ shrimp/ hour | mm leaf/ shrimp/ day |
|---|---|---|---|
| Average | 46.2 | 185 | 2219 |
| SD | 67.4 | 270 | 3235 |

**Table 8** - Shrimp leaf length harvest. Using the information from of table 2, this table shows the daily harvesting of the shrimp expressed in length of seagrass.

Combining the tables 4, 5 and 8 results in table 9. Table 9 estimates the amount of grams of DW harvested by the shrimp in the field. This amount can be compared to the average production of seagrass leaves, see table 7.

**Shrimp DW leaf harvest**

| | mm leaf/ shrimp/ day | m² leaf/ shrimp/ day | g DW leaf/ shrimp/ day | g DW leaf/ m²/ day |
|---|---|---|---|---|
| Average | 2219 | $1.62 \times 10^{-02}$ | $4.91 \times 10^{-01}$ | 0.22 |

**Table 9** - Shrimp DW leaf harvest. Using the above information on width (table 5) and DW per m² surface area of the leaves, an estimate of how much DW the shrimp harvests per day is made.

Using all the information above we can now calculate how much of the daily production of seagrass is harvested by the shrimp (Table 10). The harvest of the above ground, the green part of the leaf, and the total leaf production are displayed in table 10. The harvest of the above ground leaf is a measure of the

amount of leaf that is actually available to the shrimp, because the rest of the leaf is below the surface of the sediment.

### Shrimp harvest leaf production

| Harvest by shrimp | Leaf production | Harvest shrimp of produced leaf | Harvest shrimp of produced above ground leaf |
|---|---|---|---|
| g DW leaf/ m²/ day | g DW leaf/ m²/ day | % | % |
| 0.22 | 1.26 | 17.2 | 40.1 |

**Table 10-** Shrimp harvest leaf production. Using the information from tables 2-8 the average harvest of produced leaf and the average harvest of produced leaf material above ground is calculated, because the production under the sediment was unavailable for the shrimp to harvest. Table 4 shows that only ~42% of the leaf is produced above ground and thus available for harvesting.

The shrimp also harvests loose leaves, but, as seen in figure 14, in lesser quantities than fresh leaf material. Table 11 gives an overview of the amounts and types of loose leaves found in the seagrass beds where the shrimps were observed.

### Loose leaves composition

| | Total length (mm) | Average width (mm) | Surface area (m²)/ (m²) | Percentage % | DW (g/m²) |
|---|---|---|---|---|---|
| **Thalassia** | 18688 | 8 | 0.1225 | 96.21 | 2.2500 |
| **Halodule** | 205 | 4 | 0.0007 | 0.55 | 0.0136 |
| **Halophila** | 350 | 10 | 0.0030 | 2.32 | 0.0336 |
| **Cymodocea** | 332 | 4 | 0.0012 | 0.93 | 0.0210 |
| **Rhyzomes** | 103 | 0 | N.A. | - | 0.0633 |

**Table 11 -** Unattached loose leaves collected within a 50x50 cm quadrant in a mixed seagrass bed. This table shows the composition in a primarily *Thalassia hemprichii* bed. 2.25g DW per m² of all loose leaves are *T. hemprichii*; only ~0.13 g DW/m² are loose leaves of other species.

Considering that most of the loose leaves consists of *Thalassia* material we assume that the material harvested by the shrimp also mainly consists out of *Thalassia* loose leaves. The same calculation was done as those shown in the tables 4-10, but this time for "loose leaves".

**Leaf surface area and weight (loose leaves)**

| Sample # | Leaves counted # | Total leaf SA mm² | DW of counted leaves g | Leaf weight per m² SA g/m² leaf | DW leaf g/m2 |
|---|---|---|---|---|---|
| 1 & 2 | 46 | 18410.5 | 0.61 | 33.19 | 1.22 |
| 3 & 4 | 45 | 21311 | 0.66 | 31.02 | 1.32 |
| 5 & 6 | 94 | 40338.5 | 1.88 | 46.49 | 3.75 |
| 7 & 8 | 93 | 33039 | 1.11 | 33.61 | 2.22 |
| 9 & 10 | 107 | 40062 | 1.37 | 34.13 | 2.73 |
| Average | | | | 35.68677 | 2.25 |
| S.D. | | | | 6.155189 | 1.05 |

**Table 12** – Leaf surface area and weight (loose leaves). All leaves were taken from a 50x50 cm quadrant and two quadrants were pooled to establish DW.

**Shrimp loose leaves length harvest**

| | mm leaf/ shrimp/ 15 min | mm leaf/ shrimp/ hour | mm leaf/ shrimp/ day |
|---|---|---|---|
| Average | 5.0 | 20 | 239 |
| SD | 19.5 | 78 | 938 |

**Table 13** – Shrimp loose leaves length harvest. Calculated the amount of length of loose leaves harvested by the shrimp per hour and per day.

**Shrimp DW loose leaves harvest**

| | mm leaf/ shrimp/ day | m² leaf/ shrimp/ day | g DW leaf/ shrimp/ day | g DW leaf/ m²/ day |
|---|---|---|---|---|
| Average | 239 | $1.91 \times 10^{-03}$ | $6.83 \times 10^{-02}$ | 0.03 |

**Table 14** – Shrimp DW loose leaves harvest. The average amount of loose leaves harvested by shrimp per day.

**Shrimp harvest loose leaves**

| Harvest by shrimp | Loose leaves available | Harvest of loose leaves |
|---|---|---|
| g DW leaf/ m2/ day | g DW leaf/ m2/ day | % |
| 0.03 | 2.25 | 1.3 |

**Table 15** – Shrimp harvest loose leaves. Percentage of loose leaf DW harvested by shrimp per m².

## 3.6   Nutrient concentrations

The nutrient concentrations of the sampled water, which were taken at several places in the field, were calculated using the ammonium and reactive phosphate protocols mentioned in Methods. Unfortunately the correct wavelength

filter was unavailable, reactive phosphate: 880nm filter available, exact peak at 885nm, and ammonium: 620nm filter available, exact peak at 640nm. A wavelength filter close to the correct wavelength was used to establish these concentrations as seen in figure 15 and 16.
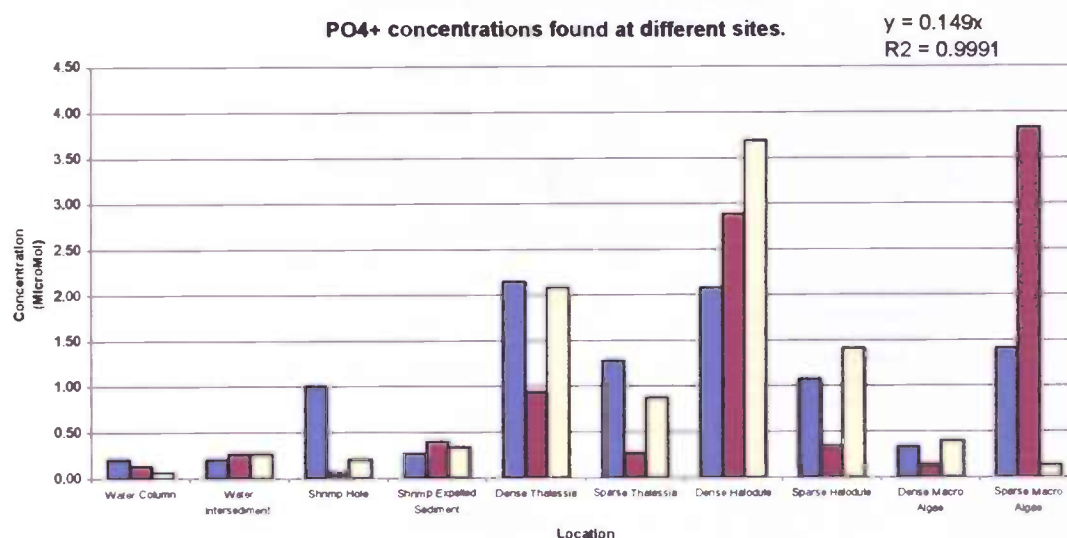


**Fig. 15** – $PO_4^+$ concentrations in µmol/l in different parts of a mixed seagrass bed. All measurements were taken at 10 cm depth in the sediment, excluding the Shrimp Hole, Shrimp Expelled Sediment and Water Column samples, and three replicas were taken per site. Concentrations were calculated using a standardized set of concentrations which had a line of best fit of y = 0.149x and an accuracy of $R^2$ = 0.9991.



**Fig. 16** – $NH_4^+$ concentrations in µmol/l in different parts of a mixed seagrass bed. All measurements were taken at 10 cm depth in the sediment, excluding the Shrimp Hole, Shrimp Expelled Sediment and Water Column samples, and three replicas were taken per site. Concentrations were calculated using a standardized set of concentrations which had a line of best fit of y = 0.0101x and an accuracy of $R^2$ = 0.9924.
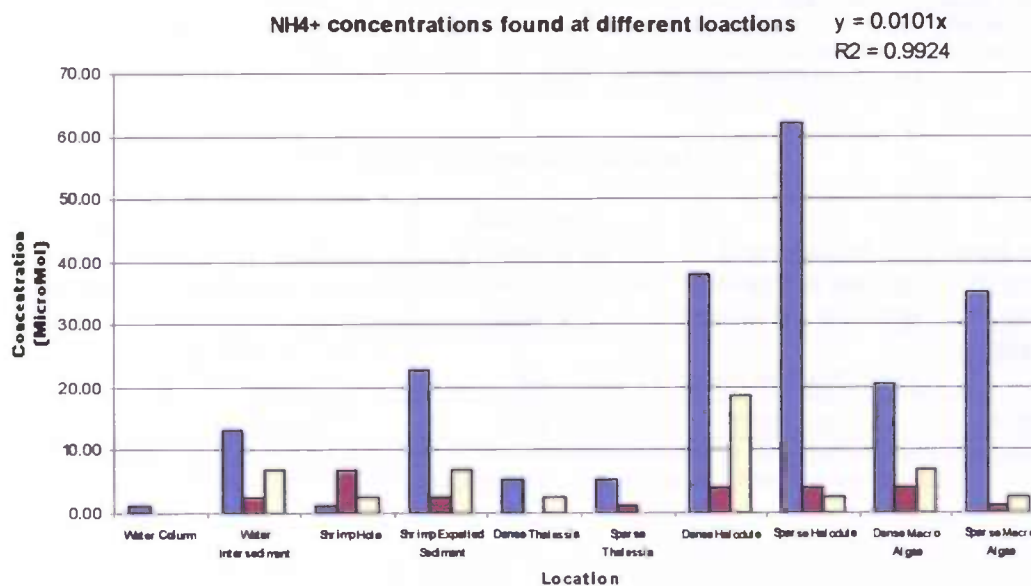
In Nijmegen, at the RU, the Nitrogen and Carbon content were determined (see Fig. 17). These can be used to establish how much was harvested by the shrimp, see table 9, 14, and 15.
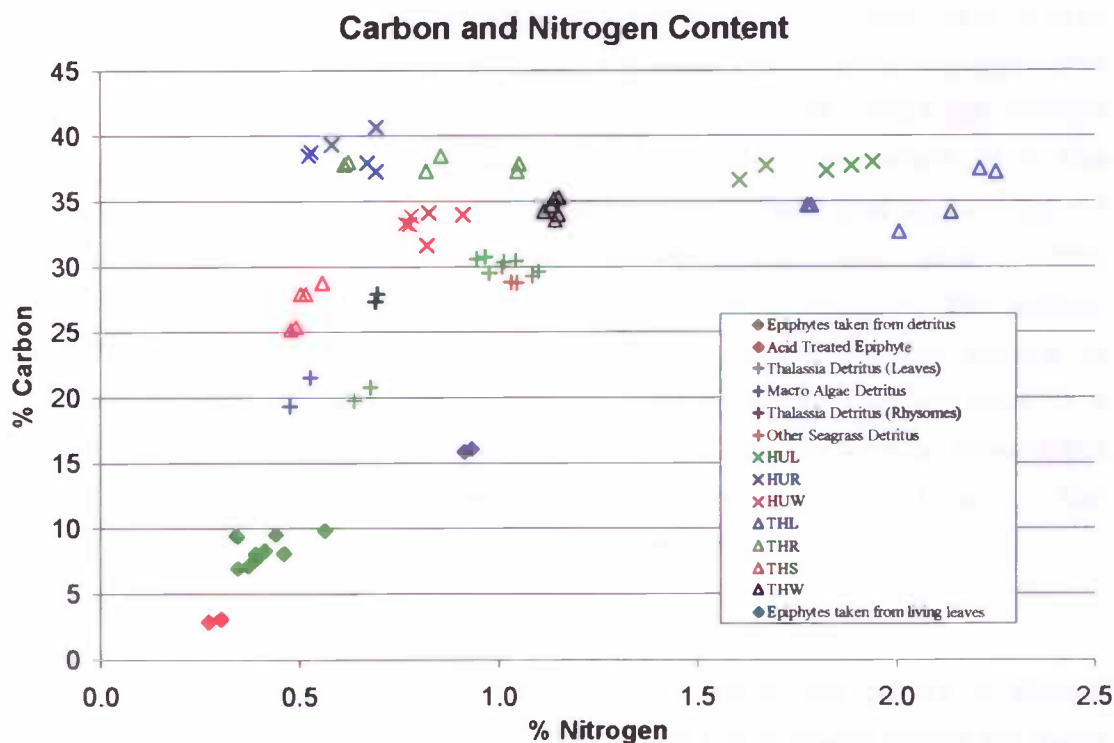
## Carbon and Nitrogen Content



**Fig. 17** – Carbon content compared to Nitrogen content for a variety of samples. Analysis was done at the Radboud University (RU) using CNS (see above). The first 2 letters of the coding refer to the seagrass species: HU= *Halodule uninervis* and TH= *Thalassia hemprichii*. The third letter refers to a part of the plant: L=leaf, S=Sheath, R=Rhizome, W=Roots unless stated otherwise. For example: THL= *Thalassia hemprichii* Leaf.

## Nitrogen harvested by shrimp

| Nitrogen content in *Thalassia* leaf | Harvest of *Thalassia* leaves by shrimp | Nitrogen content in *Thalassia* loose leaves | Harvest of *Thalassia* loose leaves by shrimp | Total harvest of Nitrogen by shrimp |
|---|---|---|---|---|
| % | g DW N/ m$^2$/ day | % | g DW N/ m$^2$/ day | g DW N/ m$^2$/ day |
| 2.03 | 4.40x10$^{-03}$ | 0.95 | 2.86x10$^{-04}$ | 4.69x10$^{-03}$ |

**Table 15** – Nitrogen harvested by shrimp. The total harvest of nitrogen, in milligrams per m$^2$ per day, by alpheid shrimps in a seagrass bed, dominated by *Thalassia hemprichii*.

## 4.    Discussion

With reference to the Introduction of this research report, we became interested in studying the cycling of nutrients on seagrass beds and the involvement of the alpheid shrimp *Alpheus edamensis*. Up till now there is only scares scientific information available about the influence of burrowing organisms - such as the alpheid shrimp - in seagrass beds and how they affect the nutrient cycle. We believe that these burrowing organisms play an important roll in the creation of a significant sink associated with seagrass beds and in the nutrient conservation strategies in seagrasses. Therefore the main goal of this experimental research was to establish the amount of seagrass the alpheid shrimp collect on a daily basis. In this research context we also wanted to observe the alpheid shrimp's behavior (see Table 1) in the seagrass beds as a starting point for future scientific research. In this section we want to present the most challenging issues in our field of research for discussion, based on the results of our experimental research activities, respectively.

### Shrimp Density

One of the effects of the storm is a decrease in the density of alpheid shrimps and *Axius acanthus* (see Table 3). During the research period we found densities of the alpheid shrimp to be $0.44 \pm 0.34$ m$^{-2}$ (see Table 2) on average. Earlier measurements, done by Stapel and Erftemeijer (2000) on a island nearby Bone Batang, resulted in estimated densities of $10.3 \pm 5.0$ m$^{-2}$ for burrows, and approximately 50% inhabited with alpheid shrimp. This could be an indication that the population of alpheid shrimp in the area is either experiencing a sharp decline, which fluctuates very strongly throughout the years or seasons (Erftemeijer and Herman, 1994), or that the anthropogenic influence on Barang Lompo, e.g. nutrient input, allows for a much larger population to sustain itself. It may also have other causes, but these are unknown up till now.

### Harvest of the daily production

This research recorded the harvest of seagrass by a burrowing shrimp during the wet season where the seagrass bed experienced a lot of physical stress due to high-energy currents and waves caused by a tropical storm at the time of the research. The results are supportive of the sink theory proposed by Griffis and Suchanek (1997) and Stapel and Erftemeijer, (2000) and the role of the burrowing shrimp by its harvesting of leaves.

Figures 8-13 show the activity of the shrimp, from many different perspectives. The tide and the time of day do not seem to influence the level of activity (Fig. 8-10). These findings are in contrast with what was found by Stapel and Erftemeijer in 2000. They found a daily rhythm with a harvesting peak at noon. We found daily rhythm with no peak at noon, but we did find a harvesting peak at lower tides (see Fig. 8 d1). A possible reason for that could be a higher "disturbance" frequency at higher tides (see Fig. 8 g1). This is also reflected in the successes in leaf harvest; about a 4% when the disturbance is very high to 52% success rate of cutting seagrass when the disturbance is very low (see Fig 12 and 13). The shrimps' activity is considered to be constant and the harvest of the leaves constant with slight deviations on a daily basis. We therefore did not integrate over the day like Stapel and Erftemeijer (2000) to get our total daily amount of seagrass material harvested.

Most of the shrimp's activity (see Table 1) is spend on sediment reworking (~90%) and sediment expulsion dominates this with 50 to 80 percent (see Fig.11 and 14). The rest of the activity is spend on the attempt to cut seagrass, which isn't always successful. Out of all attempts, between 4% to 52% are successful in cutting and retrieving fresh seagrass leaves depending on the disturbance frequency. Loose leaves are harvested to a lesser extent, ranging from 0% to 12% of all attempts to cut seagrass. We added the "cutting of seagrass" and "loose leaves harvesting" as a rate of success to the number of "attempts to cut seagrass" (see Fig. 12), because the shrimp only leaves the burrow with the intention to retrieve seagrass. It seems that in this process it "stumbles" on a fresh shoot or can stumble upon a piece of loose leaf and harvests that instead of continuing to "attempt" to cut seagrass. We did not look at the amount of loose leaves were available, or at the density of seagrass in the direct neighborhood of the burrow entrance.

The alpheid shrimp burrows are relatively deep. This depth was established by making burrow casts of the alpheid shrimps burrow (these are still being processed for photography). Once reassembled and photographed these burrow casts can be compared to burrow casts of other seagrass collecting shrimp like the thalassinid shrimps studied by Griffis and Suchanek (1991). This can shed light on what happens inside the burrow, and provide clues as to what the fate of the harvested plant material is. All the harvested material in this burrow most likely did not get washed away during the heavy storm, because of the depth of storage, though we have no evidence to support this theory. It seems highly unlikely that the storm was able to remove 70 centimeters of sediment in depth,

uprooting all the seagrass in the process, after which more sediment was deposited on top of the just uprooted seagrass.

Figure 17 shows that the nitrogen content of the leaves is almost two times higher than the content in any other part of the plant and four times higher than the N-content in the sheath. In our research the leaf nutrient compositions are showing around 2% for nitrogen and 35% for carbon in leaves, but Erftemeijer and Herman (1994) showed that these ratios fluctuate through the seasons. Around the time the heavy storm passed over the area the percentages for carbon, nitrogen and phosphor shift, as does biomass dry weight. This makes it impossible to use a fixed growth and production rate based on long-term research. Growth rates would best be determined *in situ* (see Table 7).

*Nutrient concentration*

The water samples of the nutrient concentration inside the burrow and in the expelled sand show no difference of intra-sediment concentration. The highest nutrient concentrations were found in dense areas of seagrass (see Fig. 15 and 16). The water column sample is consistent with the nutrient-poor environment as expected. Deeper in the sediment, in a location where more plants are situated, nutrients would be more readily available due to loose leaves, detrital decomposition, e.g. external cycling (Hemminga *et al*. 1999). Though Stapel *et al* (1997) showed that most phosphate is located in the top layer and ammonia becomes more readily available at greater depths, we suspect that the heavy storm had either deposited a large layer of sediment on the seagrass bed, or thoroughly mixed the top layer (~10 cm) of sediment. The reason to suspect this sediment deposition took place, was because the rhizomes were much deeper in the sediment than before the storm (pers. obs.). This is also reflected in a large part of the leaves (~57%) not being pigmented green (see Table 4). Furthermore, the variation between replicas and the overall low values of $NH_4^+$ (see Fig. 15 and 16) would suggest a loss of sediment-bound nutrients.

Koike and Mukai, 1983) found that the production of nitrate and nitrite in the burrow was mainly due to bacterial metabolism. Fourqurean & Schrlau (2003) found that *Thalassia testudinum* looses Carbon, Nitrogen, and Phosphor faster in buried incubations compared to sediment surface incubations. We found no evidence for higher concentrations of nutrients leaving the burrow via ventilation, instead we found the nutrient concentration to be quit similar to those found in intersediment and the water column level (see Fig. 15 and 16).

### Harvest comparison

The influence of the alpheid shrimp as we found in the seagrass system on Bone Batang is less during the raining season than the results, which Stapel and Erftemeijer (2000) mentioned in their pilot study. According to our findings the alpheid shrimp is responsible for harvesting ~17.2% of the daily leaf production (see Table 7) and 40.1% of the above ground leaf production (see Fig. 3). In total the alpheid shrimp harvests 4.7 mg of Nitrogen per $m^2$ per day. Estimates by Stapel and Erftemeijer (2000) are 32 mg N $m^{-2}$ $d^{-1}$ respectively, which is almost 7 times more in DW than in our findings. Stapel and Erftemeijer found that the shrimp harvested 53% of the total daily leaf production, which is three times higher than our findings of 17.2%.

### Nutrient sink theory

In the period we did our investigations we observed a smaller role for the alpheid shrimp in nutrient cycling, than Stapel and Erftemeijer did in their research (2000). Our fieldwork took place in exceptional weather circumstances, which had a large impact on nutrient compositions, both sedimentary (see Fig. 15 and 16) and leaf (see Fig 17), on the growth rate of *T. hemprichii* (see Table 7) and on the density of the shrimp (see Table 3). It is therefore not entirely surprising that our research findings show different results when compared with those of Stapel and Erftemeijer (2000), although their previous research was a very important source of information for our actual research design. Nevertheless, the alpheid shrimp's harvesting still proves to be a pathway for nutrients that are leaf bound. To what extent this harvesting of attached seagrass leaves and lose leaves play a roll in the nutrient cycling in the seagrass beds located in the Spermonde Archipelago near Makassar, Indonesia is yet to be further quantified, but is expected to be season dependent.

## 5. Conclusions and recommendations for further research

From the findings of our research we can detach the following conclusions:

- In our field study, most of the shrimps' activity seems to be dedicated to maintaining the burrow. The harvest of loose leaves is marginal compared to the harvest of fresh leaf material. It could possible be that the leaves might function both as a food source for the alpheid shrimp as well as that of materials for burrow construction, or burrow maintenance. The exact nature of the use of the leaves needs further investigation.
- The fate of the organic matter inside the burrow also plays an important role in the nutrient cycling. There is reason to believe that the main diet of the shrimp does not consist of detrivores, like it's gobiid fish partner, but of loose leaves and epi- and interstitial fauna (Karplus 1987).
- Decomposition rates of leaves are attributed to their physical and biochemical properties, and therefore can differ per species of seagrass. This information on the *Thalassia hemprichii* leaves is still lacking. It is therefore important to look at processes inside the burrow to gain a better understanding of the nutrient cycling in the seagrass bed in relation to sedimentary fauna.
- From personal observation we learned that burrows surrounded by fresh seagrass had a higher rate of harvesting than those burrows with seagrass a bit further away from the burrow. This would indicate that seagrass density and shrimp density, which was whittled down by the storm, play an important role in the efficiency of nutrient cycling.
- The findings of this experimental, 3-months lasting field research, and the findings done before strongly suggest that the intensity and quantity of nutrients cycled by the seagrass harvesting shrimp are very much season-dependent. Only a long-time survey that lasts a couple of years could provide us with viable and trustworthy knowledge of the nutrient cycling that is created by the harvest of seagrass on location.

### Some recommendations for further research

This research has left a few open questions that proved to be interesting for future research. These items concern both the *Alpheus edamensis* and *Thalassia hemprichii* seagrass beds.

We spend some time trying to catch the alpheid shrimp and its gobiid fish partner. Though many different types of techniques were used, none of them proved successful. The main problem with catching these shrimp is its gobiid fish partner and their capability to retract back into the safety of the burrow at high speed. A second problem to be tackled is the instability of the burrow. A technique that proved very effective for catching the *Axius acanthus* resulted only in the collapse of the alpheid shrimp's burrow. The stomach contents of another alpheid shrimp, *A. bellulus*, consisted of fairly large amounts of unidentified materials. It could also be possible that shrimps feed on bacteria growing on the harvested seagrass material (Karplus 1987). Capturing and analyzing gut content and processing the tail muscles needs more detailed investigation and could provide a better insight into thé diet and, indirectly, the fate of harvested seagrass and loose leaves.

How it is possible that seagrass beds exist in nutrient poor environments while lacking any form of nutrient efficiency normally associated with such an environment is still unknown. The mystery of how these sinks or sources are created and how they are sustained is slowly being unraveled.

6. Bibliography

Aerts, R. 1990 Nutrient use efficiency in evergreen and deciduous species from heathlands. *Oecol.* 84, 391-397

Chapin, F.S. 1980 The mineral nutrition of wild plants. *Ann. Rev. Ecol. Syst.* 11, 233-260

den Hartog, C., 1970. *The sea-grasses of the world.* Amsterdam: North Holland Publishing Company.
Duarte, C. M. Middelburg, J. J. and Caraco N. 2005 Major role of marine vegetation on the oceanic carbon cycle. *Biogeosci.* 2, 1–8

Erftemeijer, P.L.A. and Herman, P.M.J. 1994 Seasonal changes in environmental variables, biomass, production and nutrient contents in two contrasting tropical intertidal seagrass beds in South Sulawesi, Indonesia. *Oecol.* 99, 45-59.
Fourqurean, J.W. and Schrlau, J.E. 2003 Changes in nutrient content and stable isotope ratios of C and N during decomposition of seagrass and mangrove leaves along a nutrient availability gradient in Florida Bay, USA. *Chem. and Ecol.* 19(5), 373–390

Granger, S. and Iizumi, H. 2001 Water quality measurements methods for seagrass habitat. *In* Short, F. and Coles, R.G. Global seagrass research methods. Elsevier, Amsterdam

Griffis, R.B. and Suchanek, T.H. 1991 A model of burrow architecture and trophic modes in thalassinidean shrimp (Decapoda: Thalassinidea). *M.E.P.S.* 79, 171-183.

Hemminga, M.A. Marbà, N. and Stapel, J. 1999 Leaf nutrient resorption, leaf lifespan and the retention of nutrients in seagrass systems. *Aqua. Bot.* 65, 141-158.
Karplus, I. 1987 The association between gobiid fishes and burrowing alpheid shrimps. *Oceanogr. Mar. Biol. Ann. Rev.* 25, 507-562.
Koike, I. and Mukai, H. 1983 Oxygen and inorganic nitrogen contents and fluxes in burrows of the shrimps *Callianassa japonica* and *Upogerbia major*. *M.E.P.S.* 12, 185-190.
Man J.G de, 1888 Bericht über die im indischen Archipel von Dr. J. Brock gesammelten Decapoden und Stomatopoden. *Archiv für Naturgeschichte* 53: 215-600.

Lin, H.J. and Shao, K.T. 1998 Temporal changes in the abundance and growth of intertidal *Thalassia* hemprichii seagrass beds in southern Taiwan. *Bot. Bull. Acad. Sin.* 39, 191-198

Moncreiff, C.A. and Sullivan, M.J. 2001 Trophic importance of epiphytic algae in subtropical seagrass beds: evidence from multiple stable isotope analyses. *M.E.P.S.* 215, 93-106.
Richmond M.D. 2002 A field guide to the seashores of Eastern Africa and the Western Indian Ocean Islands. 2nd edition, Eurolitho, Milano, 2002, through Italgraf, Västerås, Sweden.

Stapel, J. and Erftemeijer, P.L.A. 2000 Leaf harveting by burrowing alpheid shrimps in a *Thalassia* hemprichii meadow in south sulawesi, Indonesia. *Biol. Mar. Medit.* 7, 282-285.

Stapel, J. and Hemminga, M.A. 1997 Nutrient resorption from seagrass leaves. *Mar. Biol.* 128, 197-206

Stapel, J. Hemminga, M.A. Bogert, C.G. and Maas, Y.E.M. 2001 Nitrogen ($^{15}$N) retention in small *Thalassia* hemprichii seagrass plots in an offshore meadow in South Sulawesi, Indonesia. *Limnol. Oceanogr.* 46(1), 24-37.

Stapel, J. Manuntun, R. and Hemminga, M.A. 1997 Biomass loss and nutrient redistribution in an Indonesian *Thalassia* hemprichii seagrass bed following seasonal low tide exposure during daylight. *M.E.P.S.* 148, 251-262.

Stapel, J. Nijboer, R. and Philipsen, B. 1996 Initial estimates of the export of leaf litter from a seagrass bed in the Spermonde Archipelago, South Sulawesi, Indonesia. *Seagrass Biology: Proceedings of an International Workshop* 155-162.